NASA Astrobiology Institute • NAI

Annual Report Year 3

July 2000 - June 2001

Future
Origins
Microbes
Meteorites
Mars
Hydrothermal
Extremophiles
Archaea
Evolution
Prebiotic
Carbon
Europa
Biogeochemistry
Extrasolar
Water
Cyanobacteria

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Year 3 Annual Report

NASA Astrobiology Institute
Ames Research Center
MS 240-1
Moffett Field, CA 94035-1000
U.S.A.
http://nai.arc.nasa.gov

Director
Baruch S. Blumberg

Associate Director
Rosalind A. Grymes

Editors
Karen E. Dodson
Wendy Dolci
Lisa Faithorn
Edward M. Goolish
David Morrison
Janet L. Morrison
Krisstina Wilmoth

Publications
Janet L. Morrison

Graphic Design and Production
Julie Fletcher

Technical Support
Shige Abe
Karen C. Bradford
Jeni Joiner
Katherine Lew
Nicole Tamer

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At the end of each report there is a "Back to Table of Contents" button. Click on it and it will return you to the Contents page.

All websites are active within this document.

Year 3 contents

Letter from the Director 4
Introduction 7
Map of Team locations 13

Summary of Lead Team Research and Education & Public Outreach

Ames Research Center 14
Arizona State University 46
Carnegie Institution of Washington 84
Harvard University 132
Jet Propulsion Laboratory (1) 160
Johnson Space Center 194
Marine Biological Laboratory 232
Pennsylvania State University 272
Scripps Research Institute 322
University of California, Los Angeles 338
University of Colorado, Boulder 356

New Team Executive Summaries

Jet Propulsion Laboratory (2) 382
Michigan State University 386
University of Rhode Island 392
University of Washington 398

NAI Focus Groups

Astromaterials 404
Europa 406
EvoGenomics 409
Mars 412
Mission to Early Earth 415
Mixed Microbial EcoGenomics 419

NAI Central Education & Public Outreach

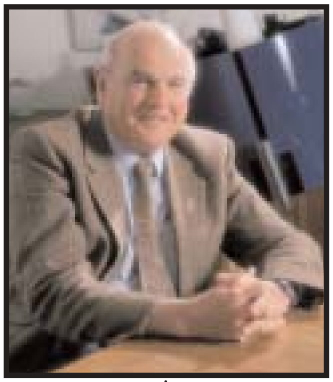
NAI EPO 423

NAI/NRC Postdoctoral Fellows

August 2000 Fellows 431
April 2001 Fellows 432

Appendices

Director's Science Council 434
Astrobiology Roadmap 435
NAI Team Projects & Focus Groups/Roadmap Objectives 437



Letter from the Director

Baruch S. Blumberg

The NASA Astrobiology Institute (NAI) was established to encourage and fund astrobiological research in basic science and to support the space-venturing missions of NASA. Humans have a long-term interest – probably pre-dating written history – in the questions, "Are we alone in the Universe? Is life a unique event that occurred only on our own Earth, or do we exist in a life-rich Cosmos?" These, coupled with the other fundamental questions, "How did life begin?" and "What is the future of life in the Universe?" encompass the subject matter of astrobiology. In the last half-decade, technological developments and the promise of research-focused space flight have allowed, and will continue to support, a scientific approach to these questions to parallel their investigation by other means.

By 1996 interest in astrobiology was fueled by several exciting scientific revelations, including: 1) Images of Europa returned by the Galileo spacecraft indicating the possibility of liquid water under its cracked, icy surface; 2) Detection, in a meteorite from Mars (ALH 84001) of material that could be of biological origin; and 3) The discovery of new planets around extrasolar stars. In September of that year, the First Astrobiology Workshop was convened at NASA Ames Research Center (ARC) at Moffett Field, CA to take an initial step toward the definition of astrobiology. In March 1997 ARC was designated as the Lead Center for Astrobiology and, in the same year, NASA formalized plans to establish the NASA Astrobiology Institute. On October 31, 1997 a Cooperative Agreement Notice (CAN), that is, a request for research proposals, was issued. More than fifty US research institutions responded, including universities, nonprofit research centers, NASA Centers, and other government agencies. The proposed projects were investigator-initiated basic science, constrained only by the wide scope of astrobiology itself. A requirement for submission was the representation of two or more academic disciplines within the proposing team. From its outset, therefore, NAI basic research was interdisciplinary. Other attributes required in the proposals were a strong education and public outreach program, as well as significant financial and other contributions from the participating institutions.

In July of 1998 eleven teams were chosen for five years of funding, initiating a core group for NAI research. Also in July, several hundred scientists from the general scientific and astrobiology community were invited to the Astrobiology Roadmap Workshop held at ARC. As a product of this workshop, the science community identified ten science goals, seventeen objectives, and four principles integral to the operation of the astrobiology research program. A definition for astrobiology was agreed on: "The study of the origins, evolution, distribution, and future of life on Earth and in the Universe." G. Scott Hubbard was appointed Interim Manager in August, and, in November, all individual members of the NAI were invited to the First NAI General Meeting. In May 1999 I was appointed the Institute's first Director by Harry McDonald (Director of ARC), Daniel Goldin (the Administrator of NASA), and Edward Weiler (Associate Administrator for

Space Science). The office of the NAI Director and his staff – termed NAI Central – was established at ARC. A formal association with the NAI's first international partner, the Centro de Astrobiología in Torrejon de Ardoz, Spain, soon recognized the international reach of astrobiology research.

Excellence in multidisciplinary basic and applied research is the first priority of the Institute. NAI was established as an interdisciplinary activity, with the various disciplines traditionally using somewhat different styles of the scientific process. The historical sciences – for example, astronomy, geology, and paleontology – are particularly strong on the inductive observation of events and processes that have already happened, while chemical and biological sciences rely to a significant extent on the experimental testing of hypotheses. In the inductive phase of research, data are collected first, and then from this information hypotheses are formulated. In the deductive phase, the hypothesis is formulated first, then data are collected to test it. These processes go on sequentially or in parallel, often in a seamless progression, usually yielding complex and interacting data sets. Astrobiology is a fascinating amalgam of these approaches.

Considering the basic science goals of astrobiology, and the need to apply them to the needs of NASA missions, the science of the NAI Lead Teams is unlikely to progress in direct accordance with the original research proposals, or be limited by the Astrobiology Roadmap as written in 1998. This flexible approach allows for the development of new ideas over the period of the five-year Cooperative Agreements. The NAI Principal Investigators, (that is, the scientist in each Lead Team responsible for the coordination of the overall research program) comprise the NAI Executive Council (EC). Regular communication with the Director and the staff of NAI Central is maintained by monthly video conferences, frequent face-to-face meetings of the EC, and at biennial meetings of all the individual members of the NAI. An additional meeting, the Astrobiology Science Conference, is jointly sponsored by ARC and NAI for all members of the astrobiology community. Held in alternate years, the first of these took place at Ames Research Center in April 2000, and the next will be in April 2002.

A second Cooperative Agreement Notice was announced in June of 2000, and four additional Lead Teams were selected from among 28 competitive proposals. Affiliate relationships have also been established with three international partners in addition to the Centro de Astrobiología in Spain: the United Kingdom, Australia, and France. Discussions are in place with several other potential international partners. These arrangements with other astrobiology organizations have resulted in many collaborative projects with the NAI, and others are planned.

From its beginning, NASA has been goal directed and dependent on the successful completion of ambitious space missions. Even a few decades ago these missions would have been considered miraculous. The mission to the Moon was generated, in part, by competition with the former Soviet Union to demonstrate technical and scientific excellence. The goal was evident and could be seen on most clear nights – to achieve it required outstanding technical and engineering skills. This can be said for many of the subsequent planetary and outer space missions of NASA and other space agencies. Instruments for scientific discovery are in place at this moment, and many more will be in the future. Basic scientific research, with a major emphasis on astrobiology, is becoming more and more prominent in space programs. A primary mission of NAI, and astrobiology in general, is to help develop the basic science required for these missions. As the rapidly maturing field of astrobiology evolves, this new discipline will have increasingly significant influence on the development of future NASA space missions.

Over the three years of its existence, certain features of the NAI have emerged. The funds are considered as Cooperative Agreements and not contracts. The individual scientists direct the research they will conduct within the broad limits set by NASA mission requirements. We do not expect that they will adhere precisely to the original proposal since it is the nature of basic science that new data and ideas emerge as the research progresses. The Implementation Plan for NAI vests authority for leadership and management in the Director who, in turn, is responsible to the Director of ARC and to the appropriate officials at NASA Headquarters. However, the Executive Council in its advisory capacity has a major role, not only in the direction of the science, but in the management of the Institute as well. Responsibilities of NAI Central include assisting the EC in these roles and the individual scientists in their research. The NAI Central staff makes every effort to respond to the needs of the scientists by providing easy access by mail, telephone, video, and in-person meetings. Although NAI can only fund a limited number of Lead Teams, many of the programs supported by the Institute are open to the general scientific community, and scientific collaboration outside of the NAI is encouraged. We believe that this new field of astrobiology will continue to flourish and, as it does, we are committed to the inclusion of students and scientists at Minority Institutions in both our educational and research programs.

International collaboration is central to our effort; no single nation can accomplish the goals of this large and growing field. The intellectual and financial resources of many nations will be needed to realize the program effectively. Although research is our major activity, we at the NAI have the responsibility to train and motivate the young-scientists-to-be who will complete the long-term research that is characteristic of this field. We must also inform the public and convey to them the excitement and relevance of this field. We believe that we also have a role, along with the rest of the scientific community, in establishing the field of astrobiology as a rigorous, broad, exciting, and effective scientific enterprise.

I am pleased to submit to you the Third Annual Report (2000-2001). These Lead Team and Focus Groups reports detail the scientific and educational progress of Institute-sponsored projects and highlight milestones in the maturation of the NAI as a collaborative global community.

Sincerely,

Year 3

Introduction

Research, Education, and Collaboration

The NASA Astrobiology Institute is NASA's investment in supporting the emergent field of astrobiology by encouraging broad interdisciplinary research, providing infrastructure resources to the community, and developing information exchange conduits to influence mission planning and technology directions. The NAI was conceived as a new approach to funding and managing science; the NAI awards financial support for 5 year periods to research teams selected through a rigorous peer-review process. Selected Teams become participants in the "virtual" or distributed Institute. The NAI's Director provides advocacy and leadership to the Institute, and through the staff at NAI Central (located at NASA Ames Research Center) directs the distribution of research support and assures the Institute's continuity, effective operations, and professional and public communications. The NAI's science activities are conducted in the distributed laboratories and field sites of the 700 Members (residing at over 100 collaborating institutions), organized through the Principal Investigators (PI's) of the fifteen Lead Teams. These PI's, together, comprise the Executive Council of the NAI, the Institute's technical guiding body.

The Annual Report of the NASA Astrobiology Institute emphasizes the discoveries and research accomplishments of the Institute Teams and their Members. This document also affords an opportunity to record key elements in the annual progress of the Institute. In the first three years, the Institute established its initial architecture and the selection of the first two groups of participating Teams, including the selection of four additional Teams following our second Cooperative Agreement Notice solicitation (CAN-2). In response to CAN-2 (June 2000), the twenty-eight submitted proposals were competitively reviewed (December 2000) and selections were announced (March 2001). More information on the new Lead Teams can be found in this document and at http://nai.arc.nasa.gov. During this initial implementation phase of the NAI, several approaches have contributed substantially to creating an interactive community and supporting a flow of intellectual innovation. Among these are: investigator-initiated Focus Groups, NAI Central-initiated award augmentations, responsive funding opportunities, and partnerships supporting international participation. Underlying themes integral to the Institute concept are represented by our commitment to education, student training, and public outreach, and our experimentation with, and evaluation of, collaborative technology tools. Accomplishments in all these areas contributed to the growth and impact of the NAI during the past year.

A key element in the creation of this collaborative and innovative community, the NAI's Focus Group program illustrates and realizes our commitment to interdisciplinary exchange, idea generation, and participation opportunities extended to scientists not formally supported through NAI membership. The two Focus Groups chartered in Year 3 also demonstrate the NAI's support of solar system exploration as well as fundamental research directions. The Europa Focus Group (endorsed September 2000, Chair, R. Greeley, ASU and Director's Science Council) engages scientists from diverse disciplines to consider exploration scenarios based on science objectives. The Astromaterials Focus Group (endorsed March 2001, Chair, D. McKay, JSC) will take a comprehensive and, again, interdisciplinary approach to studying extant meteoritic, cometary, and interstellar dust particle samples. In addition to the activities of these newly chartered groups, those initiated in prior years continue to be active and influential. The Mars Focus Group held a meeting to overflow crowds at the biennial General Meeting. Having influenced the process of landing site selection for the 2003 Mars mission, Chair J. Farmer (ASU) has encouraged members to expand the Focus Group's interests to terrestrial field work, instrumentation, and biosignatures research. The Mission to Early Earth (MtEE) Focus Group (Co-chairs A. Anbar, JPL and HAR, and S. Mojzsis, CUB), proposed a field investigation in Australia, jointly led by the Co-chairs and R. Buick, in the summer of 2001. This group demonstrates astrobiology's ability to connect space exploration with virtual time travel; in seeking to understand the early Earth, MTEE investigators look back through time at a planet very different from the Earth we experience today. Introductions effected through joint field investigations and through Focus Group meetings provide a continuing mechanism for innovative interdisciplinary exchange that links the Institute's Members through mutual research interests and opens a conduit between the Institute and the broader astrobiology community.

Another operational approach of the NAI provides flexible resource and funding support to new investigations, instrumentation needs, international scientific exchanges, and field work. A significant feature of this support is its timely responsiveness to Member-initiated requests. A substantial portion of the Director's Discretionary Fund was allocated, beginning in Year 2 and distributed in Year 3, to an internal research Augmentation Opportunity. Both the EvoGenomics Focus Group (Co-chairs B. Hedges, PSU, and J. Lake, UCLA) and the EcoGenomics Focus Group (Co-chairs M. Sogin, MBL, and D. Des Marais, ARC) received timely support through this mechanism. Over three quarters of a million dollars were distributed in additional awards coincident with the original 5-year Cooperative Agreements with the first group of participating Teams, based on external peer review of proposals submitted from within the Institute. These two Focus Groups were not the only beneficiaries. Additions to existing Lead Team projects were awarded, recognizing the dynamic nature of astrobiology, and indeed all, basic research investigations. In addition to the growth in the Lead Team component of the Institute, several major equipment acquisitions were enabled by partnered funding from NAI Central: an ion microprobe instrument located at the University of Wisconsin (JPL1 Lead Team) and a protein chip microarray facility at the Carnegie Institution of Washington. Both are available to members and colleagues. In addition to initiating and maintaining this receptivity to requests for seed funding and critical continuation support (with active assistance from the NASA Astrobiology Program), we also encourage and support travel and intellectual exchanges amongst our members and with our colleagues distributed internationally.

Year 3

Astrobiology, as a global and emerging science, demands a perspective that transcends national boundaries. In expanding our international partners program, we added two new affiliate members: the United Kingdom Astrobiology Network and the Australian Centre for Astrobiology. We also received several inquiries from international networks of scientists in our third year, one of which became an official request for Affiliate status. This action was concluded in October 2001 when the Groupe de Recherche en Exobiologie became an Affiliate member of the NAI. Other scientific organizations interested in formalizing relations represent astrobiologists in Russia, Mexico, and a network of scientists in twelve countries comprising the European Exo/Astrobiology Network Association. A wide variety of research interactions exist between NAI members and our international colleagues: joint research, field investigations, workshop support, and student exchange. In recognition of the policy and programmatic relevance of these relationships, NAI Central participated in a workshop and associated field trip to the Rio Tinto with our colleagues from the Centro de Astrobiología in September 2000, celebrated the announcement of our Affiliate partnership with the United Kingdom Astrobiology Network at the U.S. Embassy in London in June 2001, and attended the initiation of the Australian Centre for Astrobiology, at Macquarie University, Sydney, and an associated two-day international workshop in July 2001.

Conferences, symposia, and workshops have historically been, and will continue to be, one of the major ways in which new relationships and collaborations are developed within the scientific community. The NAI considers, therefore, that it has a responsibility to support these types of activities when they are directed at advancing the field of astrobiology. Activities that the NAI has supported in the past year include two special sessions at the Geological Society of America and the Geological Society of London Conference on Earth System Processes, which was held June 24-28, 2001 in Edinburgh, Scotland. The two sessions supported were entitled, “Archean Earth and Contemporary Life: The Transition from an Anaerobic to an Aerobic Marine Ecosystem,” and “The Role of Hydrothermal Systems in Biospheric Evolution.” Both sessions were chaired by NAI Members but the invited speakers were drawn from the international scientific community.

Education and Public Outreach

Educating and training the next generation is an important aspect of NAI’s mission, essential to ensuring continuity and longevity of the field of astrobiology. Many of our Members train new researchers directly in their academic programs and laboratories. While some of these courses and programs are called “astrobiology,” many of them reside within traditional astronomy, biology, chemistry, geology, and planetary science departments. In this way, NAI Members are growing the field of astrobiology both as an independent discipline and through the expansion of traditional approaches. In support of our Members’ students, a series of Director’s Scholarships was inaugurated in Year 3: the Director’s Travel Scholarship, for travel to another Team location or a field investigation, and the Director’s Research Scholarship, for support of a student's experimental activities.

The scope of NAI's commitment to education involves students of astrobiology at all levels, including those who have received their doctorate degree. The Institute, from its very inception, as stated in its Implementation Plan, has given high priority to post-doctoral education in astrobiology. In addition to the many postdoctoral students supported by the NAI's Cooperative Agreements with the Lead Teams, the NAI initiated, in Year 2, a competitive NAI Postdoctoral Fellowship Program administered by the National Research Council of the National Academy of Sciences and the National Academy of Engineering. In Year 3, six NAI Fellows were selected (August 2000) in the first application opportunity and an additional six were chosen (April 2001) later in Year 3 (see pages 430-432). These Fellows have a home research base at one of the NAI member institutions, but have the flexibility to interact across the entire Institute. The NAI encourages and supports these Fellows to ensure success both in their current research and in their future careers - as these individuals are expected to lead the next generation of astrobiologists.

In April 2001, the Institute held its second biennial General Meeting, hosted by the NAI Lead Team at the Carnegie Institution of Washington. As at our first meeting, a singular emphasis was placed on representation from the junior Members of the NAI, graduate students and postdoctoral fellows. Specific support of their attendance through generous travel awards was provided, and sessions soliciting their input and feedback on NAI operations were scheduled at the request of the National Academy of Sciences' Committee on the Origin and Evolution of Life. Another indication of our emphasis on student recognition is the inauguration of the Gerald Soffen Memorial Award, given for the best student research paper/poster submitted to the NAI General Meeting. The first recipient of this award was Jennifer Eigenbrode (PSU).

Complementing our investments in undergraduate and graduate programs, NAI also educates students, teachers and the general public. We are building a future community of astrobiologists while expanding the public's understanding of the nature and importance of our work. NAI's Education and Public Outreach (EPO) program is distributed throughout its Lead Teams. Each Team directs a local effort with specific emphasis on that Team's research and expertise while contributing to larger collaborative projects. These activities are outlined in the Team reports. The NAI Central office coordinates, directs, and funds cross-cutting activities which encompass all of astrobiology and can be offered on a national level. These include websites, print products, and curriculum supplements, as well as educational programs and activities, internships, presentations, and exhibits. Details of these specific projects are outlined in NAI Central's EPO report (see page 424).

A particular element combining NAI's research and outreach emphases, and initiated in Year 3, is our undertaking to engage the participation of Minority Institutions. Extensive discussions have been held during the development of this initiative. These have included the full range of cognizant offices and individuals at NASA HQ and NASA ARC, our Executive Council and other Lead Team scientists, and representatives from the Minority Institutions (Historically

Black Colleges and Universities, Hispanic Serving Institutions, and Tribally Controlled Colleges). A number of alternatives have been considered addressing K-12 audiences, undergraduate and graduate students, and college faculty. A final determination of the most effective approach to draw Minority Institutions productively into the Institute's efforts will be made in Year 4. This will likely involve competitively selected faculty sabbaticals for qualified researchers from Minority Institutions to be pursued in partnership with NAI scientists, in the laboratory and in the field. Opportunities such as this, as well as information regarding the Focus Groups, NAI's Lead Teams, the Postdoctoral Fellows program, and the Annual Science Reports, are publicized at meetings of professional societies to better enhance the community's knowledge of, and participation in, the Institute.

Collaborative Technology Architectures

The biennial General Meeting is a valuable and much-anticipated opportunity for dialog. Other such exchanges occur throughout the year. NAI Central and the Executive Council of the Institute meet monthly (including our international colleagues and NASA HQ sponsors) by videoconference and three to four times each year in person. The EPO leads gather via video- and teleconference, as do the Collaborative Technology leads. The Focus Groups, and other special interest meetings and seminars, engage face-to-face, by videoconference and telecon, and exchange information by e-mail. These interactions contribute to the impression, and the reality, that the operational side of the NAI is a distributed function.

The array of tools and technologies developed by government and private industry to support electronic communication and collaboration is continually expanding and evolving. Once initial contact is made, perhaps through face-to-face encounter, subsequent effort is needed by potential collaborators to establish and maintain the work practices that can result in synergy and resource maximization. An important part of NAI's mandate is to provide a technology architecture that effectively facilitates remote collaboration. During Year 3, important strides were taken in this direction as challenges were identified and addressed. Three significant challenges addressed during this year were: on-going technical difficulties with the Polycom™ videoconferencing system, widespread member dissatisfaction with the *Postdoc* document sharing and archiving system, and insufficient understanding about, and technical support of, NAI Members' articulated needs.

By January of 2001, a number of technical problems with the multicontrol unit (MCU) at Ames were identified by the NAI Technical Operations team, resulting in a diagnosis of incompatibility between the MCU unit and the Polycom™ systems installed at each NAI lead institution. Negotiations with the vendor eventually resulted in replacement of the MCU in June of 2001 with a model that supports up to 24 participating sites at once. NAI routinely hosts videoconferences with a larger number of participating sites than any other of the vendor's customers, and we have become a development testbed for them. The new unit can simultaneously support participants from the Polycom™ conference room systems and certain desktop systems, a feature we are currently evaluating. As Year

4 begins, the next generation MCU and equipment deployed to the new NAI lead institutions (the JPL2 Team shares facilities with JPL1) is finally providing satisfactory videoconferencing performance.

Postdoc, a NASA-Ames designed software system for document sharing and archiving, was acquired as a knowledge management tool and information repository. It provides a range of features theoretically of great utility for members of a virtual institute. However, the vast majority of NAI Members found it difficult to access or navigate. Widespread criticism of *Postdoc* led to two Collaborative Research initiatives. The first was discontinuing *Postdoc* while researching the features and functionalities that NAI Members expressly require. The second was launching a systematic study of commercial off-the-shelf (COTS) computer tools and technologies. The study is being conducted by consultants with expertise in collaboration technologies and virtual team building in partnership with NAI's Collaboration Research Support Group. It will produce a set of solutions for NAI from the wide range of COTS options examined.

Over the first three years of NAI's operations, NAI Central staffers have increased their understanding of the Members needs and their expectations of the virtual institute. A comprehensive knowledge capture was undertaken through a Systematic Needs Assessment. The first part of this community-wide assessment involved site visits to the NAI lead institutions. Key themes emerging from these interviews led to a Needs Assessment Survey distributed to Members at the end of the summer 2001. A comprehensive report, with recommendations, will be distributed when the data are tabulated and analyzed.

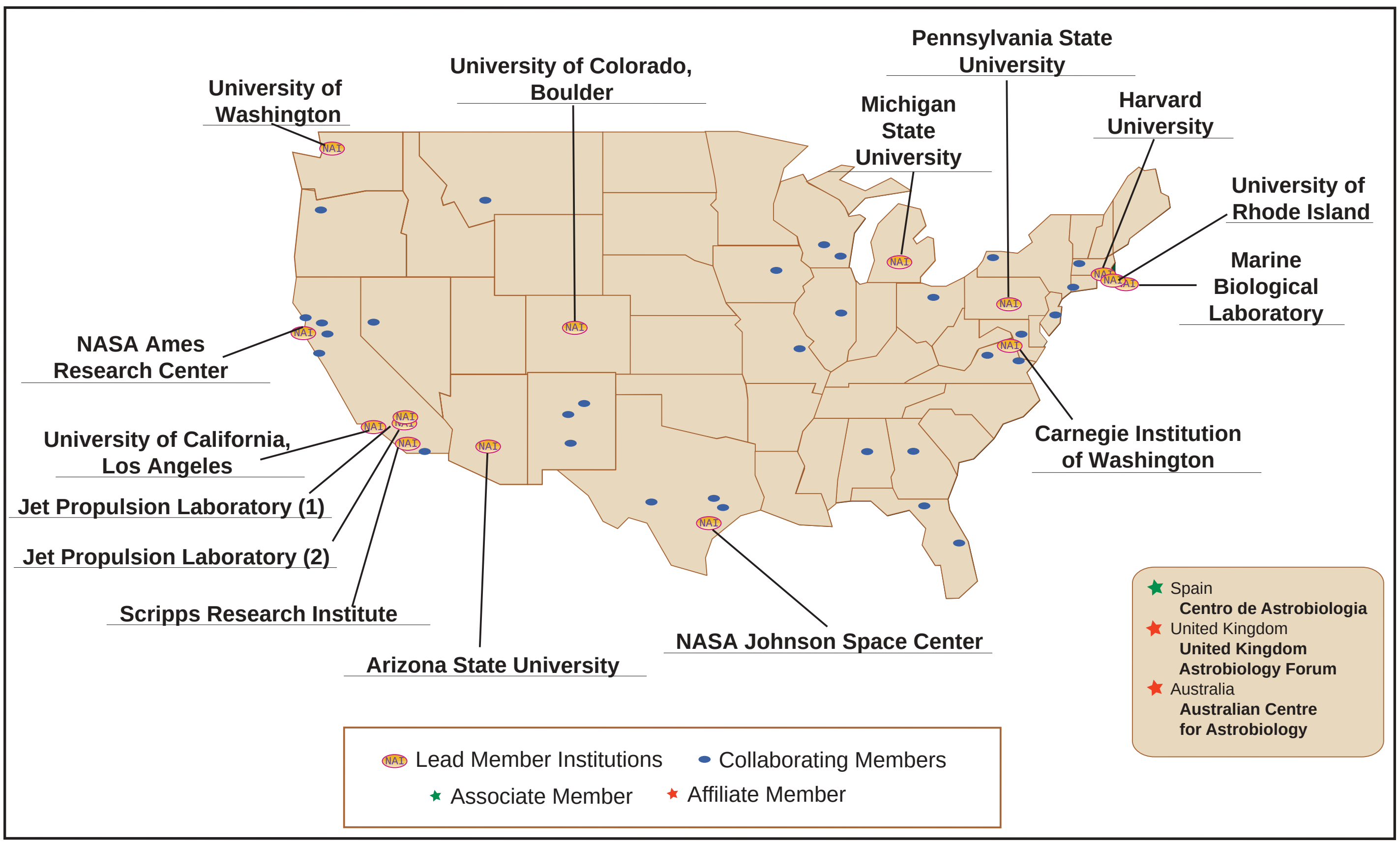
Conclusion

The NASA Astrobiology Institute now represents over 700 Members and maintains connections around the world through our four international Associates and Affiliates. Over the past three years, pioneering organizational efforts have established the Institute and demonstrated the effectiveness of its novel approaches to science management. We are forging new partnerships, new research directions, new collaborative behaviors, and learning as we go. The lessons learned and the solutions discovered illustrate new ways of supporting science, scientists, and cooperative consortia.

This document reflects the history and inception of NAI (in the Director's Introduction), the contributions of NAI Central and an Institute-wide perspective on accomplishments, and the current status of research progress (in the following science reports from the Lead Teams and Focus Groups). This *Annual Report* covers the period from July 2000 to June 2001, the third year of the NAI's Cooperative Agreement Awards. Each report outlines the general intent and current accomplishments of the Team's work, identifies the Team's Members, and correlates the Team's projects with specific objectives of the Astrobiology Roadmap.

Map of Team Locations

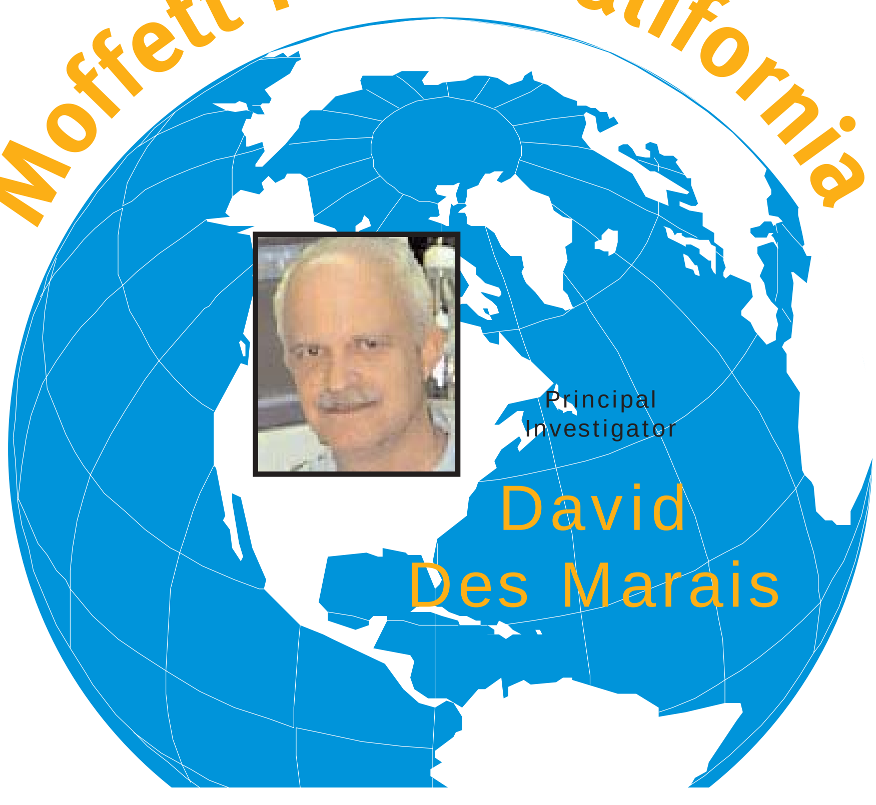
Year 3



Ames Research Center

NASA Astrobiology Institute

Moffett Field, California



Principal Investigator

David Des Marais

Ames Research Center Team Members

Maria Lucia Absy
INPA Manaus

Marcelo Acha
INIDEP (Argentina)

Dan Albert
University of North Carolina

Louis Allamandola
Ames Research Center

Brad M. Bebout
Ames Research Center

Max Bernstein
Ames Research Center

Dan Buckley
University Of Connecticut

Lidia Susana Burry
INIDEP (Argentina)

Steven Carpenter
Orbital Science Corp.

Richard Castenholz
University Of Oregon

Kenneth Cullings
Ames Research Center

Jeffrey N. Cuzzi
Ames Research Center

Hector D' Antoni
Ames Research Center

David W. Deamer
University of California Santa Cruz

David J. Des Marais
Ames Research Center

Donald DeVincenzi
Ames Research Center

Jesse Dillon
University Of Oregon

Mykell Discipulo
Ames Research Center

Moira Doty
Ames Research Center

Jennifer Dungan
Ames Research Center

Jason P. Dworkin
Ames Research Center

Jack D. Farmer
Arizona State University

R. Freedman
Space Physics Research Institution

Ferran Garcia-Pichel
Arizona State University

Raul Guerrero
INIDEP (Argentina)

Robert Haberle
Ames Research Center

Tori M. Hoehler
Ames Research Center

Mary Hogan
University of California Santa Cruz

Gerda Horneck
DLReV Institute of Aerospace (Germany)

Colleen Howell
Ames Research Center

Miguel-Angel Huerta-Diaz
University of Ensenada Baja CA

Linda L. Jahnke
Ames Research Center

Jeff Johansen
John Carroll University

James F. Kasting
Pennsylvania State University

Anthony Keefe
Harvard University

Kevin Kelly
University Hawaii

Janos Lanyi
University Of California Irvine

Carlos A. Lasta
INIDEP (Argentina)

Antonio Lazcano Araujo Reyes
UNAM (Mexico)

Jack Lissauer
Ames Research Center

Kathleen L. Londry
University of Manitoba

Nora Madanes
University of Buenos Aires

Rocco L. Mancinelli
Ames Research Center

Jorge Marcos
University of Barcelona

Christopher McKay
Ames Research Center

Paula Messina
California State University San Jose

Ellen Metzger
California State University San Jose

Hermes Mianzan
INIDEP (Argentina)

Scott Miller
Ames Research Center

Ante Mlinarevic
California State University San Jose

Eduardo Musacchino
INIDEP (Argentina)

Michael H. New
Ames Research Center

Uli Nuebel
Montana State University

Oliver de Peyer
National Institute for Medical Research

Peter Pilewskie
Ames Research Center

Andrew Pohorille
Ames Research Center

Chris Potter
Ames Research Center

Lee E. Prufert-Bebout
Ames Research Center

M. Rabbette
Ames Research Center

Christopher D. Raleigh
Ames Research Center

R. Pamela Reid
University of Miami

Petra Rettberg
DLReV Institute of Aerospace (Germany)

Nina D. Rosenberg
Lawrence Livermore National Laboratory

Lynn J. Rothschild
Ames Research Center

Lourdes Lloret y Sanchez
UNAM (Mexico)

Scott Sandford
Ames Research Center

Peter Schultz
Brown University

Ricardo Seda
University Turabo (Puerto Rico)

Norman Sleep
Stanford University

Stephen Smerdon
National Institute for Medical Research

Roger E. Summons
Massachusetts Institute Of Technology

Jack Szostak
Harvard University

Margaret A. Tolbert
University of Colorado Boulder

Owen B. Toon
University of Colorado Boulder

Matilde Trivi
INIDEP (Argentina)

Catherine J. Tsairides
Ames Research Center

Kendra Turk
University of California Santa Cruz

Pieter T. Visscher
University Of Connecticut

David Ward
Montana State University

Kimberly Warren-Rhodes
Ames Research Center

Nilda Weiler
National Patagonic Center

Richard Weinstein
University of Tennessee

Michael A. Wilson
Ames Research Center

Ray Wolstencraft
Royal Observatory (Scotland)

Bridget Wyatt
California State University San Jose

David D. Wynn-Williams
British Antarctic Survey UK

Richard Young
Ames Research Center

Kevin Zahnle
Ames Research Center

Richard Zare
Stanford University

M. Zolensky
Johnson Space Center

Executive Summary

The research efforts of the Ames team integrate a variety of disciplines around three scientific themes that address the context for life, the origin and early evolution of life, and the future of life.

Context for Life. We investigate both the chemistry and the environments conducive to life's origin. First, we trace, spectroscopically and chemically, the cosmic evolution of carbon compounds from the interstellar gas and dust to protoplanetary nebulae, planetesimals, and finally onto habitable bodies. Second, we probe the history of abiotically produced molecules of biological significance. Both investigations rely on spectral and chemical studies of realistic laboratory analogs tightly coupled with quantum chemical calculations followed by astronomical searches.

We investigate the habitability of planets by identifying and quantifying those factors that collectively determine the inner and outer limits of the circumstellar habitable zone. For example, (1) water must have been delivered to the planet; and (2) climatic conditions must allow surface liquid water to persist. Thus we focus on the origin and physical state of water, a study that depends on the sources of the water, the cycling of water and other volatiles between the surface and interior of a planet, and the detailed climate of the planet.

Origin and Early Evolution of Life and its Biosignatures. Do all habitable planets in fact become inhabited? We are identifying specific segments of this problem that are amenable to computational and laboratory investigation. We are testing the hypothesis that the most primitive protocells were structures built of evolving components related to those present in contemporary cells, but functioning without genomic control. We are defining simple biomolecular systems that can perform essential cellular functions, and we will determine conditions under which they can work together in a cellular environment.

We are investigating early microbial ecosystems by combining paleohistorical studies with experimental investigation of representative contemporary microbial ecosystems, and with model building. An improved understanding of long-term evolution of our own biosphere and the biogeochemical cycles that influence the environment will help us to assess the prospects for survival of other biospheres and to develop a strategy to find them by interpreting their biosignatures. Such biosignatures will assist our search for a potential martian biosphere, and to recognize possible spectroscopic signatures of inhabited planets around another star. We are also examining the effects of varying levels of oxygen upon the photochemistry of atmospheric constituents.

Future of Life. We investigate the effect of rapid environmental change on ecosystem properties and the potential for survival and biological evolution beyond the planet of origin. We are defining environmental factors that drive ecological change in South America. We also analyze preserved records of past change ultimately to predict future trends. We are exploring the effects of various forms of radiation upon the survival of life in extreme environments, including space. Our work includes developing methods for assessing radiation damage, examining specific biota for radiation resistance, and conducting exposure experiments that include space flight.

Training, Education, and Public Outreach. Our approach involves several elements. Source material is developed through a partnership between our scientists and participants in the professional training and development programs. This material is translated into curricula, web site content, public presentations, and other education or public outreach products by professional curriculum developers and communication specialists. Products are distributed to selected target groups (i.e., students, educators, and the public, including underrepresented minorities).

Highlights

- Vesicles assembled spontaneously from molecules formed in an interstellar ice simulation (Dworkin et al [2001] Proc. Nat. Acad. Sci. 98, 815-819.). Similar vesicles perhaps played key roles abiotically and thus have great astrobiological importance. This work garnered extensive positive international media attention (e.g., front page coverage on the Washington Post and The International Herald Tribune; radio interviews on NPR and in Canada, television, etc.).

- The photochemically-driven oxidation of polycyclic aromatic hydrocarbons (PAHs) in interstellar ices can explain the presence and deuterium enrichment of oxidized aromatics in meteorites. Some of these compounds (such as naphthoquinone) play key roles in the biochemistry of some archaea.

- Hydrothermal circulation may occur in the martian regolith and may significantly thin the surface layer on Mars at some locations, due to the upwelling of warm convecting fluids driven solely by background geothermal heating. This study is relevant to understanding the seepage channels discovered by the Mars Global Surveyor spacecraft now in orbit about Mars.

- Studies of the first proteins of non-genomic origin suggest that functional proteins are sufficiently common in protein sequence space that they can be discovered by entirely stochastic means, such as presumably operated when proteins were first used by protocells.

- The production of adenosine triphosphate (ATP) using light as the external source of energy was coupled to an ATP-dependent metabolic reaction. However, the coupling was efficient only under a narrow range of conditions that were favorable for both reactions. This work indicates that one of the main challenges in the origin of life was the integration and coordination of different cellular processes.

• A theoretical model indicated that collections of proteins could grow and evolve even in the absence of a genome. This process was markedly accelerated when it was coupled to energy-driven activation of monomers.

• Microbial mats produce quantities of hydrogen, carbon monoxide and methane that indicate a potentially important role for archean photosynthetic microbial mats in shaping both the composition and oxidation state of the ancient atmosphere.

• Microbial biofilms influence the accretion, lamination and early lithification of carbonate stromatolites. These processes can collectively create biosignatures for the microbial mat ecosystem.

• An extensive review article addressing extremophiles was published in Nature magazine. (Rothschild, L.J. & R.L. Mancinelli [2001] Life in extreme environments. Nature [London] 409: 1092-1101)

Roadmap Objectives

#1
Sources of Organics on Earth

#2
Origin of Life's Cellular Components

#11
Origin of Habitable Planets

#13
Extrasolar Biomarkers

Project

Chemical Building Blocks

Senior Project Investigator(s):
L. Allamandola, S. Sandford, M. Bernstein

Accomplishments

We investigated formation of biogenic compounds in cosmic ices and have found evidence for the production of many very interesting biogenic compounds with and without polycyclic aromatic hydrocarbons (PAHs). The PAH carbon compounds are widespread throughout space. Our team is the first to study their ice photochemistry under interstellar (IS) and early solar system conditions. Our publication on producing self-organizing, membrane-forming (primitive protocell?) molecules under harsh IS conditions generated enormous public response. We also made significant progress on PAH photochemistry in ices, studying side-group addition and isotopic enrichment. Together, these results may have important implications for the origin of life.

Highlights

• Our work on the generation of vesicles from spontaneous assembling of molecules formed in an interstellar ice simulation (Dworkin et al., 2001; Proc. Nat. Acad. Sci. 98, 815-819) was of great astrobiological significance and garnered an exceptional amount of positive media attention including pieces on the front pages of The Washington Post and The International Herald Tribune; radio interviews on NPR's "All Things Considered", Canada's "Quirks and Quarks", and others programs, as well as TV news coverage.

• In our most recent work on the oxidation of PAHs in interstellar ices we showed that interstellar photochemistry can explain the presence and deuterium enrichment of oxidized aromatics in meteorites. Some of these compounds (such as naphthoquinone) play key roles in the biochemistry of some archaea.

NASA Mission Involvement

Scott Sandford is a science Co-Investigator on the Stardust discovery mission. Launched in 1999, this mission will return samples of interstellar dust from our solar system as it flies by a comet called Wild-2 in January 2004. Stardust will return to Earth in 2006.

Scott Sanford is currently proposing for the Astrobiology Explorer (ABE). This mission is a space-based IR (infrared) telescope that would search for signatures of organic molecules considered indicative of the chemical processes integral to life as we know it.

Future Directions

• Chemically analyze the cosmic ice analogs and follow their photochemical and isotopic evolution, with emphasis on identifying the biogenically interesting species produced

• Identify the collective properties of the membrane (protocell) forming molecules

• Expand the PAH photochemical studies to realistic interstellar and cometary ice analogs

• Compare the compounds made in our interstellar ice simulations to those delivered to Earth by meteorites and interstellar dust particles

Habitable Planets

Project

Senior Project Investigator(s):
R.E. Young

Roadmap Objectives

#11
Origin of Habitable Planets

#12
Effects of Climate & Geology on Habitability

Accomplishments

Part of our work has examined the possible evolution of meteorites that might have played a role in bringing water and other volatiles to the early Earth. Results support a scenario in which low-temperature aqueous alteration of an anhydrous CM parent body and melt water from H_2O and CO_2 ices produce the altered assemblage observed in CM carbonaceous meteorites.

Three-dimensional simulations were conducted to model thermohydrologic processes on a planet with a frozen surface heated from below. The results suggest that hydrothermal circulation may occur in the martian regolith and may significantly thin the surface layer on Mars at some locations due to the upwelling of warm convecting fluids driven solely by background geothermal heating.

Laboratory and theoretical studies were accomplished concerning how carbon dioxide ice clouds affect the greenhouse effect, and hence the outer boundary of the habitable zone. Study of the microphysical properties of carbon dioxide clouds show that such clouds are unlikely to play an important role in the early greenhouses on Earth or Mars. Essentially the clouds warm the atmosphere, which tends to dissipate the clouds. The model was based on lab studies of the nucleation and growth of carbon dioxide. In circumstances where carbon dioxide clouds might have existed in the early martian atmosphere, we find that carbon dioxide clouds could be strongly warming, but the clouds can cool the surface if they are low and/or optically thick.

A review by Kasting of Ward and Brownlee's "Rare Earth" hypothesis was published in Perspectives in Biology and Medicine. Ward and Brownlee's book argues that Earth-like planets are rare, based on a number of biological, geological, and astronomical arguments. Several of these arguments are challenged in the review by Kasting.

We have examined the carbon dioxide cycle on the Earth immediately after it formed. The question of whether a hot (~100ºC) surface could long be maintained was also investigated. That Earth formed hot has been more-or-less accepted since at least the time of Lord Kelvin. The issue is how long did it stay hot? No stable feedback mechanism that could maintain a surface temperature in the vicinity of 100ºC was identified.

Analysis of data returned from the Clouds and Earth's Radiant Energy System (CERES) instrument onboard the Earth Observing System (EOS) satellites shows the same behavior as we found in data from the predecessor Earth Radiation Budget Experiments (ERBE) over the warm pool in the Pacific Ocean. Namely, the outgoing infrared (IR) radiative energy flux maximizes and then decreases as a function of increasing sea surface temperature, a signature of the runaway greenhouse effect.

Highlights

Three dimensional simulations of thermohydrologic behavior on a planet with a frozen surface heated from below were completed (Travis, Rosenberg, and Cuzzi). The results suggest that hydrothermal circulation may occur in the martian regolith and may significantly thin the surface layer on Mars at some locations due to the upwelling of warm convecting fluids driven solely by background geothermal heating. This study is relevant to understanding the seepage channels discovered by the Mars Global Surveyor spacecraft now in orbit about Mars.

Cross Team Collaborations

We have been actively collaborating with Jim Kasting (Pennsylvania State University Team), as well as Brian Toon and Maggie Tolbert (University of Colorado, Boulder Team), on studies of the microphysics and radiative transfer properties of CO_2 ice clouds.

These collaborations have resulted in the incorporation of their microphysics and radiative transfer models into the Mars circulation model developed at Ames. This research work is done to assess whether CO_2 ice clouds might have been a significant factor in determining the outer boundary of the habitable zone in our solar system. The same collaborations will also be employed to study organic hazes, which may have been important for understanding Earth's early climate, and perhaps the early climate of Mars as well.

NASA Mission Involvement

Our project work includes plans to derive stratospheric temperature profiles for different O_2 and O_3 amounts for early Earth's atmosphere. This calculation is highly relevant to NASA's proposed Terrestrial Planet Finder mission.

The radiative transfer code is being developed for study of habitable planet GCMs (General Circulation Models ... planet atmospheric circulation). It will be used in conjunction with the existing photochemical model of James Kasting (Pennsylvania State University NAI Team) and the LBL (Line-by-Line) radiative transfer model to derive synthetic IR (infrared) spectra of atmospheres containing various amounts of O_2.

Jack Lissauer is on the Kepler Mission team as a Co-I. The Kepler Discovery Mission 2006 spacecraft will use photometric observation to detect Earth-like planets in Earth-like orbits around stars in the disk of our Galaxy.

Future Directions

- Study the possible effects of organic haze layers on paleoclimate: Laboratory and theoretical studies will be conducted to characterize organic haze particles that may have been important in the climatology of Earth's early atmosphere, as well as perhaps that of Mars.

- Develop a radiative transfer code suitable for use in a habitable planets general circulation model (GCM): We will use this code in conjunction with our existing photochemical model and the LBL (Line-By-Line) radiative transfer model to derive synthetic IR spectra of atmospheres containing various amounts of oxygen. The code will be used in a Mars GCM to study early Mars climatology and the effects of clouds.

- Study further the partitioning of carbon dioxide between reservoirs on the Hadean Earth: These studies are relevant to understanding the thermal structure of early Earth's atmosphere, which influences atmospheric chemical and photochemical processes.

- Model the rise of oxygen in the early Earth's atmosphere: Theoretical understanding of the rise of oxygen suffers from an absence of quantitative reasoning. To this purpose we are devising a simple model that can address quantitatively the various fluxes and reservoirs that describe Earth's carbon cycle and the buildup of oxygen.

- Analyze the latest satellite data concerning the super greenhouse effect over the warm pool of the Pacific Ocean: Simulate the outgoing IR flux at the top of the atmosphere using climatological humidity profiles to explain the observed flux limitation as a function of sea surface temperature. This is relevant to understanding the runaway greenhouse effect on other planets.

- Explore water redistribution throughout a planetary body as it heats up, as a function of the assumed thermal source responsible for supplying the heat, the size of the parent body, rock properties such as porosity and permeability, and the initial quantity and distribution of ice. We plan to take advantage of a newly developed modeling code, MOR.ICE, and use it for the simulations.

Roadmap Objectives

Project

Early Metabolic Pathways

Senior Project Investigator(s):
A. Pohorille

#2
Origin of Life's Cellular Components

#3
Models for Life

Accomplishments

The main, long-term goals of this work have been (a) to develop protein enzymes representative of those that existed on the early Earth, (b) to couple their catalytic activity in model protocells to external sources of energy and nutrients, and (c) to determine conditions under which such protocellular systems can evolve using theoretical modeling.

Functional proteins that efficiently bind adenosine triphosphate (ATP) were selected from a very large library of random sequences. These proteins have been characterized in detail. It appears that they form folded structures. They were shown to be highly selective – they bind ATP but not guanosine triphosphate or cyclic ATP. They require zinc ions, but not magnesium ions, to function and contain conserved cysteine residues. This suggests that their structure may be similar to those of zinc finger proteins.

A simple bioenergetics system consisting of bacteriorhodopsin and ATP synthetase incorporated into phospholipid liposomes was coupled to thermophilic ADP-forming acetyl-CoA synthetase, which in the presence of acetate, CoA and magnesium ions produces acetyl-CoA and converts ATP back to ADP, leading to a light-dependent accumulation of acetyl-CoA. The conditions for maximum production of acetyl-CoA were optimized.

It was demonstrated through theoretical modeling that proteins in protocells could not only grow but in the process yield distributions of catalytic efficiencies quite different than the starting distributions. This is the first example of protocellular evolution in the absence of a genome. The model was extended to include peptide activation and activated ligation. These extensions increased the rate of growth in the catalytic potential of the protocell, as well as the maximum length of peptides produced. This suggests that protocells capable of capturing and storing energy and then using that energy to activate peptides for further polymerization might have been able to increase their catalytic potential rapidly.

Highlights

- Studies on the first proteins of non-genomic origin suggest that functional proteins are sufficiently common in protein sequence space that they may be discovered by entirely stochastic means, such as presumably operated when proteins were first used by protocells.

- The production of ATP using light as the external source of energy was coupled to an ATP-dependent metabolic reaction. However, the coupling was efficient only under a narrow range of conditions that were favorable for both reactions. This clearly demonstrates that one of the main challenges in the origin of life was the organization of different cellular processes such that they could work in concert.

- Using theoretical modeling it was shown that collections of proteins could grow and evolve even in the absence of a genome. This process was markedly accelerated when it was coupled to energy-driven activation of monomers.

Cross Team Collaborations

Dave Deamer, a member of both the Pohorille and Allamandola teams, collaborated with Jason Dworkin, Scott Sandford, and Louis Allamandola to establish biophysical properties of photochemical products from an interstellar medium simulation.

Despite the initial reactants being very simple compounds such as CO (carbon monoxide), CH_3OH (methanol) and NH_3 (ammonia) embedded in thin films of water ice at 10 degrees Kelvin, it was discovered that a remarkable number of more complex molecules were synthesized with molecular weights ranging up to 400 kD. Some of these were amphiphilic molecules with physical properties resembling those present in carbonaceous meteorites, including the ability to form membranous vesicles.

A paper on this research (Dworkin et al.) appeared in January 2001 in the special Astrobiology issue of the Proceedings of the National Academy of Sciences USA. It attracted considerable media attention, including a front page article in the Washington Post and interviews for BBC and CNN.

Future Directions

- Selection of new proteins of non-genomic origin
- Coupling the energy generating system to chemical reactions that produce vesicle-forming material
- Extension of the current model of protein evolution to include several functions and their coupling

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#13
Extrasolar Biomarkers

Project

Early Microbial Ecosystems

Senior Project Investigator(s):
D. J. Des Marais

Accomplishments

We are investigating the microbial populations and ecological processes that influence adaptation and evolution, and that create signatures of life (e.g., biomarkers such as gases, chemical compounds and morphologic features in sediments). In microbial mats at Guerrero Negro, Mexico, the budgets of oxygen and carbon have been found to vary between subtidal and intertidal mats, reflecting, in part, differences between the relative importance of oxic respiration versus sulfate respiration during organic degradation. Significant methane production occurred in unexpected places, such as the near-surface layers of subtidal mats and in the presence of high sulfate concentrations. Substantial hydrogen production occurred in mat surface layers, and is perhaps attributable to fermentation and also nitrogenase activity. Such hydrogen fluxes indicate that benthic microbial ecosystems might have played a significant role in determining the oxidation state of the Earth's early atmosphere. The budgets of volatile sulfur gases, such as dimethyl sulfide, indicate that a delicate balance exists between biological and nonbiological processes of production and oxidation. A variety of low molecular weight thiol biomarkers was catalogued. Green nonsulfur bacteria from Yellowstone and

Guerrero Negro mats revealed a diverse, more deeply divergent population of phototrophs than previously represented by *Chloroflexus* spp. The roles played by ultraviolet light in the structure, function, and evolution of cyanobacteria was extensively summarized in a published review article. The Ames greenhouse facility "Archean Gardens" was enhanced to create important research opportunities involving samples collected on field trips and involving information systems technology at Ames. Long-term monitoring of mats transplanted from Baja California indicates that this facility can successfully maintain hypersaline mats in their near-original state for several months after field collection. Lithified laminae in Bahamian carbonate stromatolites are formed by biofilm communities that alternately dissolve and precipitate carbonate during early diagenesis. Laminated stromatolitic fabrics arise due to successions between different types of biofilm communities. Recent observations of the phylogeny of photopigments, together with the geologic record of stromatolites, indicate that anoxygenic phototrophic bacteria arose sometime prior to 2.8 Ga ago, and prior to the development of cyanobacteria.

Highlights

- A study of the taxa and distribution of bacteria related to *Chloroflexus* spp. has revealed a broader diversity than was previously recognized for green nonsulfur photosynthetic bacteria (Nuebel et al., Applied and Environ. Microbiol., in revision).

- Microbial mats produce amounts of hydrogen, carbon monoxide and methane that indicate a potentially important role for photosynthetic microbial mats in shaping the oxidation state of the ancient atmosphere (Hoehler, et al., Nature, in press).

- Microbial biofilms influence the accretion, lamination and early lithification of carbonate stromatolites. These processes collectively serve as biosignatures for the microbial mat ecosystem (Reid et al., Nature, 406:989, 2000).

Field Trips

Guerrero Negro, Baja California Sur, Mexico, May 30 – June 12, 2001.

A major field trip was conducted to Guerrero Negro, Baja California Sur, Mexico. Attendees included B. Bebout, S. Carpenter, D. Des Marais, J. Dillon, F. Garcia-Pichel, J. Farmer, T. Hoehler, M. Hogan, M. Huerta-Diaz, S. Miller, K. Turk, and P. Visscher.

Cross Team Collaborations

Our research on early microbial ecosystems has involved collaborative interaction with four particular NAI Teams, as described below.

Arizona State University Team

A field trip to Guerrero Negro, Baja California Sur, Mexico (June 2001) marked the beginning of an interdisciplinary study of the hypersaline microbial mats at Guerrero Negro. This study carries implications for our understanding of the early evolution of microorganisms and animals, as well as for the interpretation of biomarker gases and sedimentary organic matter.

MBL Team

Our activity in the Ecogenomics Focus Group with MBL Team members is just beginning. It will address the structure and function of microbial communities from three perspectives: biogeochemistry, population distributions, and gene expression at the ecosystem level.

University of Colorado, Boulder Team

Novel methods have been developed by the research group of Dr. Norm Pace for investigating sequences of DNA molecules extracted from natural microbial populations. These new data are creating a broad, new perspective on the diversity of microbial life. Samples of subtidal photosynthetic microbial mats are collected to determine the richness of microbial species present. It is anticipated that numerous, previously unidentified microorganisms will be revealed.

University of Washington Team

Sulfate-reducing bacteria play major roles in microbial mats. They are both consumers of fermentation products and hydrogen, as well as producers of sulfide that sustains many other microbiota. Our interactive group will identify and determine the distribution of various sulfate-reducing bacteria species and also examine the association between populations of cyanobacteria and sulfate-reducing bacteria.

NASA Mission Involvement

These studies will strengthen the systematics for interpreting the microbial fossil record and thereby enhance astrobiological studies of martian samples. Our models of biogenic gas emissions will enhance models of atmospheres that might be detected on inhabited extrasolar planets. This work therefore addresses the question: How can other biospheres be recognized?

The Co-PI, Des Marais, has been extensively involved in mission planning and preparation. Accordingly, he establishes linkages between this research and NASA programs and missions. He is a member of both MEPAG (Mars Exploration Payload Analysis Group) and TPF-SWG (Terrestrial Planet Finder-Science Working Group). He participated in the Mars Program reorganization effort during the past year, particularly during key workshops in the summer of 2000. He is chair of the TPF-SWG subgroup on Biomarkers and is currently leading the preparation of a major white paper to articulate the goals and objectives for seeking biosignatures and evidence of habitability in extrasolar planets. Des Marais is an interdisciplinary scientist for astrobiology on the Mars Exploration Rover mission, which is scheduled for a 2003 launch. In this capacity and as member of the Science Operations Working Group, Des Marais will participate in the strategic planning and operation of the 2003 rovers.

Year 3

Key programmatic and mission-related activities during NAI Year 3 are listed below:

7/18/2000 Lunar and Planetary Institute, Houston, TX. Mars Exploration Workshop (3 days), Program Planning

8/21-24/00 JPL, Pasadena, CA. Mars Synthesis Meeting - Concepts and Approaches for the Robotic Exploration of Mars: Recommendations for basic Mars science (4 days)

9/18/2000 JPL, Pasadena, CA. Science Operations Working Group Meeting (2 days)

9/26/2000 JPL, Pasadena, CA. Terrestrial Planet Finder Science Working Group (2 days)

12/12/2000 JPL, Pasadena, CA. Terrestrial Planet Finder Science Working Group (2 days)

1/24/2001 Center for Mars Exploration. Mars Landing Site Workshop (3 days)

3/29/2001 JPL, Pasadena, CA. Mars Exploration Rover Working Meeting (SOWG) - Mars Yard Rover Test (2 days)

4/30/2001 JPL, Pasadena, CA. Mars Exploration Rover Working Meeting (SOWG) - Mojave Desert (8 days)

5/23/2001 JPL, Pasadena, CA. Mars Exploration Rover Working Meeting (SOWG) (3 days)

Future Directions

- Explore the impact of environmental parameters such as temperature and availability of oxidants upon the structure and function of subtidal mats, with an emphasis on their production of biomarker compounds, including gases

- Conduct parallel genetic studies of photosynthetic populations to establish relationships between population richness, biomarkers and ecosystem function

- Conduct a long-term study of the response of microbial mats to declining sulfate concentrations, using the “Archean Gardens” greenhouse testbed facility. This test will simulate the low-sulfate conditions that existed in the marine environment during the Archean Eon.

- Collaborate with Drs. M. Huerta Diaz, B. Thamdrup, C. Johnson and B. Beard to explore the biogeochemistry of sulfur, iron, and other metals in microbial mats and related sediments

- Culture and identify sulfate reducing bacteria obtained from the cyanobacterial mats at Guerrero Negro. This work will be coupled to genetic studies to identify which taxa play major roles in these hypersaline mats.

Roadmap Objectives

Project

The Future of Life: Rapid Rates of Change

Senior Project Investigator(s):
H. D'Antoni

#14
Ecosystem Response to Rapid Environmental Change

Accomplishments

We have identified areas in Patagonia for research on environmental events introducing rapid changes; a larger proposal is in progress. A paper is being prepared on modern pollen dispersal as an ecological indicator of retrogressive biogeochemical cycles. Another paper is in progress on global forcing factors on South American ecosystems. A digital movie is being prepared to show environmental change in South America at monthly scales. We continue to work on data mining and analysis for ecological applications. A dissertation is in progress on fine-scale ecological research in Patagonia. One of our team also has a book in press on the climates of Amazonia.

This year has been crucial in identification of cause-effect relations of the South American biota and the forcing factors of the environment. Predictability of the forcing factors and the vegetation responses were demonstrated. Work is in progress on the multivariate analysis of results.

The large international team continues to produce valuable results. The INIDEP team created an aggressive program to collect ecosystem signals throughout the Rio de la Plata watershed and formed a large team of collaborators from national and provincial agencies. The Ecuadorian team is tracing the history of run-off water for human survival in a research project that overlaps with our astrobiology interests in this area. We are radiocarbon dating the archeological evidence (26 samples submitted to the Geochronology Lab of University of Arizona) and cooperating with our Ecuadorian colleagues in areas of common interest, such as identifying sites of massive forest extinction in Patagonia and dating evidence using the potassium-argon technique.

Highlights

- First time retrogressive biogeochemical cycles assessed

- Remote sensing ecosystem data as predictor of physical variables of the environment

- Physical variables of the environment as predictors of ecosystem's net primary productivity

Field Trips

Southern Patagonia, Argentina, January, 2000.

Field Work was carried out in Southern Patagonia, with Nilda Weiler, National Patagonic Center (Argentina). This was a search of geoforms indicative of major ecological disturbance.

Ecuador's Dry Forest, September 2000.

Fieldwork was carried out in Ecuador's Dry Forest, with Jorge Marcos, Polytechnic School of the Littoral, Ecuador. This was a search of indicators of ancient human actions on the environment in connection with ENSO effects.

NASA Mission Involvement

This research project connects most directly with the retrogressive study of biogeochemical cycles and carbon cycle questions within NASA's Earth Science Enterprise.

Our research pursues understanding of the science relating to Objective #14 of the Astrobiology Roadmap. This objective is: "Determine the resilience of local and global ecosystems through their response to natural and human-induced disturbances." Our research project also meets some of the goals of NASA's Earth Science Applications Program.

Future Directions

- Describe past environmental events affecting the long-term ecosystem process of South America

- Measure human impact on biogeochemical cycles in the dry forests of Ecuador and compare ENSO and non-ENSO years (collaboration with Marcos' team in Ecuador)

- Measure ENSO effects on the Rio de la Plata watershed (collaboration with Lasta's team in Argentina)

- Connect selected sites to the changes of South American ecosystems through time

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#10
Natural Migration of Life

#12
Effects of Climate & Geology on Habitability

#14
Ecosystem Response to Rapid Environmental Change

#15
Earth's Future Habitability

#16
Bringing Life with Us Beyond Earth

#17
Planetary Protection

- Evaluate atmospheric correction to further understand the connections of environmental forcing factors and the effects on the biota

- Initiate research in the biomolecular signatures of ecosystems at different phenologic phases

Project

Life Beyond the Planet of Origin

Senior Project Investigator(s):
L. Rothschild

Accomplishments

Progress during the first part of this project began September 1, 2000. Several team members have isolated halophiles from nature, and testing has begun to determine radiation resistance, as part of a study to evaluate linkages between mechanisms of resistance to salt and to ionizing radiation. A new study has begun on the DNA damage resistance of *Dunaliella salina*, an extremely halophilic eukaryotic alga. We have explored new techniques to assess damage resistance, and ways to measure gene activation upon rehydration of yeast.

Previous experiments have shown that halophilic organisms isolated from salterns and intertidal environments can survive a two-week exposure to the space environment. Using the space simulation facility at the DLR in Cologne, Germany, we have completed a series of ground simulation experiments using a halophilic cyanobacterium from the genus *Synechococcus* spp. We have exposed this organism to vacuum desiccation and ultraviolet (UV) radiation. These experiments reveal that these organisms can survive exposure to UV radiation and vacuum desiccation for at least six months and probably longer.

Highlights

- Definitive survey of extremophiles published in Nature. [Rothschild, L.J. & R.L. Mancinelli, 2001; Life in extreme environments, Nature (London) 409:1092-1101]

- Opportunity to define astrobiology to the public at large through outreach and publication of the entry for astrobiology for the McGraw-Hill Encyclopedia of Life Sciences (Rothschild)

Field Trips

Cargill Salt Company, San Francisco Bay, June and August, 2000.
Purpose: To isolate halophiles. Participants: Rettberg (DLR), Lloret y Sanchez (UNAM), Mancinelli (SETI Institute) & Rothschild (NASA ARC).

China, November 2000.
Purpose: To isolate halophiles. Researcher: Warren-Rhodes (NASA ARC).

NASA Mission Involvement

ESA (European Space Agency) was the first to send out an Announcement of Opportunity (AO) to fly biological samples that could be exposed to the space environment in a controlled manner aboard the space station, Mancinelli wrote a proposal in response to this announcement. His proposal was reviewed and accepted to fly on the ESA external platform of the ISS (International Space Station).

The Mancinelli portion of this ISS experiment is being partially funded by NAI. Up until two weeks ago, NASA had not released an Announcement of Opportunity to fly equivalent experiments aboard the International Space Station. We plan to propose to this AO and suggest organisms and experiments based on the work conducted through NAI.

Future Directions

- Characterize halophiles and desiccation resistant organisms for resistance to UV and oxidizing damage. Specifically we will focus on natural isolates, *Dunaliella*, and possibly other eukaryotes. Techniques developed in Rothschild's lab in the last year, ranging from fluorescence detection of reactive oxygen species to damage, will be applied.

- Using the space simulation facility at the DLR (German Aerospace Center) in Cologne, Germany, expose most resistant organisms to UV radiation and simulated space vacuum; these organisms include eukaryotes and prokaryotes.

- Initiate gene chip studies to identify genes activated by rehydration. This will be conducted on yeast.

- Integrate and initiate work by other team members, for example, desert crusts as model systems (Johansen), theoretical studies (Wolstencroft), and effects of impact on survival (Shultz).

Education & Public Outreach

Ames NAI Team Poster

The Ames team created a poster for the NAI Annual Meeting that illustrated the partnership between the research team and outreach. Charting the origins, evolution and distribution of life in the universe is an enterprise that requires the participation of astronomers, biochemists, biologists, and Earth and planetary scientists. The Ames team includes investigations from all of these disciplines, therefore its research contributes knowledge that strengthens our understanding of the linkages between our origins, evolution and destiny. This team provides a unique contribution to public education and outreach as a source of continuing knowledge that informs teachers, excites students and involves the general public.

Astrobiology Academy

The NASA Ames Astrobiology research team participates in the Astrobiology Academy by presenting lectures to the students, offering lab tours, and hosting student interns from the Academy. The students have a research mentor from Ames who works with them as they develop their projects and meets with them to oversee the research developments.

Astrobiology: The Search For Life Curriculum

TERC and NASA have developed an interdisciplinary high school course using the science of astrobiology as its unifying, underlying structure. Through a series of inquiry-based activities designed according to the theme of the search for life on other planets, students will explore diverse concepts in chemistry, physics, biology, Earth and space science, and engineering. These concepts are truly integrated for ninth grade students as a sequential, coherent stream of units, chapters, activities, and long term investigations.The astrobiology curriculum is currently being field tested nationally by educators and students.The lead NAI teams participating in the national review include the Ames Research Center, Pennsylvania State University and Jet Propulsion Laboratory. This astrobiology product is scheduled for publication with a national publishing company in January 2002. The national publisher will provide extensive marketing, dissemination and professional development for prospective users of the curriculum reaching an estimated eight thousand classrooms.Sample activities, recent developments, and an outline of the course are located at
http://astrobio.terc.edu

Product Reviews

Astrobiology: The Search For Life

The curriculum product review for "Astrobiology: The Search for Life" was organized with TERC, the Ames Astrobiology team, Pennsylvania State University, and Jet Propulsion Laboratory. Catherine Tsairides, Outreach Project Lead for the

Ames team directed the science review with the lead teams, researchers, and TERC. In March the curriculum was reviewed by the Origins education evaluator and an Origins Scientist. The JPL team is reviewing portions of the program that focus on instrumentation while the Ames and Penn State teams are looking directly at the science content and relevant research in each lesson.

EXOQUEST-Experts in Science ASTROBIOLOGY

This multimedia product developed at NASA Classroom of the Future provides students with the opportunity to ask the same kinds of questions and conduct the same kinds of research as scientists from NASA and other organizations around the world. EXOQUEST creates a link between students and scientists as a means of integrating NASA's experience and expertise into the middle and high school curriculum. Students use EXOQUEST interface to travel on virtual journeys to destinations in the solar system and beyond. At each destination students conduct investigations that include hands-on and simulated experiments. These investigations pose problems that focus on different areas of research, providing an interdisciplinary approach to science and the scientific method. Activity modules include: Extreme Environments, Ocean on Europa, Search for Extrasolar Planets, Water in the Solar System, Extraterrestrial Communications. The “Experts in Science” video consists of excerpts from interviews with twenty-seven astrobiology researchers. This piece was developed for middle school students and will be marketed as a career video for distribution with a publisher.

Outreach Review Panel

The Ames Astrobiology team has been represented on the review panels focusing on the outreach proposals and projects submitted for funding. The team has had representation on the following panels, Cooperative Agreement Notice-NAI, Space Telescope Science Institute, and Office of Space Science-Product Review.

Coursework

Hector D'Antoni presented "Astrobiology and Past Environments" at Guayaquil, Ecuador, the University of Mar del Plata and the University of Puerto Rico, Mayaquez. His research module focuses on Rapid Rates of Change.

Distribution-Astrobiology Educator Guide

The Astrobiology Educator Guide was produced to provide teachers (grades 5-10) a way to explore astrobiology in their classrooms. Conferences, presentations, workshop, exhibit, poster session and meetings were the venue for distribution of the astrobiology educator guide. These included the following activities: Arizona State University teacher workshop, Tempe, Arizona, American Geophysical Union, San Francisco, California, American Astronomical Society, San Diego, California, NASA Educator Workshops, NASA Ames Research Center, Moffett Field, California, Lab and Greenhouse Tours, NASA Ames Research Center, Moffett Field California, Astronomical Society of the Pacific, San Francisco, California, Student Presentation Ravenswood School District, East Palo Alto, California.

Educational Presentations/Workshops

The Ames Astrobiology Research team gave 12 presentations, hands-on workshops, and lectures targeted at students of all levels, university professors and educators. The themes address the value of scientific training in participating in space exploration, as well as the exciting details of astrobiological research. Workshops were given in CA, MT, AZ and NY.

FOCUS-Microbial Mats and Stromatolites

Brad Bebout created a focus article for the NAI website illustrating what microbial mats are and why NASA is interested in studying them. This site continues to be an ongoing piece for the general public and educational groups to access.

Hampton University Grant

Dave Des Marais is the Technical Monitor for the grant that Hampton University is receiving. He provides technical advice and reviews the progress of the development of a course in astrobiology. The team hosted a graduate student from Hampton University on her visit here to Ames. Brendlyn Faison of Hampton University was presented with astrobiology course outlines from our lead teams and the Astrobiology Educator Guide was sent as a product for them to review as they began development of their own astrobiology coursework at Hampton University.

Hewlett Packard Business Executive Lab Tour

A team of fifty corporate presidents, vice-presidents, managers, and software engineers requested a tour and presentation so that they could learn more about the astrobiology program. This international corporate team is affiliated with Hewlett Packard Corporation. They were interested in business partnerships and database development. Dave Des Marais and Tori Hoehler presented astrobiology and led the lab tours with Barry Blumberg, the Director of the NASA Astrobiology Institute, as the keynote speaker.

Human Biology #107, Astrobiology and Space Exploration

Human Biology, HB 107, is a formal course taught at Stanford University. It consists of 20 seventy-five minute lectures taught by several Ames space and life scientists. The topics range from the origin of the universe, the stars, planetary systems and origin and early evolution of life, and then address gravitational biology and the quest for humanity to move beyond Earth and inhabit the heavens. The students are exposed to a breadth of topics that encourage broad interdisciplinary inquiry into our place in the universe and our destiny. Despite these broad themes, the course still offers details about how research into our origins and how we engineer our survival and future in space are achieved at the technical level. The course thus stresses the integration of science and technology, and encourages students to span disciplines in their own future careers.

Marine Science Institute

The Marine Science Institute (MSI) is a nonprofit science and environmental education organization offering hands-on programs that excite students of all ages about science. The Institute has been offering programs for students throughout Northern California since 1970. Lynn Rothschild and her research team participate in developing and presenting Ames astrobiology research to the students during hands-on activities and during teacher training workshops throughout the year. Students spend a half-day as scientists, discovering the San Francisco Bay Estuary's ecosystem and discussing their own role within it. Students collect and examine plankton, run hydrology tests, and observe wetland ecology from the ship. They delve through mud samples, discovering the fascinating invertebrates that thrive at the bay's bottom. This program offers students the chance to enjoy the natural vitality of this area while learning valuable scientific skills.

Palo Alto Rotary Club

The Ames Astrobiology team developed a partnership with the Palo Alto Rotary Club. Pat DeVaney of Stanford University worked with Ames to develop a science program for the gifted population in East Palo Alto. These students were selected by their teachers as students that have potential for college with an emphasis on math and science. We outlined a program in collaboration with teams from Life Sciences, SOFIA, the Mars project and astrobiology to provide hands-on lessons and practicing scientists in the Mars, technology, and astrobiology missions while Catherine Tsairides presented the Astrobiology Resource Guide activities. Ames Outreach provided the students with a field trip to Ames and Space Camp setting up a fund raiser for Space Camp so that the Rotary Club could sponsor students to attend the week long Space Camp at Ames Research Center.

Presentations-Informal Education

The Ames Astrobiology team is involved in high leverage partnerships with public organizations that reach millions of members of the general public. We have developed partnerships with the national park system and museums across the country, making a long term sustained commitment to integrating education and outreach through collaborations with these agencies, Marine Science Institute, Redwood City, CA, Grand Teton National Park, Jackson Hole, WY, Kilauea Visitor Center, AfterDark in the Park Hawaii, Kilauea Visitor Center, Hawaii Volcanoes National Park, Hawaii, Varian Medical Systems, Palo Alto, CA, MIT Media Lab, Cambridge, MA, Eugene Natural History Society, Eugene, OR, California Academy of Sciences, San Francisco, CA, Yellowstone National Park, WY.

Professional Presentations

The Ames Astrobiology team presents their research and provides information on astrobiology to professional societies and conferences nationally and internationally. These included the following: ASEE Summer Fellowship, Ames Research Center, American Academy of Microbiology, Tucson, AZ, Gordon Research Conference, Plymouth, NH, Lunar and Planetary Institute, Houston TX, Artificial Life Symposium, Portland, OR, Mars Exploration Payload Advisory Group, Jet Propulsion Laboratory, Pasadena, CA, Marine Biological Laboratory, Woods Hole, MA, Ames Research Center, Moffett Field, CA, Center for Mars Exploration, American Society for Limnology and Oceanography, Rutherford Appleton Laboratory, UK.

Public Presentations

The Ames Astrobiology team participates in public outreach by providing astrobiology lectures, presentations, exhibit material, and demonstrations.

Research Mentoring

Lee Prufert-Bebout mentored the research project by Joanna Naymark "Isolation of Cyanobacteria and study of the Formation of Stromatolites, Ancient Fossils" as part of the Menlo School Biotechnology Research Course. This course was started and taught by Tanya Buxton in collaboration with Gene Connection, a San Mateo County partnership under BABEC (Bay Area Biotechnology Education Consortium) http://www.babec.org/SCCBEP,
http://www.smcoe.k12.ca.us/geneconnection/. This collaboration was also facilitated by the Palo Alto Chapter of the Association for Women in Science. Des Marais' team also mentored a research project by Rebekah Shepard on the connection between cyanobacterial distribution and lithification in modern marine stromatolites and Marlene Sosa on identifying ecological patterns of occurrence of pharmacologically active cyanobacterial metabolites.

Tours of Microbial Mats

The Ames Astrobiology team has a unique laboratory tour available to visiting researchers, NASA officials, local education groups, and outside organizations. Dave Des Marais, Brad Bebout and Tori Hoehler are the lead researchers that prepare their presentations based on the type of group that is scheduled to tour the labs. The public is given a unique opportunity to view the samples and hear from the principal investigators on the project. The tours occur on a continuous basis throughout the year with requests coming from the NAI , Public Affairs, Education and Public Outreach, and other NASA centers.

Yellowstone National Park

The Ames Astrobiology team has been working at Yellowstone National Park and would like to extend the research project into an integrated plan with outreach. Yellowstone National Park has close to three million visitors of all ages experiencing this diverse environment. The NASA Ames Astrobiology team, with other NAI Lead Team partners, sees the potential for investing their energy in an activity that has a large impact. The National Park Service is actively engaged in programs and projects development of educational materials and activities that contribute to education and public understanding of science. Collaborating across the Teams and with the National Park Service provides astrobiology research with the potential to reach audiences up to three million per year. The Ames Astrobiology team with Lynn Rothschild and David Ward from of Montana State in partnership with Arizona State and Jack Farmer propose bringing astrobiology into the Interpretative Research Program at Yellowstone National Park. The Ames Astrobiology team with its partners will support the NASA mission by providing exposure to the general public to the supporting enhancement of knowledge and skills, and to provide access to NASA information in science, mathematics, technology, engineering, and geography.

Publications

Back to Table of Contents

Chemical Building Blocks

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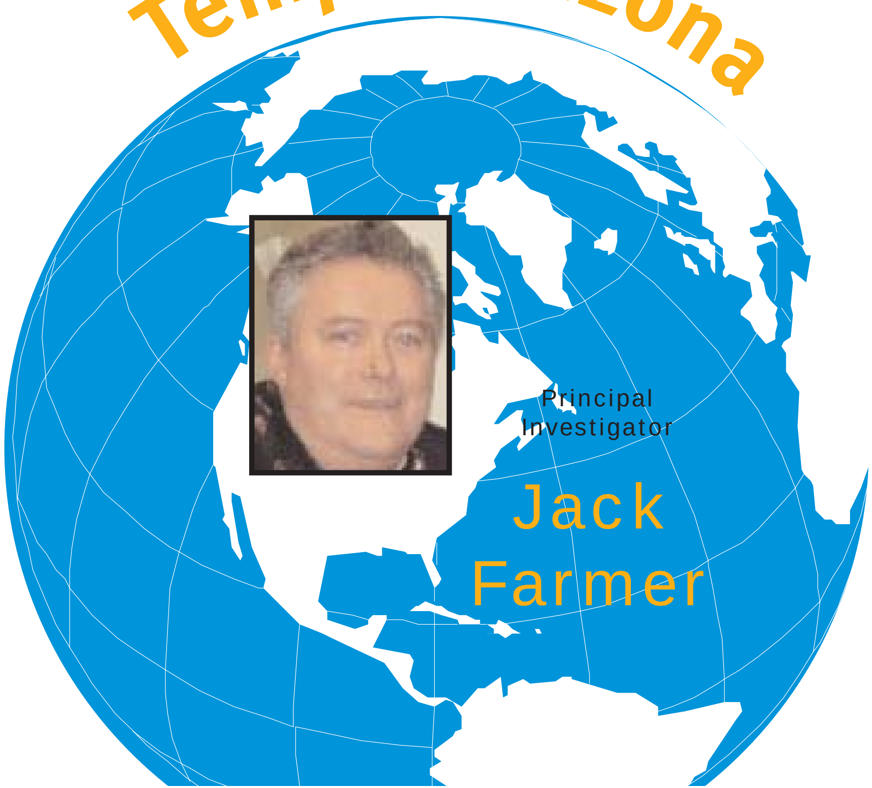
Back to Table of Contents

Year 3

Arizona State University

NASA Astrobiology Institute

Tempe, Arizona



Arizona State University Team Members

James Allen
Arizona State University

Andrew Armstrong
California Academy of Sciences

Dawn Ashbridge
Arizona State University

Alice Baldridge
Arizona State University

Carl Bauer
Indiana University

Brad M. Bebout
Ames Research Center

Gretchen K. Benedix
Virginia Tech.

Robert Blankenship
Arizona State University

Karl Booksh
Arizona State University

Daniel Brune
Arizona State University

Donald Burt
Arizona State University

Brandon Canfield
Arizona State University

Evan Carson
Arizona State University

Philip R. Christensen
Arizona State University

Frank Chuang
USGS Flagstaff

Antonio Cruz
UNAM (Mexico)

Melissa del Rosario
Arizona State University

Pet Della-Betta
Arizona State University

David Des Marais
Ames Research Center

Tami Diedrich
Arizona State University

Thomas E. Dowling
Arizona State University

Jeremy Draghi
University of Connecticut

Eileen Dunn
Arizona State University

Christopher Eagle Hawk
Arizona State University

Luis Eguiarte
UNAM (Mexico)

James J. Elser
Arizona State University

Ana Escalante
UNAM (Mexico)

Laura Espinoza
UNAM (Mexico)

William Fagan
Arizona State University

Sarah Fagents
Arizona State University

Jack D. Farmer
Arizona State University

Patricio Figueredo
Arizona State University

Tom Foltz
Arizona State University

Wayne Frasch
Arizona State University

Ferran Garcia-Pichel
Arizona State University

Darcy Gentleman
Arizona State University

J. Peter Gogarten
University of Connecticut

Julia Goreva
Arizona State University

Carolina Granados
UNAM (Mexico)

Ronald Greeley
Arizona State University

Reema Gulati
University of Connecticut

John Devens Gust
Arizona State University

Victoria Hamilton
Arizona State University

John R. Holloway
Arizona State University

Carolyn James
University of Puget Sound

Steve Kadel
Arizona State University

Anne Kelsen
Arizona State University

Jim Klemaszewski
Arizona State University

Paul Knauth
Arizona State University

David A. Kring
University of Arizona

Marcia Kyle
Arizona State University

Vanessa Lancaster
Arizona State University

Laurie A. Leshin
Arizona State University

Michael Lince
Arizona State University

Blair Lindford
Arizona State University

Donald R. Lowe
Stanford University

Trent McDowell
Arizona State University

Kat McFadden
Arizona State University

Eli Meir
University of Washington

H. Jay Melosh
University of Arizona

Susanne Meschter
Arizona State University

Wendell Minckley
Arizona State University

Jeffry Moersch
University of Tennessee

Ana L. Moore
Arizona State University

Thomas Moore
Arizona State University

John Moreau
Arizona State University

Julia M. Muccino
Arizona State University

David M. Nelson
Arizona State University

Peggy O Day
Arizona State University

Gary Odell
University of Washington

Robert Pappalardo
Brown University

Meredith Payne
Arizona State University

Elizabeth Pierazzo
University of Arizona

Beverly Pierson
University of Puget Sound

Sandra Pizzarello
Arizona State University

F. Gerald Plumley
University of Alaska

Jason Raymond
Arizona State University

Peter Roopnarine
California Academy of Sciences

Valeria Routt
Arizona State University

Steve Ruff
Arizona State University

Bruce N. Runnegar
University of California Los Angeles

Victor Scarpa
University of Puget Sound

Thomas G. Sharp
Arizona State University

Reshma Shial
University of Connecticut

Valeria Souza
UNAM (Mexico)

Carol Tang
California Academy of Sciences

Mary Turnispeed
College of William and Mary

Cindy Vandover
College of William and Mary

Hebe Vanegas-Farfano
Arizona State University

Willem Vermaas
Arizona State University

Pieter T. Visscher
University of Connecticut

Kenneth M. Voglesonger
Arizona State University

Brian Wade
Arizona State University

Sheri White
Woods Hole Oceanographic Institute

David Williams
Arizona State University

JoAnn Williams
Arizona State University

Lynda B. Williams
Arizona State University

Teri Williams
Arizona State University

Jin Xiong
Indiana University

Hua Zhang
University of Alaska

Olga Zhaxybayeva
University of Connecticut

Executive Summary

Accomplishments

During the third year of ASU's Astrobiology Program (The Origin, Evolution and Distribution of Life in the Solar System) we achieved substantial progress toward meeting our research and educational goals. This work involved a team of some eighty-five research Co-I's, collaborators and students located at seventeen institutions. It resulted in twenty-two publications in first-tier peer-reviewed journals and six additional publications still in press. The following highlights outline the major achievements of the ASU team during this review period.

• In the study of carbonaceous meteorites, Laurie Leshin determined that hydrogen isotope values for magmatic water entrapped in a martian meteorite indicate the potential for ~2-3 times more water in the crust today than previously thought, thus increasing the potential for life.

• In laboratory simulations of deep sea vent (black smoker) environments, the research team of John Holloway and Peggy O'Day showed that methanol, an important pre-biotic organic molecule, can be synthesized under natural geologic conditions occurring on the Earth's seafloor and should be a byproduct of the large amounts of carbon dioxide and hydrogen liberated by volcanic eruptions on the seafloor. They also used synchrotron X-ray tomography to determine the volume of micropores in a modern sulfide chimney from an active seafloor vent system. Such micropores may provide important sites for concentrating smaller organic molecules needed for the synthesis of more complex precursor molecules needed for the origin of life.

• Robert Blankenship's team reported a new molecular phylogenetic analysis of the genes for photosynthesis which was published in a featured cover article in *Science* magazine (Xiong et al. 2000).

• In July 2000 a group of nine scientists in Robert Blankenship's team made several dives on the submersible ALVIN to hydrothermal vents of the Endeavor field on the Juan de Fuca Ridge. Samples were collected and an analysis of pigments and cultures indicate that chlorophyll-like pigments are ubiquitous in the ocean and that small numbers of photosynthetic organisms are present both near vents and throughout the water column. The work has been accepted for publication in the journal Science.

• In a study of fossilization processes in thermal springs in Yellowstone National Park, Jack Farmer and collaborators determined that mat-sinter frameworks store large amounts of photosynthetic oxygen over a depth profile of several centimeters during peak day-time production. This discovery explains the commonly observed preservational modes in hotspring environments where surface encrustation of microbial cells and filaments is followed by rapid degradation of organic matter leaving behind external molds and other microtextural information.

• Thomas Sharp and John Moreau used methods of Analytical Transmission Electron Microscopy to show that the kerogencomprising organically preserved microfossils in the Gunflint Iron Formation are associated with nano-scale mineralogical and ultrastructural signatures that indicate that the organic materials enhanced the nucleation of amorphous silica during fossilization, inhibiting recrystallization and growth of crystallites during diagenesis. The ultrastructural features resulting from this inhibition reaction comprise a new type of biosignature.

• Paul Knauth and Donald Lowe found that oxygen isotope ratios in unaltered Archean cherts suggest climatic temperatures vastly warmer than those of today (surface temperatures of 50º - 80ºC).

• Paul Knauth and Donald Burt published a letter in Science which showed that eutectic brines, a logical consequence of current ideas about martian geologic and atmospheric history, can readily account for the recently observed seep features found at many high latitude locations on Mars.

• During 2000-2001, a large interdisciplinary team lead by James Elser began a study of extreme desert spring environments near Cuatro Cienegas, an ancient arid basin in central Mexico. By studying living algal-cyanobacterial mats and stromatolites and their grazers, they hope to gain clearer insights into the structure and energy flow of simple ecosystems comparable to those that emerged at the end of Proterozoic.

• Phil Christensen and team obtained new results from the Thermal Emission Spectrometer (TES instrument) presently mapping from Mars orbit. An important result was the discovery a region at Sinus Meridiani covered by hundreds of square kilometers grey, coarse-grained crystalline hematite. On Earth this mineral is only known to form in the presence of large amounts of water, indicating that this site was probably hydrologically active earlier in the planet's history. On this basis, Sinus Meridiani is now a leading candidate site for the 2003 Mars rover mission.

• Remote sensing analog studies in Death Valley led by Jeffrey Moersch and Jack Farmer showed that at TES spatial resolution of 3 km/pixel, carbonates and sulfates are difficult to detect in mixtures. However, at 100 m/pixel (the resolution to be obtained by the THEMIS instrument presently in route to Mars), both mineral groups were easily detected.

• Site selection studies for the Mars 2003 mission by Jack Farmer, Ron Greeley and David Nelson produced a list of high priority landing sites for astrobiology that were presented to mission planners. In addition, completion of 1:2M scale geological map of the Elysium Basin-Terra Cimmeria region of Mars (which includes several candidate sites for 2003) revealed a prolonged geological history of aqueous sedimentary activity.

• Ronald Greeley and collaborators synthesized a general review of the geology of Europa and made calculations to determine the amount of heat necessary to account for some of the dark features seen on the surface of this icy jovian moon. Standards were derived for the geological mapping of Europa and applied to one part of the surface. Studies were completed of two areas where sub-ice materials have been brought to the surface by impact processes.

- ASU carried out a diverse E&PO effort last year which included several campus-wide events. More than 125 science teachers and students attended a poster session held in conjunction with a public lecture given by NAI Director Barry Blumberg. The lecture event drew >2500 attendees. The teacher workshop included an NAI-hosted display and a review of recent curriculum materials in astrobiology.

Roadmap Objectives

#1
Sources of Organics on Earth

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

#9
Life's Precursors & Habitats in the Outer Solar System

#11
Origin of Habitable Planets

#12
Effects of Climate & Geology on Habitability

Project

Cosmochemistry of Carbonaceous Meteorites

Senior Project Investigator(s):
L. Leshin

Accomplishments

In this project we seek to understand the origin of pre-biotic organic molecules in carbonaceous chondrite meteorites and the origin and distribution of water (and other volatiles) on Mars.

In the past year we have had the unexpected pleasure of being able to study in detail a new, exciting meteorite fall, the Tagish Lake carbonaceous chondrite. We have examined the organic and inorganic inventory of this new, very primitive meteorite, leading to two published extended abstracts.

In the past year, we published a cover article in Geophysical Research Letters on the origin and evolution of water on Mars, based on an analysis of hydrous minerals in martian meteorite QUE 94201. In this work, we found the minerals to contain a mixture of water from two different sources, as delineated by their D/H ratios. First, the minerals contain primary "magmatic" water from the martian interior, with a D/H value ~twice terrestrial. This water was partially equilibrated with martian "meteoric" water, with a D/H value ~5 times terrestrial, as previously measured in the martian atmosphere. The big discovery here was the D/H value of magmatic water, which is much higher than previously assumed. This result has implications for the history of water on Mars, which probably involved a two-stage evolution, where early hydrodynamic escape was followed by Jeans escape of the atmosphere, a process that continues to the present day. In addition, the data imply that there may be ~2-3 times more water in the martian crust today than previously thought, which bodes well for martian biological potential.

Highlights

Hydrogen isotope values for magmatic water entrapped in a martian meteorite suggest there may be ~2-3 times more water in the crust today than previously thought, thus enhancing the potential for life.

NASA Mission Involvement

The analyses performed with this project help to focus analytical techniques for any NASA sample return missions. Such missions include Genesis and Stardust (now operational), plus an eventual Mars sample return mission.

For NASA's Mars Scout mission program, ten of the most promising mission concepts (for possible launch to Mars in 2007) have been selected to receive funding for six months of continued studies. Dr. Leshin is the Principal Investigator for SCIM (Sample Collection for Investigation of Mars), which is a mission concept in this select group of 10 for a possible Mars Scout mission.

Future Directions

- Publish papers on carbonates in CM (Carbonaceous Meteorite) chondrites and new work on Tagish Lake.

- Make further connections between inorganic and organic geochemistry in carbonaceous chondrites.

- Continue work on martian meteorites, focusing on the hydrous phases found in the new Los Angeles meteorite, and the history of martian volatiles including magmatic, crustal and surface inventories.

Roadmap Objectives

Project

Environmental Conditions of Early Earth

Senior Project Investigator(s):
L. P. Knauth

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

#12
Effects of Climate & Geology on Habitability

Accomplishments

This year our research focused on the climatic temperature of the early Earth at the time of the oldest microfossil record. Oxygen isotope data for the complete spectrum of cherts in the 3.5 Ga Swaziland Sequence of South Africa were examined in terms of the sedimentologic and structural context. We were able to show that the oxygen isotopic composition of many of these cherts was clearly set during the early burial history in the Archean and was not re-set later as so many have casually asserted. The data indicate that climatic temperatures were drastically warmer than those of today and must have been in the range of 55°-80°C. Early life on Earth was a high-temperature affair, consistent with the thermophilic nature of the earliest organisms in the universal rRNA tree of life. Early high temperature life was not restricted to vent areas.

In response to the announcement of apparently recent aqueous seeps on Mars, this past year we deviated from the original research goals to develop an argument that these seeps, if aqueous, must be brines with freezing points lower than –55°C. In reviewing current ideas about martian history, it is clear that if 1) Mars outgassed a hydrosphere, 2) lost most of its water via photodissociation/gravitation escape, and 3) subsequently froze, then eutectic brines should be present in the present martian subsurface.

In response to the heavy emphasis given to the Snowball Earth scenario at the 2000 NAI meeting, we also diverted from original plans to critically evaluate claims that the oxygen isotope composition of Neoproterozoic carbonates was re-set and should be ignored and also the extraordinary claims that carbon isotope excursions are global indicators of massive death of the biosphere. We analyzed new stratigraphic sections of the Beck Spring Dolomite in the Death Valley region of California and showed that oxygen and carbon isotope variations are correlative and that the excursions are actually local diagenetic events related to subaerial exposure of the carbonate.

Highlights

- New results indicate that oxygen isotope ratios in certain cherts in the Archean are indicative of climate temperatures vastly warmer than those of today (surface temperatures between 50º and 80ºC).

• The presence of eutectic brines are a logical prediction of current ideas about martian geologic/atmospheric history and can readily account for the recently observed seep features found at many high latitude locations on Mars.

• Carbon isotope excursions in Neoproterozoic carbonates can, in part, record local alteration events indicating a need for caution regarding recent extraordinary claims of global extinction during a putative Snowball Earth.

Field Trips

Death Valley, California, Fall 2000.

Field work was done during the fall of 2000 by Blair Lindford as part of an MS thesis work on Neoproterozoic carbonates in the Death Valley, CA, area.

Cross Team Collaborations

This project involves 2 areas of collaboration with the UCLA Team:

• Archean work was done in collaboration with Donald Lowe (Stanford University), who is a member of the UCLA team.

• Preliminary isotopic analyses and planning for field work in Australia were done in collaboration with Bruce Runnegar (UCLA team).

NASA Mission Involvement

Work on martian brines suggests that there may be many locations on Mars where hypersaline waters may exist in the shallow crust of Mars, thus creating potential near surface oases for life. This has important implications regarding site selection for future astrobiological missions to Mars.

In addition, our experiments with eutectic brines will refine our understanding of the potential of such environments to sustain life and/or preserve biosignatures of a subsurface martian biosphere. Finally, our work with brines is also helping to identify the kinds of measurements and instrumentation that will be needed to detect such environments on Mars during future missions.

Future Directions

• Complete and submit manuscript regarding eutectic brines on Mars and their astrobiological implications. This will be done with Prof. Donald Burt (ASU) who will be spending his sabbatical year at the Lunar and Planetary Institute in Houston.

• Complete and submit a manuscript on the climatic temperature of the Earth at 3.5 Ga

• Do fieldwork in NW Australia and extend the oxygen isotopic work recently completed for South Africa, to the Archean of Australia. This will be done in collaboration with UCLA partners Bruce Runnegar and Don Lowe. Petrography and isotopic work will be done in our laboratory here at ASU.

- Attempt to synthesize martian eutectic brines and measure their electrical conductivity and D/H ratio. If the brines have the high conductivity expected, they should be vastly easier to detect with remote geophysical methods than is pure water. This approach could provide a test of our brine hypothesis and has the potential to impact future Mars missions.

Roadmap Objectives

#3
Models for Life

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#14
Ecosystem Response to Rapid Environmental Change

Project

Evolution in Microbe-Based Ecosystems: Desert Springs as Analogues for the Early Development and Stabilization of Ecological Systems

Senior Project Investigator(s):
J. Elser

Accomplishments

Last year we made progress by initiating studies at our field study sites in Cuatro Cienegas, central Mexico. We worked with our UNAM collaborators to obtain full scientific permits for project work in Mexico. During a 10-day expedition in December 2000, we completed field site reconnaissance and sampling and continued ongoing genetic studies of fish populations across environmental gradients started the previous year with seed funding from ASU. This was followed-up with a longer expedition in March 2001 in which several additional project components were initiated. Sampling sites for a chosen headwater-terminal basin flow path (Churince) were established. Two 2x2 nutrient enrichment (nitrogen, phosphorus fertilization) experiments were performed at Laguna Intermedia (Churince system) to compare a diatom-dominated mat system to one dominated by cyanobacteria. The experiment involved the laboratory incubation of isolated mat samples under natural illumination.

Fifty-five localities within five separate drainage systems were sampled for analysis of diversity of archaea and bacteria using molecular techniques. Many of these have been successfully isolated using selective media and brought into culture and frozen for detailed genetic analysis. Population genetic studies have been initiated for widespread taxa across important environmental gradients. For studies of stromatolitic phototrophs at Cuatro Cienegas, we have carried out extensive sampling in the field area and successfully developed methodologies for efficient nucleic acid extraction from encrusted/calcified microbial communities. Characterization of phototrophic components of stromatolite building microbial communities using molecular techniques is underway, and morphogenesis studies have been initiated. We thin-sectioned fixed oncolite samples and started basic paleontological, mineralogical and microfabric studies of those and previously collected samples.

Population genetic studies of pupfish (Cyrpinodon) across environmental extremes were continued. Cyprinodon specimens were collected from throughout the basin, providing necessary perspective for examination of horizontal gene transfer between *C. atrorus* and *C. bifasciatus*. Temporal sampling of physicochemical data and Cyprinodon within areas of hybridization was initiated. We developed morphological, systematic, and molecular protocols to analyze the evolution and ecology of hydrobiid snails from Cuatro Cienegas. All physicochemical measurements and Cyprinodon samples are currently being prepared for analysis. A database has been set up to integrate data on landmark morphometric analyses, relationships to environmental factors (i.e., temperature, pH, salinity), and existing molecular DNA analyses. Our project also involves modeling of food-web evolution. We have completed formulating the architecture of our model for ecological communities evolving under stoichiometric constraints, which we will use to compare to empirical observations of the living systems under study. We have nearly completed the mathematical derivation of mass conservation equations and are also nearing completion of encoding the model into C++.

Highlights

During 2000-2001, our diverse team of scientists, including limnologists, microbial ecologists, evolutionary biologists, biosedimentologists and paleontologists began an intensive study of unique system of desert springs near Cuatro Cienegas, an ancient arid basin in central Mexico. By studying living algal-cyanobacterial mats and stromatolites and their grazers (primarily Cyprinodon fish species and hydrobiid snails) we expect to gain clearer insights into the structure and function of simple microbial mat-grazer ecosystems similar to those that arose during the major biosphere transition that occurred at the end of Proterozoic just after the first macroscopic grazing animals appeared. It is hoped that the highly integrated bio-geological studies that are currently underway will lead to a better understanding of the ecological and environmental forces that shaped and eventually stabilized Earth's earliest complex food webs.

Field Trips

Chihuahuan Desert, Central Mexico.

Field work focuses on desert hot springs in the Chihuahuan Desert of central Mexico near the town of Cuatro Cienegas, Mexico. Information about the field area can be found at http://www.utexas.edu/depts/tnhc/.www/fish/dfc/cuatroc/.

Cuatro Cienegas, December 2000 and March 2001.

Field expeditions to Cuatro Cienegas occurred during December 2000 (2 weeks) and March 2001 (3.5 weeks). Participating were: Elser (ASU), Minckley (ASU), Dowling (ASU), Garcia-Pichel (ASU), Souza (UNAM), Eguiarte (UNAM), Kyle

(ASU), Wade (ASU), Carson (ASU), Meir (UW). The December expedition involved reconnaissance for several PIs and continued genetic sampling of fish populations (Dowling, Carson). During March, monitoring of our main study sites was initiated, a 2-week nutrient enrichment experiment was performed (Elser, Kyle), sampling of Archaea and Eubacteria in diverse habitats was accomplished (Souza, Eguiarte), sampling of cyanobacteria mats and stromatolites for molecular studies completed (Garcia-Pichel, Wade), and further genetic sampling of fish populations was performed (Carson).

Cross Team Collaborations

F. Garcia-Pichel, J. Farmer, J. Elser, and B. Wade (all with the ASU Team) are collaborating in a study of the ecology and biomineralization of floating cyanobacterial colonies at Escobedo in the Cuatro Cienegas basin.

Additionally, Farmer, Garcia-Pichel, McDowell, and Wade (all with the ASU Team) have been working together in preliminary studies on the morphogenesis of algal-cyanobacterial oncoids in the basin.

In a third interactive group (within the ASU Team), Elser, Garcia-Pichel, and Wade are collaborating in experimental studies involving unmineralized algal mats in Laguna Intermedia in the Churince drainage of Cuatro Cienegas.

C. Tang (ASU Team) has been interacting with Monica Medina, who was an NAI postdoc in Mitch Sogin's lab (MIT) and is currently employed at the Joint Genomics Institute in California. The two will be collaborating on molecular analyses of snail populations from Cuatro Cienegas.

Future Directions

- Continue sampling of Churince system for environmental variables and algal C:N:P stoichiometry.

- Perform P-fertilization experiment to examine effects of higher algal P-content on growth of grazing snails.

- Continue sampling and analysis of physicochemical data and Cyprinodon to determine whether patterns of gene transfer are associated with environmental parameters. We will also prepare for publication tests of population genetic structure for *Cyprinodon atrorus* and *C. bifasciatus*.

- Isolate the DNA for at least 30 archaeal strains per site, analyze the 16S rRNA, the intergenic spacer of the RNA genes, and a neutral gene to assess genetic diversity, differentiation between sites, and linkage disequilibrium within and between sites.

- Identify the phylogenetic and taxonomic groups of the Eubacteria using biochemical and genetic tools (16S rRNA). We have more than 4000 strains that need to be classified from 50 sites. Perform community analysis on these data. Obtain the population genetic parameters of the 2-5 key species that are widespread in different sites.

- Finish molecular fingerprinting of phototrophic communities. Initiate characterization of rest of bacterial communities including sulfate reducers, sulfide oxidizers.

- Document the physical relationships between microbial biofilms and primary mineral precipitates using SEM (scanning electron microscopy) on fixed and critical point dried samples, along with correlative light microscope observations of petrographic thin sections of fixed and stained microbial biofilm/mineral interfaces.

- Document the range of primary microfabrics for a range of stromatolite morphotypes (including oncoids) and determine their primary mineralogy. Assess how primary microfabrics and mineralogies are altered during early diagenesis using materials collected from older parts of the basin. Microsample stromatolitic carbonates for isotopic studies. Assess the nature of competitive interactions between stromatolite-forming microbial mats and their grazers and competing macroalgae to understand the effects on stromatolite morphology and microfabric.

- Complete morphometric analysis on previously collected hydrobiid snails, conduct field work to collect new snail specimens from selected localities, conduct pilot molecular work on hydrobiid snails to assess placement within molluscan phylogeny, conduct initial molecular analyses to assess genetic variability and differentiation among species and populations, and develop sampling protocol for stable isotopic analyses of shell material.

- Complete the mathematical derivation and encoding of the food-web evolution model, and begin exploring key questions associated with the evolution of ecological complexity under biogeochemical constraints.

Exploring for Habitable Environments on Europa

Project

Senior Project Investigator(s):
R. Greeley

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#9
Life's Precursors & Habitats in the Outer Solar System

#12
Effects of Climate & Geology on Habitability

Accomplishments

We have made substantial progress in deciphering the origin and evolution of the surface of Europa, and in characterizing the potential niches appropriate for astrobiology: 1) A general review of Europa was synthesized for a meeting of terrestrial sea ice experts, some of whom subsequently became involved with the Europa Focus Group, 2) calculations were made to determine the amount of heat necessary to account for some of the dark features seen on the surface of Europa (assuming that they result from ice sublimation and the formation of a lag deposit), some of these areas might be favorable for astrobiology, 3) standards were derived for the geological mapping of Europa and applied to one part of the surface, and 4) studies were completed of two areas where sub-ice materials have been brought to the surface by impact processes.

Highlights

A general review of Europa was synthesized and calculations made to determine the amount of heat necessary to account for some of the dark features seen on the surface of Europa. Standards were derived for the geological mapping of Europa and applied to one part of the surface, and studies were completed of two areas where sub-ice materials have been brought to the surface by impact processes.

Cross Team Collaborations

Collaborative research for this project has been conducted with Robert Pappalardo (Brown University), currently a member of the University of Colorado (Boulder) and relocating there.

We plan to continue the collaboration, including preparation of a general review paper on Europa for the Cambridge University Press Special Volume series. This publication will relate to the Jupiter Conference held in July 2001 at the University of Colorado, Boulder.

NASA Mission Involvement

This project related to NASA's future Europa Orbiter, tentatively scheduled for launch in 2008 and operation in 2010. Results from our study are directly relevant to this NASA mission. Our studies relate to targeting high priority areas on Europa for critical remote sensing data from the orbiter, as well as for future landed operations.

Future Directions

For the next year, we plan a synthesis of the global geology of Europa, outlining the key terrains and surface units and deriving a general geological history of the surface. Results would then be used to target sites favorable for exobiology for future exploration.

Exploring Mars for Past or Present Life

Project

Senior Project Investigator(s):
P. Christensen, J. Farmer, R. Greeley

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

#11
Origin of Habitable Planets

#12
Effects of Climate & Geology on Habitability

Accomplishments

This research project seeks to promote the astrobiological exploration of Mars through planetary exploration and terrestrial analog studies. Over the past year, efforts have been focused in three major areas: 1) Continued orbital mapping of the martian surface in the mid-infrared in search of aqueous mineral deposits formed earlier in martian history when water was present at the surface (such deposits are important targets for surface exploration to search for a record of prebiotic chemistry and /or fossil biosignatures of martian life). 2) Remote sensing analog studies of evaporite basins on the Earth to establish spatial and spectral resolution thresholds for the detection of aqueous minerals from Mars orbit. 3) Site selection for future landed missions to explore for past or present martian life. Progress in each of these areas is discussed below.

1) Exploration for aqueous mineral signatures was continued the past year using data from the Thermal Emission Spectrometer (TES), presently mapping in Mars orbit. A substantial effort was made to define the distribution of coarse-grained (specular) hematite detected last year. This mineral is the most compelling aqueous mineral signature so far discovered at Mars. Specular hematite has now been detected at three sites: Sinus Meridiani, Aram Chaos and Candor Chasma. These results have established the potential importance of specular hematite as a pathfinder mineral for ancient aqueous environments on Mars. The largest hematite deposit discovered so far is located at Sinus Meridiani. This place has emerged as a leading candidate landing site for landed missions to be launched in 2003. Another important result concerns the origin of putative paleolake deposits at a site known as White Rock. Previous authors have suggested this deposit to be a lacustrine evaporite based on geological setting and albedo. Evaporites hold obvious interest for martian astrobiology. Analysis of the TES data suggested an absence of aqueous minerals, but is more consistent with an aeolian origin, thus calling into question the hypothesis of an aqueous origin for White Rock.

2) We have completed field work for remote sensing analog studies in the Badwater Basin of Death Valley. The field component involved the collection of ground spectra using portable spectrometers. We also made significant progress in processing several infrared remote sensing data sets (AVIRIS, MASTER and ASTER) for the study area. This was accomplished using ENVI, a remote sensing

analytical software. Analysis involved developing sound atmospheric corrections and binning measurements from the high resolution data to achieve spatial resolutions comparable to TES and THEMIS. We then ran classification algorithms, including multivariate statistical analyses that allowed us to produce mineral maps of the end member compositions at spatial resolutions for TES and THEMIS. These end-member maps were then compared to spectral libraries to predict the distribution of key aqueous mineral groups, such as carbonates, sulfates and borates. The main conclusion was that while TES spatial resolution of 3 km/pixel appears too coarse to reliably detect these minerals, they are clearly detected at the 100 m/pixel spatial resolution of THEMIS (presently in route to Mars). Analog comparisons are still incomplete in that A) we have yet to model the effects of the lower spectral resolution of THEMIS on detectability and B) we still need to complete ground truth studies to establish what minerals are actually present and in what abundances. This last year we completed the field work for the ground truth component of the study, and most of the lab work. Lab work involved X-ray diffraction analysis of field samples and lab spectral analysis using a TES analog instrument. We are close to completing thin section petrographic analyses that will establish actual mineral abundances through statistical point counting methods.

3) Site selection for upcoming Mars missions is an important aspect of our research, and the past year we have focused this effort in two areas: A) Site identification and characterization for the 2003 mission to explore for ancient aqueous sediments and B) Exploration for high latitude polar sites where recent volcanism and associated hydrothermal activity may have occurred. We have now completed a 1 : 2M scale photomosaic and geologic map of the Elysium Basin – Terra Cimmeria region of Mars, an area which shows evidence of a prolonged aqueous sedimentary history. The goal is to describe the detailed hydrological history of the area and compare it to other similar-aged terrains previously mapped by others. Included within the map area is Gusev crater, an important astrobiological site we advocated at the 2003 landing site workshop in January and which has now been short-listed as a potential landing site for the 2003 mission. The second aspect of the site selection work involves the exploration for sites of volcano-ice interaction along the margins of the North Polar Cap of Mars. This past year we completed reconnaissance of the North Polar region, examining all available Viking and Mars Orbiter Camera (MOC) images. For this work we relied heavily on use of the Space Photography Lab an imaging archive located on the ASU campus. This work resulted in the identification of three major sites, one a small volcanic field, with associated outflow channels, another site we interpret to be a pseudocrater field where explosive volcanism occurred as volcanic flows were erupted onto ground ice, and an alluvial plain in the Chasma Boreale region thought to have formed by subglacial outfloods. Current work is focused on obtaining topographic (MOLA) data for each of the sites above and producing integrated data sets as a basis for detailed mapping. The sites we have discovered are suspect hydrothermal environments where subsurface ecosystems may have been sustained for a prolonged period. Thus, they are considered good places for future landed missions to explore for biosignatures sequestered in ground ice.

Highlights

• Thermal Emission Spectrometer (TES) results produced a leading candidate site for the 2003 Mars rover mission: Gray, crystalline hematite covering hundreds of square kilometers in the Sinus Meridiani region of Mars has been identified using TES data from the Mars Global Surveyor mission. Because of the link between coarse grained hematite and water, Sinus Meridiani has emerged as a strong candidate landing site for one of the two Mars Exploration Rovers to be launched in 2003.

Field Trips

Death Valley and the Mono Basin.

Field studies form an integral part of the Mars analog studies in this research module, and this past year were focused on work in Death Valley and the Mono Basin of eastern California.

Cross Team Collaborations

Collaborative interactions with other NAI members have been mainly through our participation in the NAI Mars Focus Group activities described elsewhere in this report.

NASA Mission Involvement

The Co-Investigators with this project are all active in Mars mission planning, implementation, and data analysis.

Co-I Christensen is the PI for the TES (Thermal Emission Spectrometer) instrument on MGS (Mars Global Surveyor), as well as the THEMIS (Thermal High Emission Infrared Spectrometer) instrument recently launched on the Mars 2001 Odyssey mission. He is also PI for the mini-TES, a mid-IR spectrometer that will be launched in 2003 as part of the Mars Exploration Rover missions.

Co-I Farmer is an active member of the Site Selection Steering Committee for the 2003 Mars rover mission, and he was a member of the 2005 Science Definition team. Farmer also heads the NAI Mars Focus Group, which provided recommendations on landing site selection for astrobiology this past year.

Co-I Greeley heads the Mars Exploration Payload Advisory Group, which has been developing a long-range exploration plan for Mars.

For NASA's Mars Scout mission program, ten of the most promising mission concepts (for possible launch to Mars in 2007) have been selected to receive funding for six months of continued studies. In this select group of ten is the CryoScout mission concept, with Frank Carsey (Jet Propulsion Laboratory) as PI and Jack Farmer as Co-I.

Future Directions

- A high priority will be placed on submitting manuscripts for publication based on the remote sensing analog studies in Death Valley and site selection studies in the Elysium Basin and North polar region of Mars.

- We plan to expand work in Death Valley to other playa lake basins (e.g. Fish Lake playa and Deep Springs Valley) to cover a broader range of evaporite minerals (Moersch, Farmer, Baldridge). This will include the development of spectral libraries for a wide variety of naturally occurring and synthetic evaporite minerals.

- We will begin field work in Iceland to map and sample several types of features formed by volcano-ice interactions (Farmer and Payne). The focus of this study will initially be to characterize the paleoenvironments of ancient subglacial features and identify volcanic and sedimentary facies formed under conditions habitable for life. These facies will be explored for biosignatures (alteration textures) in volcanic glasses similar to those that have been recently described for deep sea basalts.

- We will also undertake refined mineral mapping of the martian surface (Christensen, Ruff and Hamilton). TES data have been used to generate initial maps of the distribution of global geologic units. Improved surface-atmosphere–separation results and more localized analyses will lead to improvements in our knowledge of the mineralogy of the Martian surface. Following orbital insertion this Fall, the THEMIS (Thermal Emission Imaging System) instrument will also begin mapping at a much higher spatial resolution. This should significantly improve our ability to detect surface deposits of aqueous minerals and provide a basis for more refined site selection for astrobiology.

- We will expand our search for aqueous sediments to include other putative paleolake basins on Mars (Moersch, Ruff, Baldridge, Christensen, Farmer). Roughly 150 paleolake basins have been identified on Mars based on geomorphic evidence. TES and THEMIS data will be well suited to identify aqueous minerals in these settings, this providing additional data for testing paleoenvironmental hypotheses derived from geomorphic observations.

- Ongoing contributions to landing site selection for astrobiology will include efforts to assess the geomorphology, mineralogy and surface characteristics of candidate landing sites using MOC, TES and starting this Fall, THEMIS data. Convolved data sets will be produced for several of the highest priority sites tentatively targeted for landed missions in 2007 and beyond. Among these sites are putative paleolake basins at Sinus Meridiani, Eos Chasma, Gusev and Gale Craters, and possible hydrothermal sites at Apollonaris Patera, Candor Chasma, Elysium, and Aram Chaos.

Microbial Fossilization Processes in Extreme Environments

Project

Senior Project Investigator(s):
J. Farmer

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

Accomplishments

The goal of this project is to understand the taphonomic processes that govern the fossilization and long-term preservation of biosignatures in extreme sedimentary environments dominated by microbial life. Progress was made on several fronts in the past year. Results of work reported last year to determine the taphonomic controls on preservation in Yellowstone hotsprings is now in a draft manuscript, and completion is anticipated by the end of the summer.

The past year we began new studies in conjunction with the EcoGenomics Focus Group (EGFG).

1) We completed previous work on determining the origin of laminated fabrics on submerged cyanobacterial mats in saltern environments at Guerrero Negro, Baja Sur, a mat system that accretes at a rate of about 1 cm/yr.

2) As a part of the Guerrero Negro study, we have also begun to assess the role of meiofaunal grazers in the overall mat economy. To advance this project, samples were collected in conjunction with a group diel experiment conducted by the EGFG. The goal was to determine whether there is vertical migration of the meiofauna during diel excursions of oxygen-sulfide in the surface mat environment. Samples were fixed in the field and returned to the lab where they were dehydrated and embedded in low viscosity resin. Using standard methods, 20-micrometer-thick petrographic sections (of both stained and unstained materials) were prepared for light microscopy. In addition, fixed samples were critical-point dried for observation by FESEM. Detailed light microscopy and FESEM are presently underway.

3) In addition to studies of submerged mat systems found in salterns at Guerrero Negro, we also began a new study of microbial mats in natural lagoon supratidal and tidal channel environments. During field work in June, we deployed permanent temperature data recorders at several sites to monitor the frequency and duration of tidal submergence for these mats over the next 6 months. We also collected mat-sediment samples to determine whether mats show grain size selectivity during the trapping and binding of detrital sediments brought in during tidal cycles. (This has implications for the recognition of mat-mediated detrital sediments in the ancient record). Upon returning to the lab, mat samples were oxidized to remove all organic matter, and the size frequency distribution of mat-associated detrital grains was determined by mechanical sieving. (Data are presently being

analyzed by statistical methods to quantify grain size differences between different mat types). In addition, mat-sediment samples were prepared for light microscopy using the procedures described under 2) above.

Highlights

Studies of rapidly mineralizing thermal springs have shown that microbial biosignatures preserved in those environments consist of primarily microstructural information (cellular and filament molds), with little or no preserved organic matter. This is rather unexpected given the rapid rate of entombment of organisms commonly observed in mineralizing springs, a process that intuitively should rapidly arrest degradation favoring the preservation of organic matter sequestered within precipitated mineral phases. Oxygen microsensor studies of hot-spring microbial mats have provided an explanation. The photosynthetic bacteria that dominate these ecosystems produce large quantities of oxygen which becomes entrapped and concentrated within bubbles in the slime matrix of mats and porous sinter framework. Oxygen levels exceeding 900 times the atmospheric concentration have been measured during periods of peak photosynthesis, indicating a super-oxygenated environment down to depths of several cms within mat-sinter frameworks. Stromatolites (layered sedimentary structures formed by mineralizing microbial mat systems) comprise the primary record of benthic life on Earth during the Precambrian. These important biolaminated structures have been traditionally attributed to the activities of photosynthetic cyanobacteria and typically lack an associated cellular microfossil record. Although microsensor approaches are just now being applied to the study of mineralizing mat systems, the presence of high concentrations of entrapped oxygen in the stromatolitic sediments of thermal springs suggests a potential explanation for the paucity of organic microfossils commonly observed in ancient stromatolites.

Field Trips

Guerrero Negro, Baja Sur, May and June, 2001.

The projects associated with this research are field intensive. Over the last year this involved visits to hypersaline marine environments in Guerrero Negro, Baja Sur (Farmer, Garcia-Pichel, Des Marais, Bebout, Visscher, Director Blumberg and other members of the EcoGenomics Focus Group 5/00 and 6/01). Goals of these expeditions were to conduct *in situ* measurements of natural microbial mat systems to better understand their ecology, biogeochemistry, sedimentology and paleontology.

Cross Team Collaborations

Research collaborations for this project have involved various people and teams

1) David Des Marais, Brad Bebout, and other members of the microbial mat team with the ARC Team
2) Pieter Visscher (University of Connecticut with the ASU Team)
3) Ferran Garcia-Pichel and Paul Knauth (both with the ASU Team)

NASA Mission Involvement

Understanding requirements for the preservation of fossil biosignatures is programmatically important for NASA. It helps to establish criteria for the recognition of legitimate microfossils in ancient Martian sediments, an important objective driving Mars sample return.

The recognition of factors important in controlling microbial fossilization has important applications in the development of exploration strategies for a martian fossil record. This background has been a driver in the development of payload instrumentation concepts for Mars landing missions.

Future Directions

- Complete and submit a manuscript describing the taphonomic controls on microbial fossilization in hydrothermal springs; with Brad Bebout and Pieter Visscher.

- Complete studies of laminated mat fabrics of submerged cyanobacterial mats at Guerrero Negro, Baja Sur and prepare manuscript for publication.

- Isolate and describe the dominant species of meiofaunal metazoans associated with laminated cyanobacterial mats at Guerrero Negro; begin experiments to determine the respiration rates of major meiofaunal species under different regimes of temperature, pH and sulfide; examine diagenetic trends in the preservation of mat-derived organic matter over the decadal accretionary record provided by laminated subtidal mats.

- With Ferran Garcia-Pichel (ASU), complete initial field studies of supratidal and tidal channel mat systems at Guerrero Negro to assess their role in the sedimentology of these environments and, in collaboration with Paul Knauth, apply this information to an interpretation of similar Proterozoic intertidal sediments in North America and elsewhere.

- In collaboration with Paul Knauth, begin a systematic study of problematic microfossil assemblages associated with Proterozoic karst environments of the Apache Group, central Arizona. The goals are to establish morphometric and geochemical criteria for judging the biogenicity of suspect microfossil assemblages in these rocks, based on comparisons with modern analogs.

Roadmap Objectives

#1
Sources of Organics on Earth

#7
Extremes of Life

#9
Life's Precursors & Habitats in the Outer Solar System

Project

Organic Synthesis at High Temperatures

Senior Project Investigator(s):
J. Holloway and P. O'Day

Accomplishments

Our overall goal in this research is to explore the potential for abiotic synthesis of organic molecules important to the origin of life under geologically realistic conditions of pressure, temperature and chemical composition. To this end we have detailed how volcanic diking-eruptive events on the seafloor can lead to short-term high levels of carbon dioxide and hydrogen gases. We then determined experimentally that those gases and water react at typical seafloor hydrothermal conditions to form methanol, a significant organic precursor molecule, in the presence of the mineral magnetite. Building on kinetic studies of the conversion of smectite clay to illite, we have begun experiments to follow the conversion of methanol to more complex organic molecules in the presence of smectite/illite clays.

We constructed and tested a new high-pressure reactor to safely study organic synthesis reactions in sulfide systems. The operating parameters for this system were determined using thermodynamic calculations as a guide.

The internal structure of sulfide chimneys in seafloor systems may provide sites for concentrating prebiotic organic molecules. The recent collection of a large, living chimney has provided excellent samples to examine microporosity using synchrotron X-ray tomography, and preliminary results have been reported showing porosity down to the limit of resolution of the technique, about ten micrometers.

We have established a collaboration to develop analytical techniques based on optical fiber probes to allow chemical analysis of small volumes in experimental apparatus for organic synthesis and in natural environments at hydrothermal conditions.

These accomplishments were included in our proposed goals for the third year. However, we have not made as much progress on the artificial seafloor hydrothermal system experiments as planned due to the need to redesign and construct portions of the system. We also expended a significant effort in improving our techniques for analyzing organics in aqueous solutions.

Highlights

- Demonstrated that volcanic eruptions on the seafloor, the most common form of volcanism on Earth, can generate large amounts of carbon dioxide and hydrogen

- In press peer-reviewed paper demonstrates that the important organic molecule, methanol, can be synthesized under natural geologic conditions occurring on the Earth's seafloor

- Constructed and tested a new high-pressure reactor to safely study organic synthesis reactions in experimental systems precipitating the sulfide minerals found in seafloor hydrothermal vents

- Used synchrotron X-ray tomography to measure the volume of micropores occurring in a young sulfide chimney from a seafloor hydrothermal system. Such micropores may have been important sites for concentrating smaller organic molecules to allow synthesis of more complex biomolecules.

Cross Team Collaborations

We have interacted with John Delaney and John Baross (both with the University of Washington Team) on geological and microbiological issues related to active seafloor black smokers.

NASA Mission Involvement

Results of our research will improve our ability to assess the potential for organic synthesis on other bodies in our Solar System (e.g., Mars and Europa) where hydrothermal processes are likely to have been active. The experimental approach we have developed permits the simulation of conditions that may exist elsewhere in the Solar System, with attending technology spin-offs to enhance the development of NASA flight experiments and instruments for future missions.

Future Directions

- Investigate the role of sulfide in abiotic synthesis of organic molecules under seafloor hydrothermal conditions, including synthesis in the presence of precipitating sulfide minerals.

- Earth (and probably other planets) has aqueous-carbonic systems consisting of liquid only, gas and liquid, and gas only. We will explore differences between abiotic synthesis reactions in each of these systems to further explore the nature and limits of synthesis.

- Clay minerals have long been thought important in the origin of life. We plan a series of experiments to determine the efficacy of clay minerals in assembling complex organic molecules from simple ones. We will begin by using methanol, which we have shown can be synthesized under natural geologic conditions.

- We will extend the mapping of porosity in a chimney from a modern seafloor hydrothermal system vent using X-ray tomographic analysis.

Roadmap Objectives

#3
Models for Life

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

Project

Origin and Early Evolution of Photosynthesis

Senior Project Investigator(s):
R. Blankenship

Accomplishments

Work on the origin and early evolution of photosynthesis is carried out by a highly interdisciplinary team of scientists from several different institutions. The overall theme is understanding the origin and early evolution of photosynthesis and its effect on the biosphere. This year, four main aspects have been emphasized: (1) molecular evolution analysis of photosynthesis genes from a wide range of organisms, (2) whole genome comparative analyses, (3) field and laboratory studies of photosynthetic organisms in iron-rich environments and (4) the search for phototrophs in non-solar environments around hydrothermal vents (living things that derive their photo-energy from non-solar sources). Each of these projects has seen significant progress, as described below.

Molecular evolution analysis of photosynthesis genes. We have carried out and published sequencing and molecular evolution analysis of photosynthesis genes from all known major groups of photosynthetic organisms.

Whole genome comparative analyses. We developed methods for doing whole genome comparisons. The results, as yet unpublished, strongly support the mosaic nature of bacterial genomes, in which different genes in an organism have distinct evolutionary histories.

Field and laboratory studies of photosynthetic organisms in iron-rich environments. Our field and laboratory studies indicate that some photosynthetic organisms can utilize reduced iron as an electron donor, which may have importance for understanding the origin of the banded iron formations and the rise of oxygen on Earth.

The search for phototrophs in non-solar environments around hydrothermal vents. In July 2000 a team of nine scientists made several dives in the ALVIN submersible vehicle to hydrothermal vents of the Endeavor field on the Juan de Fuca Ridge. Samples were collected and are in the process of being analyzed. Analysis of pigments and cultures indicate that chlorophyll-like pigments are ubiquitous in the ocean and that small numbers of photosynthetic organisms are present both near vents and throughout the water column.

Highlights

- Molecular evolution analysis of photosynthesis genes was published in a cover-featured article in Science (Xiong et al., 2000; Science 289:1724-1730).

• A team of scientists made dives to hydrothermal vents and found evidence for chlorophyll-like pigments and viable photosynthetic organisms in the deep ocean. A report on some of this work has been accepted for publication in Science.

Field Trips

Endeavour Field on the Juan de Fuca Ridge, July 16-28, 2000.

Purpose: search for phototrophs in a non-solar environment in the deep sea. Nine investigators embarked on a research expedition including ALVIN dives to hydrothermal vents at the Endeavour Field on the Juan de Fuca Ridge (16-28 July 2000) to search for phototrophic organisms. Participants: Cindy Lee Van Dover, Chief Scientist, College of William & Mary; Gerry Plumley, University of Alaska Fairbanks;Robert Blankenship , Arizona State University; Paul Falkowski, Rutgers University; Zbigniew Kolber, Rutgers University; Andrew Lang, U. British Columbia; Chris Rathgeber, U. Manitoba; Michael Lince, Arizona State University; Sherri White, Woods Hole Oceanographic Institution; Bill Smith, Medeco, Inc; Cheryl Jenkins, College of William & Mary.

Chocolate Pots hot springs, Yellowstone National Park, Summer 2000.

Purpose: Physiology of phototrophs in iron rich hot spring environments. Field work was conducted at Chocolate Pots hot springs, Yellowstone National Park (Summer 2000). The following team members attended: Beverly Pierson, Team Leader, Univ. of Puget Sound; Victor Scapa, Undergraduate Research Student, Univ. of Puget Sound; Coleen Pidgeon, Technical Assistant, Univ. of Puget Sound; Niki Parenteau, Technical Assistant; Univ. of Puget Sound.

Cross Team Collaborations

Lynn Rothchild (NASA Ames Team) and Robert Blankenship (ASU Team) are collaborating on the study of reactive oxygen enzymes in thermophilic phototrophs.

This collaboration is just beginning to produce results, and it will be continued in the coming year. The work involves both laboratory and field research. In conjunction with this project, one of Blankenship's Ph.D. students will be interning with Rothchild at Ames during the summer of 2001.

Future Directions

• A second series of ALVIN dives to hydrothermal vents is anticipated at a location to be determined. Emphasis will be on determining the environmental distribution of genes for photosynthesis using PCR (polymerase chain reaction) analysis.

• Whole genome comparisons will be expanded to include recently determined genomes of additional photosynthetic organisms.

• Molecular evolution analyses will be carried out on heme biosynthesis genes and additional chlorophyll biosynthesis genes using advanced techniques of phylogenetic analyses.

- Artificial cell experiments will be pursued by using light-driven ATP (adenosine triphosphate) synthesizing vesicles that incorporate a synthetic light-driven proton pump and an ATP synthase enzyme.

- Field work on the iron oxidizing bacteria, as well as the reactive oxygen enzymes of phototrophs in thermal environments, will be carried out.

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#12
Effects of Climate & Geology on Habitability

#14
Ecosystem Response to Rapid Environmental Change

Project

Roles of Impacts in the Origin, Distribution & Evolution of Life

Senior Project Investigator(s):
D. Kring

Accomplishments

We finished testing the new computer system designed to provide 3-D simulations of impact cratering events. We then simulated the Ries impact event that occurred 15 million years ago in what is today Germany. This impact is interesting because it involved carbonate rocks, which, when vaporized, can produce greenhouse-warming carbon dioxide. We are continuing these types of model calculations to determine how impact events can affect Earth's climate.

We began studies of impact-generated hydrothermal systems. Initial results were presented at several meetings and a paper is presently in production. We are evaluating the thermal history of these systems and their appropriateness for thermophilic and hyperthermophilic life, particularly early in life's evolution when impact events were more common.

In a collaborative project, we demonstrated that the lunar cataclysm hypothesis was correct. This implies that the Earth was severely bombarded approximately 3.9 Ga ago, coincident with evidence of the earliest evolution of life on Earth.

Highlights

Model-based studies of early impact processes have provided a positive test of the lunar cataclysm hypothesis and imply that the Earth was severely bombarded approximately 3.9 Ga ago, immediately preceding the earliest fossil evidence for life on Earth.

Field Trips

Queen Charlotte Island , Canada.

Recent fieldwork was carried out in a collaboration with Peter Ward (Univ. of Washington) to study the role of impacts in the terminal Cretaceous extinction on Queen Charlotte Island, Canada.

Cross Team Collaborations

We have just begun a collaborative study concerning the role of impact processes in planetary habitability and extinction with Peter Ward (University of Washington Team).

NASA Mission Involvement

The role of impacts in regulating planetary habitability has important applications in assessing the potential for life on Mars. Modeling of impact generated hydrothermal systems on Mars has important implications concerning site selection in the exploration for past martian life.

Future Directions

Simulation of impact cratering events using a 3D hydrocode to assess their effects on the climate and other components of the environment, both on Earth and Mars

Computer models will be used to calculate the thermal evolution of impact-generated hydrothermal systems.

We will analyze samples of hydrothermally-altered rocks collected from existing impact craters to further constrain our models of impact-generated hydrothermal systems.

In a related project involving the environmental consequences of impact cratering, we will continue to investigate the effects of impact-generated wildfires.

Roadmap Objectives

Project

Submicroscopic Study of Microbial Fossils in Chert

Senior Project Investigator(s):
T. Sharp

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

Accomplishments

The purpose of this study of microbial fossils is to improve our understanding of microbial biosignatures by developing criteria for biogenicity at the nanometer scale using new methods of electron microscopy. To achieve this goal we have been studying the mineralogy and ultrastructure of well known microfossils from the 2.0 Ga old Gunflint Iron Formation. Gunflint microfossils were initially investigated by standard methods of petrography and electron microscopy to determine whether submicroscopic features of organically preserved microfossils persisted. Our nanoscale studies have shown that microfossils are comprised of aggregated spheroidal grains (200-500 nm) of æ-quartz and brown interstitial kerogen. Analytical transmission electron microscopy (TEM) revealed that quartz spheroids are made up of smaller domains of æ-quartz separated by poorly diffracting silica phases. In contrast, the matrix silica is composed of granulitic, coarse-grained (5-10 micrometer) æ-quartz. The kerogen not only provides the optically visible biosignature in these samples, but it exerted a clear influence on the nucleation, growth and recrystallization of the silica, resulting in ultrastructural biosignatures not previously recognized.

Hydrofluoric acid (HF) vapor etching, commonly used to prepare microfossils for scanning electron microscopy (SEM) work, was used to preferentially dissolve the matrix silica of samples, leaving microfossils in positive relief. These etched surfaces were then studied using FESEM. The results of FESEM investigations suggest that morphological variations in HF acid-etched microfossils mainly depend on the distribution of kerogen and not silica phase variations between the microfossils and matrix. The presence of kerogen inhibits removal of the associated spheroidal silica, leaving a distinctive morphological biosignature. Consistent with previous work, our SEM and FESEM studies of silicified microfossils have shown the nanoscale structure of the original matter to have been substantially altered by post-mortem degradation processes.

We abandoned our attempts to develop a new vacuum-based HF etching technique (as originally proposed) because the etched surfaces produced with this technique are inferior to other methods and the temperatures required to achieve good results are high enough to be destructive to samples.

Highlights

- In organically preserved microfossils from the Gunflint Iron Formation, kerogen influenced nano-scale mineralogical and ultrastructural signatures by enhancing nucleation of amorphous silica during fossilization and inhibiting recrystallization and growth of crystallites during diagenesis.

• The kerogen of Gunflint microfossils controls the patterns of hydrofluoric acid etching but the form of the residual kerogen left after acid etching may not be representative of the morphologies of organisms at the time of silicification.

• The spatial distribution of kerogen was modified by the morphology of quartz grains during primary silicification and diagenesis, producing preservational artifacts.

Field Trips

Although fieldwork was not initially part of this project, it is very likely that additional samples will be collected from the Gunflint Formation at Mink Mountain in Ontario and from the Mescal Formation at Shell Mountain in Central Arizona. If such fieldwork is required, it will be done by the PI and a new MS student.

Cross Team Collaborations

As Principal Investigator for this project, I have recently begun a new cross-team collaboration with John Holloway and Peggy O'Day (both with the ASU Team project: Organic Synthesis at High Temperatures).

In this collaboration, I will co-supervise a student who will be investigating the morphological, crystallographic, and surface structural characteristics of magnetite used as a catalyst for organic synthesis in experimental hydrothermal systems. We will also characterize the chemistry, morphology, and structure of natural magnetites found in mid-ocean ridge basalts.

NASA Mission Involvement

This project is not directly related to planned NASA missions. However, our mineralogical and ultrastructural criteria for biogenicity will be very important in the search for biosignatures in samples returned from Mars.

Future Directions

• Apply criteria for the recognition of ultrastructural biosignatures we have discovered in organically preserved Gunflint microfossils to the study of problematical microstructures (suspect microfossils) preserved in Fe-oxide rich cherts of the Mescal Formation in central Arizona where organics were removed by oxidation

• Investigate Fe-oxide-rich samples from the Gunflint Formation found at Mink Mountain to investigate the mineralogy and ultrastructure of suspect microfossils in samples preserved under oxidizing conditions, where mineralogical replacements provide the primary biosignatures

Education & Public Outreach

Arizona State University Earth Sciences Day

Annual workshop for grade school and high school students, parents, teachers, and the general public that provides hands-on activities for children and adults to help them understand how science fits into our everyday lives. Faculty, staff, and students from the various astrobiology disciplines at ASU participated in the day-long activities (geological sciences, biology, and chemistry and biochemistry).

Astrobiology Docent Training

Training in astrobiology for docents was given at the California Academy of Sciences. Six hours of lectures were delivered to 111 attendees. NAI brochures and training materials were distributed. We estimate that each docent will reach ~15,000 museum visitors each year with astrobiology information.

Astrobiology Forum

A twice monthly forum sponsored by the ASU Astrobiology Program to promote team communication and interaction.

Astrobiology Presentation

Presentation from Scott Hubbard, Director of the Mars Program for NASA Headquarters, Washington, D.C. at ASU

Astrobiology Program Forum

A seminar with Jack Farmer on developing a formalized Astrobiology Program curriculum at Arizona State University. The purpose of this forum was to discuss present and future plans to develop an astrobiology curriculum to enhance local training and educational opportunities for ASU Astrobiology students. An overview of the present curriculum and our original proposal plan to offer a degree emphasis in astrobiology attached to degrees in geology, chemistry, biology and microbiology was presented by Jack Farmer.

ASU Astrobiology Program Retreat

This event allowed all members of the ASU Astrobiology Program (including many members from universities that are doing collaborative research) to spend a day learning about the overall ASU Astrobiology Program research activities, to update all participants on progress that is being made within each research module, and to develop new collaborative research connections.

BioForum

In June 2001 at the California Academy of Sciences in Golden Gate Park, a “BioForum” for teachers in the San Francisco Bay Area was moderated based on the NAI teaching module, "Astrobiology: Search for life in the Universe". NAI speakers were chosen for the day-long event.

Dr. Rock activity, ASU Earth Science Day

The outreach team helped with rock and mineral displays, methods of identification, visualization and microscopy demonstrations for the public at Arizona State University's Earth Science Day. Over 1200 people attended the event.

Europa Landing Site Project

Six high-school students were selected as part of the Earthwatch Institute 2000 Student Challenge Awards Program to work with ASU researchers for 15 days during the summer of 2000. During the past five years, data for Jupiter's moon Europa were acquired using multispectral and visible imaging systems, and also through the magnetometer and radioscience experiments. Using these data sets, combined with information obtained from the Voyager spacecraft in the late 1970s, a clearer understanding of geologic processes on Europa can be attained. The research goals for this project were to determine the geologic history of Europa's leading hemisphere, and to investigate interactions between the surface and subsurface in order to select sites for data acquisition by the upcoming Europa Orbiter Mission and future lander missions. Student research was conducted through reading relevant scientific literature, image analysis, the mapping of images, evaluation of geophysical models, trend analysis, and discussion.

Europa: Discovery of an Ocean

This project involved the production of an educator slide set (20 slides) which was distributed by members of the ASU Astrobiology team.

Evolution in microbe-based ecosystems:

Desert springs are analogues for the early development and stabilization of ecological systems. This project involves interdisciplinary studies of simple microbial mat-grazer ecosystems of the Cuatro Cienegas Basin in the Chihuahuan Desert of central Mexico. These ecosystems are regarded as analogues for those that arose during the major biosphere transition at the end of Proterozoic following the first appearance of macroscopic grazing animals. Based on an ASU press release following announcement of funding of the project by NAI, the project received News coverage on the web by abcnews.com (http://abcnews.go.com/sections/science/DyeHard/dyehard.html. A film was produced and broadcast highlighting the Cuatro Cienegas project on NHK Television in Japan (April 2001).

Lectures and Presentations

Jack Farmer gave the following lectures and presentations:

- Invited lecture for the Graduate Research Training Seminar series "Early Events in Photosynthesis" group, ASU campus, Topic: "Exploring for Life in the Solar System: Prospects and Challenges".
- Guest lectures were given to the Introduction to Planetary Science course offered at ASU. The first lecture was on "Basic Requirements for Life". The second lecture was on "Exploring for past or present habitable environments in the Solar System".
- Two public lectures entitled "Search for Life on Mars" and "Role of Mass Extinctions in Biospheric Evolution" were given to a local community group, "Spirit of the Senses".

Mars Field Analog Teacher Training Workshop

Field workshop offered by Jack Farmer on behalf of the ASU Mars E&PO Program. Prepared guidebook and lead field workshop for high school science teachers to study the Mono Basin area of California as a Mars Analog.

Media Interviews

Jack Farmer provided the following Media interviews.

- Florida Today: Interviewed with Kelly Young science writer for Florida Today for print article on astrobiology
- Philadelphia Inquirer: Participated in a telephone interview for Philadelphia Inquirer regarding the origin of life and discoveries of 3.8 billon year old fossils in Isua, Greenland.
- Discovery Channel, Science Daily, Canada: Conducted live television interviews from satellite studio in Phoenix to Discovery Canada concerning results from the Mars Global Surveyor mission.
- NHK Japan Video on the human exploration of Mars: met with film crew in the laboratory at Arizona State University and again at field site near Bridgeport, CA to film segments for an educational video series on the human exploration of Mars.
- Press Release, Edinburgh, Scotland: Jack Farmer and graduate student Meredith Payne prepared a press release about their poster presentation on volcano-ice interactions and the search for extant martian life.
- Chain Reaction Magazine: prepared a written interview and illustrations about astrobiology for Chain Reaction Magazine.
- Newsweek: interviewed by Erica Check of Newsweek for an article about the results of the Mar Global Surveyor.
- New York Times: interviewed by Bill Broad of the New York Times for an article about Mars water; provided illustrations for use in article.
- The Mars Society's Martian Chronicles: prepared a written interview that was published in the Mars Society Youth Chapter, National Newsletter, Massachusetts Institute of Technology, May 2001. http://chapters.marssociety.org/youth/

Seminars:

Prof. Martin Fisk of University of Oregon, gave a seminar entitled "Microbes in the Ocean Crust: Applications to the Search for Extraterrestrial Life." This talk reviewed Fisk's work on microbial alteration of seafloor volcanic glass which has emerged as a fascinating new area of geomicrobiological research.

Prof. Ferran Garcia-Pichel of the Dept. of Microbiology, Arizona State Univ. presented a seminar entitled: "Microbes that Follow the Water." A seminar on the microbiology of crusted desert soils of the Southwestern United States, a discussion of the nature of extreme ecosystems and their importance as analogs for extraterrestrial life on other planets such as Mars.

Profs. Laurie Leshin and Thomas Sharp (Dept. of Geological Sciences, Arizona State) presented a seminar entitled: "Putative signs of life in Martian meteorite, ALH 84001." A seminar on whether life existed in the Martian meteorite and the controversies surrounding this meteorite.

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Back to Table of Contents

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Back to Table of Contents

Carnegie Institution of Washington

NASA Astrobiology Institute

Carnegie Institution of Washington Team Members

Michael J. Acierno
Carnegie Institution of Washington

Conel M. O'D. Alexander
Carnegie Institution of Washington

John A. Baross
University of Washington

James Bischoff
USGS Menlo Park

Nabil Z. Boctor
Carnegie Institution of Washington

Alan P. Boss
Carnegie Institution of Washington

Charles Kevin Boyce
Harvard University

Chris Bradburne
George Mason University

Jay A. Brandes
University of Texas, Austin

R. Paul Butler
Carnegie Institution of Washington

Gavin W. Chan
Washington University

Kenneth M. Chick
Carnegie Institution Of Washington

I-Ming Chou
USGS Reston

Ines Lucia Cifuentes
Carnegie Institution of Washington

Mary M. Coder
Carnegie Institution of Washington

George D. Cody
Carnegie Institution of Washington

Steven J. Desch
Carnegie Institution of Washington

Andrew J. Dombard
Carnegie Institution of Washington

Janice S. Dunlap
Carnegie Institution of Washington

David Emerson
George Mason University

Timothy R. Filley
Purdue University

Marilyn L. Fogel
Carnegie Institution of Washington

Glenn A. Goodfriend
George Washington University

Shaun J. Hardy
Carnegie Institution of Washington

Erik H. Hauri
Carnegie Institution of Washington

Robert M. Hazen
Carnegie Institution of Washington

Russell J. Hemley
Carnegie Institution of Washington

Julie Adrienne Huber
University of Washington

Wesley T. Huntress
Carnegie Institution of Washington

Satoshi Inaba
Carnegie Institution of Washington

Charles C. James
Carnegie Institution of Washington

Catherine L. Johnson
Carnegie Institution of Washington

Samantha Joye
University of Georgia

Jonathan Z. Kaye
University of Washington

Karl Kehm
Carnegie Institution of Washington

Sandra A. Keiser
Carnegie Institution of Washington

Stephen Kortenkamp
Carnegie Institution of Washington

Kono Lemke
Stanford University

Mausmi P. Mehta
University of Washington

D Arcy Meyer
Washington University

Michelle E. Minitti
Carnegie Institution of Washington

Harold J. Morowitz
George Mason University

Kenneth H. Nealson
Jet Propulsion Laboratory

David S. Ross
USGS Menlo Park

Gotthard Saghi-Szabo
Carnegie Institution of Washington

Matthew O. Schrenk
University of Washington

James H. Scott
Carnegie Institution of Washington

Anurag Sharma
Carnegie Institution of Washington

Everett Shock
Washington University

Sean C. Solomon
Carnegie Institution of Washington

Terry L. Stahl
Carnegie Institution of Washington

Andrew Steele
Johnson Space Center

Jianhua Wang
Carnegie Institution of Washington

George W. Wetherill
Carnegie Institution of Washington

Hatten S. Yoder, Jr.
Carnegie Institution of Washington

Misha Y. Zolotov
Washington University

Executive Summary

Accomplishments

The astrobiology team led by the Carnegie Institution of Washington is studying the physical, chemical, and biological evolution of hydrothermal systems, including vent complexes associated with ocean ridges, deep aquifers, and other subsurface aqueous environments, both on Earth and on other Solar System and extrasolar bodies. Such diverse systems are important environments for life on Earth and possibly elsewhere in the cosmos. We also continue a strong observational and theoretical program related to understanding the formation and early evolution of both our Solar System and extraterrestrial solar systems.

The traditional view of life's origin on Earth has focused on processes near the photic zone at the ocean-atmosphere interface, where ionizing radiation provides energy for prebiotic organic synthesis. In the context of astrobiology, this origin paradigm restricts the initial "habitable zone" around stars to planets and moons with surface water. According to this view, subsequent adaptations on Earth, and possibly elsewhere, led to expansion of the biosphere into subsurface habitats.

An alternative hypothesis is that life-forming processes may also occur in subsurface hydrothermal environments at the water-mineral interface. This hypothesis, that life on Earth originated from oxidation-reduction reactions in deep hydrothermal zones, perhaps at or near ocean ridge systems, opens exciting possibilities for astrobiological research. If a subsurface, high-pressure origin of life is possible, then the initial habitable zone around stars is greatly expanded to aqueous environments where redox reactions can be driven along thermal and chemical gradients.

Several lines of evidence lend credibility to the hydrothermal origins hypothesis. Numerous recent discoveries of high-pressure life, especially lithotrophic prokaryotes, suggest that hydrothermal environments support abundant life. Models of the Earth's formation postulate large, surface-sterilizing impacts as recently as 3.8 billion years ago, but deep hydrothermal zones may have insulated life from these traumas. Studies of molecular phylogeny reveal that thermophilic microbes are perhaps the closest living relatives of the last universal common ancestor. Finally, hydrothermal organic synthesis experiments reveal unexpectedly facile synthetic pathways. Whether or not life originated in a subsurface hydrothermal zone, these lines of evidence, coupled with the assumed widespread distribution of such environments in our Solar System and elsewhere, point to the need and opportunity for an intense study of the characteristics of hydrothermal systems.

Our team's research activities explore the physical, chemical, and biological evolution of hydrothermal systems from these complementary fronts:

- We model planetary formation, and we detect and characterize extrasolar planets, in an effort to understand the range of objects that develop hydrothermal systems as well as the distribution of volatiles, especially water, within those objects.

- We investigate the circumstances under which hydrothermal systems form on planets and other bodies and the expected physical and chemical characteristics of those systems as they evolve.

- We study geochemical processes in hydrothermal systems, especially those that lead to abiotic organic synthesis. A particular focus is the role of mineral catalysis in these systems.

- We consider the origin and evolution of biological entities in hydrothermal systems through studies of the biochemistry of contemporary hydrothermal organisms.

A complete understanding of hydrothermal systems and their role in life's origins requires dramatic advances on all of these fronts, as well as an extensive and challenging integration of these topics. During the past year we achieved significant progress in each of these research areas, as well as increased attention to the interfaces among these theoretical, experimental, and field approaches.

Among the highlights from the past year's research in the area of planetary formation and evolution, members of our team

- Discovered 12 new extrasolar planets, including the second and third known examples of multiple-planet systems.

- Demonstrated that Jupiter-mass protoplanets can form from instabilities in a protoplanetary disk of a mass similar to the solar nebula.

- Demonstrated that Mars-size planetary embryos can form in the inner Solar System even in the presence of rapidly formed outer planets.

- Showed that the Tharsis rise on Mars was largely emplaced prior to the formation of late Noachian valley networks, and that the volcanic construction of Tharsis may have contributed significantly to the atmospheric inventory of water and other volatiles.

In the area of the evolution of organic matter and water in meteorites, we

- Demonstrated that shock waves in the solar nebula may have led to melting and chondrule formation as well as the breaking of N_2 bonds to form nitriles, which can then polymerize or contribute to amino acid synthesis.

- Developed stringent new constraints on the characteristics of the Murchison organic macromolecule from nuclear magnetic resonance experiments.

- Documented petrological evidence for pervasive premetamorphic aqueous alteration of ordinary chondrite meteorites, which implies a more sunward position of the water-ice condensation line than has been heretofore assumed.

- Discovered D-rich structural water in a post-stishovite silica phase in martian meteorites, providing additional evidence linking D-rich water and shock processes accompanying impact cratering on Mars.

- Developed improved inductively coupled plasma mass spectrometer techniques for precise measurements of Fe isotopes as a tool for identifying possible isotopic biomarkers.

In the area of experimental tests of proposed hydrothermal organic synthesis reactions, our group has

- Abiotically synthesized pyruvic acid — a key molecule for the emergence of biochemistry — under hydrothermal conditions.

- Identified a viable and robust "geochemical" ignition point for the prebiotic fixation of carbon under hydrothermal conditions.

- Synthesized vesicle-forming amphiphilic molecules from pyruvic acid under hydrothermal conditions — compounds strikingly similar to those extracted from the Murchison meteorite.

- Challenged the notion that formaldehyde/HCN chemistry led to amino acid formation on the primitive Earth by demonstrating that an early CO_2/N_2 atmosphere could not provide sufficient quantities of these precursors.

- Demonstrated that ammonia is readily synthesized from nitrate under hydrothermal conditions in the presence of a wide variety of transition-metal oxide and sulfide minerals.

- Developed experimental techniques for observations of fluid-phase behavior and *"in situ"* determination of organic reaction kinetics at hydrothermal conditions.

In the area of supporting theoretical studies of hydrothermal synthesis reactions, we

- Established a global thermodynamic framework for metabolism in thermophilic and hyperthermophilic prokaryotes.

- Demonstrated that the hydrocarbons detected in ALH84001 likely were formed abiotically through reactions of CO and H_2 in the presence of magnetite.

- Showed that the citric acid cycle may be derived from physical and chemical selection rules suggesting an emergent quality of the chemistry distinct to its environment.

In the area of biological studies of hydrothermal systems, members of our consortium

• Showed that hyperthermophilic archaea isolated from the subseafloor near deep-sea vents are phylogenetically and physiologically different from similar organisms isolated from vent sulfide structures.

• Described a novel and deeply rooted hyperthermophilic archaea which can grow on organic acids such as acetate and citrate with Fe (III) as the electron acceptor.

• Characterized the subsurface bacterial and archaeal communities at Axial Volcano, Juan de Fuca Ridge.

• Designed a molecular probe to detect the nitrogen fixation gene in archaea and successfully detected the gene in subseafloor hyperthermophiles.

• Demonstrated that Fe-oxidizing bacteria are widespread and play a substantial role in the deposition of Fe oxyhydroxide deposits in Fe-rich oxic-anoxic boundary habitats on Earth.

• Demonstrated the viability of biochemical activity at pressures exceeding 10,000 atmospheres.

• Demonstrated that calcite crystals, when immersed in a racemic aspartic acid solution, display significant adsorption and chiral selectivity of D- and L-enantiomers on pairs of mirror-related crystal growth surfaces, thereby providing a plausible geochemical mechanism for chiral selection and subsequent homochiral polymerization of amino acids on the prebiotic Earth.

Under a new project initiated this past year, members of our team are developing protein chip-based molecular recognition technology as a method for detecting life on Earth and other Solar System bodies. The Ciphergen Biosystems Protein Chip Reader utilizes the surface chemistry of small chips to capture selectively femtomole amounts of complex mixtures of organic molecules for molecular weight determination by time-of-flight mass spectrometry. Together with collaborators from several other NAI teams, our group has begun to intercompare different types of organic molecules. An initial study demonstrated that microbial proteins, biologically produced but chemically modified microbial molecules, and abiotically synthesized organic matter can be distinguished on the basis of their respective mass spectrometric characteristics.

Finally, our team hosted the second General Meeting of the NASA Astrobiology Institute at the Carnegie Institution of Washington's administrative headquarters, and one of our Co-Investigators chaired the program committee for the meeting.

In summary, our team's recent research, including discoveries of new planetary systems, exploration of possible hydrothermal regimes on other worlds, elucidation of robust hydrothermal synthetic pathways, documentation of novel microbial metabolic strategies, and finding unexpected high-pressure environments for life, inform the central questions of astrobiology. Taken together, these discoveries are changing our views of life's origin and its distribution.

Roadmap Objectives

#1
Sources of Organics on Earth

#2
Origin of Life's Cellular Components

#6
Microbial Ecology

#9
Life's Precursors & Habitats in the Outer Solar System

Project

A New Molecular Recognition Instrument for Astrobiological Applications

Senior Project Investigator(s):
M. Fogel, D. Emerson, R. Hazen

Accomplishments

Protein chip-based molecular recognition technology is being developed as a method for detecting life on Earth and other Solar System bodies. The Ciphergen Biosystems Protein Chip Reader (PBSII) is dependent on the surface chemistry of small chips to selectively capture femtomole amounts of complex mixtures of organic molecules for molecular weight determination by time-of-flight mass spectrometry. By using different chips, experiments can be designed to recognize a complex combination of molecular markers by selecting a series of chip surface chemistries to pull apart complex mixtures.

- Proteomics. We determined the protein profile of *Shewanella putrefascens*, a microorganism that is commonly isolated from geochemical gradient zones in which dissolved oxygen levels range from fully oxic to completely anoxic. Replicate analyses of the chip detected 62 peptides or proteins from 754 to 237,954 daltons, some of which corresponded to molecular weights of proteins identified from the genome. The instrument detected proteins in the lower picomole to higher femtomole range.

- Geochemical Studies. On Earth, as well as Europa, the bulk of organic carbon molecules in the world's oceans are found dissolved in seawater. However, the chemical composition of dissolved organic matter is complex and poorly understood, making it a good proxy to what might exist on Europa or Mars.

The molecular weight distribution of organic molecules was determined in dissolved organic matter (DOM) to see whether large proteins or microbial cell wall fragments could be detected from the Pacific Ocean (100 m depth) and from a riverine-derived filtrate (<0.2 microns). The molecular weight distribution precludes any intact proteins in the sample. The molecules between 1,000 and 6,000 daltons could be explained by units of microbial cell walls. Last and most important, the Surface Enhanced Laser Desorption Ionization (SELDI) technique identified a biosignature in both of the samples–the common C-14 peaks and an additional biological source in oceanic DOM not found in the riverine sample.

Highlights

We compared three different types of organic molecules, including microbial proteins, biologically produced but chemically modified microbial molecules, and abiotically synthesized organic matter. The molecular weight ranges of the three groups were found to be segregated: biological material (up to 250,000 Daltons), geochemical molecules (up to 6,000 Daltons), and abiotic molecules (up to 1,000 Daltons).

Field Trips

Belize, August and November 2000

With Biocomplexity project, including Matthew Wooller (CIW) and Samantha Joye (Univ. of Georgia); to sample microbial mats

Cross Team Collaborations

Collaboration is ongoing with the NAI teams from JPL and NASA Johnson Space Center. Additional inter-team collaborations will be initiated in the coming year.

NASA Mission Involvement

Protein chip-based molecular recognition technology is being developed as a method for detecting life on Earth and other Solar System bodies. By using different chips, experiments can be designed to recognize a complex combination of molecular markers by selecting a series of chip surface chemistries to pull apart complex mixtures.

We would like to develop this instrument concept and its capabilities for eventual adaption to space flight and the *in situ* analysis of martian soils.

Future Directions

- Expand the list of microbes investigated with this technology, to include the Archaea
- Increase the number of collaborators participating in the project
- Become more familiar with the use of antibody binding chips

Roadmap Objectives

#1
Sources of Organics on Earth

#2
Origin of Life's Cellular Components

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#9
Life's Precursors & Habitats in the Outer Solar System

#12
Effects of Climate & Geology on Habitability

#14
Ecosystem Response to Rapid Environmental Change

Project

Biological Studies of Hydrothermal Systems

Senior Project Investigator(s):
J. Baross, G. Cody, D. Emerson, M. Fogel, R. Hazen

Accomplishments

Task 1. Field Studies and Laboratory Characterization of Hydrothermal Vent Microbes

We have completed a study showing that the hyperthermophilic archaea isolated from the subseafloor are phylogenetically and physiologically different from similar organisms isolated from nearby sulfide structures. These results show that the subseafloor in vent systems is a biotope distinct from massive sulfide environments, with unique characteristics and a separate group of indigenous hyperthermophilic species. In a related study, we have described an extremely novel and deeply rooted hyperthermophilic archaea, which we are naming *Saganella petroecbolus*. This organism has simple nutritional requirements and can grow on organic acids such as acetate and citrate with Fe (III) as the electron acceptor forming magnetite. It can also grow with CO_2 and H_2. It appears that the normal lifestyle for *Saganella* is to attach to mineral surfaces and form biofilms, a characteristic we believe to be canonical for subseafloor microorganisms.

The molecular phylogenetic analyses of the subsurface bacterial and archaeal communities from the 1998 deep-sea eruption at Axial Volcano, Juan de Fuca Ridge, has been completed for samples collected in 1998, 1999, and 2000. The results from this study show that the subseafloor archaeal community at diffuse-flow vents is a complex mixture of seawater-entrained and indigenous species. Hyperthermophilic and mesophilic methanogens and Thermococcales are clearly indicator-organisms for an anaerobic subseafloor biosphere. The *Thermococcus* species, while phylogenetically closely related, have markedly different phenotypic characteristics including different protein patterns and enzyme activities. There are also a large number of archaeal sequences that are not found in seawater that appear to be unique to the subseafloor.

A molecular probe has been designed to detect the NIF (nitrogen fixation) gene in archaea and has been used successfully to detect the NIF gene in hyperthermophiles from subseafloor environments. Data from the 2000 cruise to Axial volcano indicate an extremely high diversity of both bacteria and archaea that have the NIF gene. Most of the archaeal NIF genes appear to be associated with methanogens and uncultured marine Crenarchaeota.

A preliminary description of the microbial ecology of active sulfide chimneys has been completed using a combination of molecular and microscopic analyses. Intact microbes have been observed throughout these sulfide structures, including in mineral zones thought to be at temperatures greater than 150°C. This work has also been expanded to include the newly discovered (December 2000) Lost City vent field on the Mid-Atlantic Ridge. This is a unique vent system since the fluids are devoid of sulfide and enriched in carbonate, hydrogen, and methane. The smoker structures consist of carbonates. The preliminary observations indicate unique micro- and macro-faunal communities.

Task 2. Studies of Neutrophilic, Lithotrophic, Fe-oxidizing Bacteria

Iron is potentially one of the Solar System's most abundant energy supplies, and it is the fourth most abundant element in the Earth's crust, yet only in the last 10 years has unequivocal evidence been found to support the notion that neutrophilic Fe-oxidizers are truly lithotrophic microorganisms. Our work during the past year has focused on a combination of biodiversity, morphological, and physiological studies aimed at answering fundamental questions about this unique form of metabolism.

- Biodiversity: We have discovered a rich habitat for Fe-oxidizing microbes in the rhizosphere of wetland plants where microoxic zones around the root mass of wetland plants create an ideal habitat in an otherwise anoxic subsurface for Fe(II) oxidation. As much as 5% of the total population of microbes (approximately 10^7 cells/cc of root) can be lithotrophic Fe-oxidizers. Interestingly, these root-associated Fe-oxidizers are closely related to other terrestrial and marine Fe-oxidizers.

- Morphology: A unique feature of Fe-oxidizing bacteria (FeOB) is the signature morphologies of the Fe-oxides formed as a result of their metabolism, normally in the form of a tubular sheath or a helical stalk. We have isolated a strain of FeOB from the Loihi Seamount hydrothermal vent that forms a unique filamentous Fe-oxide. Recent work by several investigators on microfossils from different sources (e.g., ancient hydrothermal vent sites or lake beds) and of different ages (up to at least 500 Myr) have shown Fe-silicate structures that look remarkably like the remnants of modern-day Fe-oxidizers.

- Physiology: We have been conducting growth studies using a bioreactor that allows precise control of O_2, pH, temperature, and Fe(II) concentration to determine the relative portion of Fe-oxidation carried out by the bacteria compared to abiological Fe-oxidation. These measurements are difficult to make, but preliminary estimates are that 30 - 60% of the Fe-oxidation is microbially mediated, the amount depending upon specific conditions of O_2 and Fe(II) concentrations.

- Field studies: For the past 15 months, we have collected field data on Fe(II), O_2, pH, temperature, and conductivity, as well as samples for molecular community analysis, from a high-iron, freshwater aquatic field site that features both low pH (pH 2 - 4) and neutral (pH 5.5 - 7) flow regimens. These data will be useful background information for future field studies aimed at coupling diversity and physiology studies.

Task 3. Possible Origin of Chirality

We initiated studies of the selective adsorption of L- and D-amino acids on calcite – work that has implications for the origin of biochemical homochirality. One of life's most distinctive biochemical signatures is its strong selectivity for chiral molecular species, notably L-amino acids and D-sugars. Prebiotic synthesis reactions, with the possible exception of some interstellar processes, yield essentially equal amounts of L- and D-enantiomers. A significant challenge in origin-of-life research, therefore, is to identify natural mechanisms for the homochiral selection, concentration, and polymerization of molecules from an initially racemic mixture. Symmetry breaking on a chirally-selective mineral surface in an aqueous environment offers a viable scenario for the origin of life. We demonstrated that crystals of the common rock-forming mineral calcite ($CaCO_3$), when immersed in a racemic aspartic acid solution, display significant adsorption and chiral selectivity of D- and L-enantiomers on pairs of mirror-related crystal growth surfaces. This selective adsorption is greater on crystals with terraced surface textures, which suggests that D- and L-aspartic acid concentrate along step-like, linear growth features. Selective adsorption of D- and L-amino acids on calcite is thus a plausible geochemical mechanism for the chiral selection and subsequent homochiral polymerization of amino acids on the prebiotic Earth.

Task 4. Emergence

We have been investigating the role of emergence in the origin of life. Natural systems with many interacting components, such as atoms, molecules, cells or stars, often display complex, emergent behavior not associated with their individual components. In some instances, as in the emergence of turbulent flow in fluids, the solid-state properties of crystals, or the periodic spacing of sand dunes, such complex behavior can be modeled *a posteriori* when appropriate interaction parameters have been determined. Other phenomena, such as the emergence of consciousness from collections of neurons or the emergence of social behavior from collections of humans, are, at least for the present, less amenable to quantitative analysis. The geochemical origin of life may be modeled as a sequence of "emergent" events, each of which adds to molecular complexity and order. Each of these steps, if properly formulated, should be amenable to experimental study. Each emergent step, furthermore, may result in characteristic isotopic, molecular, and structural "fossils" that might be measured in extraterrestrial environments that have not been subjected to reworking by biological activity.

The observed emergent behavior of highly ordered systems, including galaxies, planets, and life, points to a universal organizing principal. Natural systems tend to develop local-scale order – non-equilibrium regions of spontaneously increased free energy and decreased entropy – even as global-scale entropy increases. This universal tendency for systems to display increased order at an energy-rich interface, while consistent with the first and second laws of thermodynamics, is not formally addressed in either of those laws; indeed, some researchers have proposed that this behavior should be systematized in a "fourth law of thermodynamics."

If the chemical evolution of life occurred as a sequence of successively more complex stages of emergence, then the divide between non-life and life may be illdefined. One might establish a hierarchy of emergent properties – a progressive sequence that leads, for example, through a number of steps from a pre-biotic ocean enriched in organic molecules, to a cluster of molecules arranged on a mineral surface, to self-replicating molecular systems, to encapsulation and eventually prokaryotic life. The exact nature and sequence of these steps may vary in different environments, but any definition that distinguishes between non-living and living systems of necessity becomes more arbitrary as the number of discrete emergent steps to life increases.

The concept of a sequence of discrete emergent steps is useful and appealing in experimental and theoretical studies of the origin of life for at least two pragmatic reasons. First, a progression of steps reduces an immensely complex historical process to a succession of several, more manageable chemical episodes. Each step becomes a focused process for laboratory experimentation or theoretical modeling. Second, each of these steps may result in distinctive, measurable isotopic, molecular, and structural signatures. As we search for life elsewhere in the universe, we may thus be able to characterize extraterrestrial environments according to their degree of emergence along this multi-step path. In this context, it is useful to review experimental programs that attempt to elucidate a few of the possible geochemical steps in the emergence of life.

Highlights

Task 1

- Showed that hyperthermophilic archaea isolated from the sub-seafloor near deep-sea vents are phylogenetically and physiologically different from similar organisms isolated from vent sulfide structures

- Described a novel and deeply rooted hyperthermophilic archaea which can grow on organic acids such as acetate and citrate with Fe (III) as the electron acceptor forming magnetite

- Characterized the subsurface bacterial and archaeal communities at Axial volcano, Juan de Fuca Ridge

- Designed a molecular probe to detect the nitrogen fixation gene in archaea and successfully detected the gene in sub-seafloor hyperthermophiles

Task 2

- Lithotrophic FeOB are abundant and widespread and play a substantial role in the deposition of Fe oxyhydroxide deposits in extant Fe-rich oxic-anoxic boundary habitats on Earth.

- Comparative studies of ancient Fe-silicate microfossils and modern-day FeOB may lead to new insights about the role of FeOB in earlier Earth environments, and as potential biomarkers for discovery of life processes on other planets.

Task 3
Demonstrated that calcite crystals, when immersed in a racemic aspartic acid solution, display significant adsorption and chiral selectivity of D- and L-enantiomers on pairs of mirror-related crystal growth surfaces, thereby providing a plausible geochemical mechanism for chiral selection and subsequent homochiral polymerization of amino acids on the prebiotic Earth

Field Trips

Axial Volcano, Juan de Fuca Ridge, July 2000.
Archaeal and bacterial diversity in the hot subsurface environment using molecular phylogeny methods. Incidence and characteristics of the NIF gene (nitrogen-fixing gene) in subseafloor microbial communities. Isolation and characterization of novel hyperthermophilic archaea and bacteria. NOAA/SeaGrant sponsored cruise. Participants: J. Baross and students J. Huber and M. Mehta.

Endeavour vent field, Juan de Fuca Ridge, June 2000; September 2000.
Chief-scientists, J. Delaney and D. Kelly. Baross support to participate on these cruises comes from the NAI. The diversity of hyperthermophilic archaea and halophilic bacteria and archaea from diffuse-flow vents and sulfides using molecular and culturing methods. Participants: J. Baross and students Jon Kaye and Matt Schrenk.

Virginia, USA, May 2000.
For the past 15 months Co-I Emerson has collected data on Fe(II), O_2, pH, temperature, and conductivity, as well as samples for molecular community analysis, from a high iron, freshwater aquatic field site that features both low pH (pH 2 - 4) and neutral (pH 5.5 - 7) flow regimens. This site is located in Virginia, a 1 hr drive from the Co-I's laboratory. This will be useful background information for future field studies aimed at coupling diversity and physiology studies, as well as mineralogical studies.

New Vent and Endeavour vent field, Juan de Fuca Ridge, June 2000.
Chief-scientist, H. P. Johnson. Baross support to participate on these cruises comes from NAI and NSF LExEn proposal. Diversity of hyperthermophilic archaea from high-temperature and diffuse-flow hydrothermal fluids. Molecular methods to quantify specific groups of archaea including methanogens and determine if nitrogen-fixing gene is expressed *in situ*.

Juan de Fuca Ridge, Axial Volcano, July 2001.
Archaeal and bacterial diversity in the hot subsurface environment using molecular phylogeny methods. Incidence and characteristics of the NIF gene (nitrogen-fixing gene) in subseafloor microbial communities. Isolation and characterization of novel hyperthermophilic archaea and bacteria. Measure nitrogen fixation *in situ* and from enrichment cultures. NOAA/SeaGrant sponsored cruise. Participants: J. Baross and students J. Huber and M. Mehta.

Cross Team Collaborations

During this past year, scientists at CIW (Hazen, Fogel, Cody) have continued collaborative studies with members of the Harvard NAI team (Knoll, Boyce, Nofke) for this research project.

This research has focused on new techniques for microanalytical paleontology – the characterization of composition, isotopic signatures, and molecular species of ancient fossils. Work with doctoral student Boyce culminated in a new technique for mapping carbon distribution in fossils at the micron scale. This work included applications of X-ray microscopy and isotope analysis to ancient plant fossils.

NASA Mission Involvement

Co-I Baross became a member of the NASA Science Definition Team (SDT) for the 07 Mars Lander. This Mars lander and rover mission plan involves a mobile scientific laboratory and small scout missions.

Future Directions

Task 1

Characterization of recovered hydrothermal vent microbes and their physical and chemical environments will continue. Additional cruises to vent sites are planned for the coming year.

Task 2

More effort will be focused on understanding the physiology of FeOB and investigating the mineralogical implications of the unique morphological Fe-oxide structures of these microbes. The ultimate goal of this work is to understand the biochemical mechanism of lithotrophic Fe-oxidation. It is expected that Co-I Emerson will also interact with Co-I's Fogel and Hauri at CIW, investigating different aspects of C and Fe isotope work. These collaborations have already been initiated.

Task 3

In the next year, we plan to study the selective adsorption of other amino acids onto calcite and other minerals (gypsum). We will also study the molecular-scale mechanism of amino acid adsorption using liquid immersion atomic force microscopy, in collaboration with Henry Teng of George Washington University.

Task 4

Collaborative studies with Co-I Harold Morowitz and with Juan Perez Mercader of the Centro de Astrobiología are now planned. Ultimately, we hope to develop a mathematical framework for describing and predicting emergent properties.

In the coming year Kevin Boyce, who received his Ph.D. in May 2001, will be an NAI Fellow, working jointly with NAI scientists at Harvard and CIW. The group's objectives include developing techniques for increased resolution of carbon isotopes and application to a variety of Precambrian fossils.

Roadmap Objectives

#1
Sources of Organics on Earth

#2
Origin of Life's Cellular Components

#7
Extremes of Life

#8
Past and Present Life on Mars

#9
Life's Precursors & Habitats in the Outer Solar System

#11
Origin of Habitable Planets

#13
Extrasolar Biomarkers

Project

Hydrothermal Organic Synthesis

Senior Project Investigator(s):
G. Cody, R. Hazen, R. Hemley,
J. Bischoff, D. Ross

Accomplishments

Task 1. Abiotic Hydrothermal Synthesis

Our approach is to explore the catalytic capabilities of transition-metal sulfides for the promotion of organic reactions that have biochemical utility. Over the past year we have demonstrated that we can synthesize pyruvate under extreme conditions. Specifically, reactions involving aqueous formic acid, FeS, and alkyl thiol promote double carbonyl insertion reactions leading to the formation of alpha-keto acids. Within the product suite we identified carbonylated organometallic phases. We conclude that the formation of alpha-keto acids proceeds homocatalytically through reaction between the alkyl thiols and the organometallic phases. Within central metabolism, pyruvate lies at the junction of several key reaction pathways, including sugar synthesis, amino acid synthesis, as well as the oxidative and reductive citrate cycles. The synthesis of pyruvate is, therefore, a critically important prebiotic reaction for the emergence of biochemistry.

The facile synthesis of carbonylated FeS clusters is also important for the emergence of primitive biochemistry. First, the formation of such organometallic phases might provide the critical first step for the generation of ferrodoxin based electron networks. Second, the predominant organic metallic phase formed in our reaction is $Fe_2S_2(CO)_6R_2$. The structure of this complex is very similar to the active center (the "H-cluster") within the enzyme hydrogenase, a ubiquitous catalytic element in the generation of reducing power in anaerobic microorganisms. Note also that hydrogenase obtains the electrons necessary for reduction of a broad range of oxidants through the oxidative decarboxylation of pyruvate.

A second component of our research involves deriving the most likely first pathway for carbon fixation. Recent theories have proposed that life arose from primitive hydrothermal environments employing chemistry analogous to the reductive citrate cycle as the primary pathway for carbon fixation. This chemistry is presumed to have developed as a natural consequence of the intrinsic geochemistry of the young, prebiotic Earth. However, there has been no experimental evidence that there exists a natural pathway into such a cycle. We have now demonstrated, experimentally, a viable route for carbon fixation with clear relevance to the origins of life. The path involves the conversion of propene and CO_2 up to a tricarboxylic acid, citric acid, via the Koch (hydrocarboxylation) reaction promoted heterocatalytically using NiS in the presence of a source of CO. These results point to a simple hydrothermal redox pathway for citric acid synthesis that may have provided a geochemical ignition point for the reductive citrate cycle.

Task 2. Hydrothermal Chemistry

We are developing techniques to monitor directly hydrothermal chemistry and its effect on biological activity, including the development of experimental procedures to monitor biological activity at extreme physical and chemical conditions. Preliminary experimental results have indicated the viability of biochemical activity at very high pressures, an order of magnitude higher than previously estimated limits on life. These results further expand the concept of what constitutes a "habitable zone," shed light on whether pressure has any evolutionary component, and provide an experimental procedure for *in situ* study of life in extreme environments.

Experimental techniques were also developed to monitor directly organic reactions and the effects of fluid phase behavior on their reaction kinetics. Experiments involve direct observation of fluid behavior at extreme conditions of temperature and pressures, an order of magnitude higher pressure than in previous studies. Such studies provide an important contribution to our understanding of the phase behavior of mixed fluids, the availability of fluid species for any biochemical activity, and the formation of any metastable and weakly bonded structures (such as gas hydrates). Studies were also conducted on the stability and structures of gas hydrates, resulting in the determination of two new phases of gas hydrates at pressures exceeding 10 kbar.

Task 3. Amphiphiles

The synthesis of amphiphilic molecules capable of aqueous self-assembly into membrane-like structures, including bilayers and micelles, is an essential step in the emergence of a protocell. Mechanisms for the prebiotic synthesis and assembly of amphiphiles are thus of considerable interest. We observe amphiphile synthesis from pyruvate in a CO_2-water fluid subjected to temperatures from 250ºC to 350ºC and pressures from 0.05 to 0.2 GPa (500 to 2000 atmospheres) for 2 hours. Principal run products include acetic acid and CO_2 (from decarbonation of pyruvate) and methyl succinate (from dimerization of pyruvate and subsequent decarboxylation). We also observe up to 50% conversion of pyruvate to a water-insoluble, yellow-brown, strongly aromatic oily residue. Analysis by GCMS reveals this material to be a complex suite, including cyclic aromatic compounds, presumably formed by polymerization and subsequent cycloaddition reactions. We characterized this material by extracting and analyzing the chloroform-soluble fraction. We examined this material by 2D thin layer chromatography (TLC), which revealed a pattern of seven distinct fluorescent regions – a pattern that is qualitatively similar to that observed for amphiphilic components of the Murchison meteorite. Each of these seven TLC areas was extracted, dried, washed in a phosphate buffer (pH = 8.5), and examined by fluorescence microscopy. One of these regions consisted of a significant fraction of surface-active molecules, which form apparent monomolecular films at air-water interfaces. When placed in the phosphate-buffered aqueous solution, these molecules also organize into fluorescent, membranous vesicle-like structures. We conclude that hydrothermal processes, perhaps similar to those that occurred on the Murchison parent body, may lead to the production of amphiphilic, membrane-forming molecules.

Task 4. Amino Acid Synthesis Under Hydrothermal Conditions

We are building on our earlier work on the synthesis of amino acids from H_2CO and HCN on various common mineral surfaces. We have recently considered the question of an endogenous source of amino acids, and we have carried out calculations showing that a primitive CO_2/N_2 atmosphere in equilibrium with water and an FMQ (fayalite/magnetite/quartz) redox buffer could not have yielded sensible quantities of H_2CO and HCN for amino acid formation. The calculations show that the expected aqueous species are ammonium and formate, at concentrations well into the mM range for $CO_2 = N_2 = 1$ bar.

We thus conclude that an endogenous basis for life must be based upon those ions. Since C-C bond formation is a necessary feature of early life-related chemistry, formate as the principal reactant in Fischer-Tropsch-synthesis (FTS) is an attractive consideration. The FTS variant described by Koelbel and Trapper satisfies another critical element of the early chemistry — C-N bond formation — and it can be shown further that at temperatures below 100ºC an FMQ-controlled aqueous system provides hydrogen fugacities sufficient to produce quantities of glycine > 1 M via

$$2\ CO_2(g) + 0.5\ N_2(g) + 4.5\ H_2(g) \rightleftarrows H_2NCH_2CO_2H(aq) + 2\ H_2O$$

This approach is not without its challenges, however. The third essential ingredient of the Earth's early organic chemistry must be a process providing branching and polyfunctionality (i.e., routes leading to both carboxyl and amino substitution). Here a Fischer-Tropsch approach must be carefully appraised since conventional FTS yields solely alkanes or monofunctionalized alkanes.

Our current activities consider the likelihood that the elevated temperatures in conventional FTS trigger secondary, high-activation-energy reactions that eliminate initial polyfunctionality. These reactions would vanish kinetically at lower temperatures, and our current efforts include studies seeking such products in FTS at lower temperatures.

Task 5. Nitrogen Fixation

The reduction of nitrite and nitrate to ammonia is an important step in the prebiotic synthesis of ammonia. We have investigated the hydrothermal formation of ammonia at 300ºC and 50 MPa in the presence of a variety of transition metal oxide and sulfide minerals. We find that nitrate reduction occurs readily in the presence of most of these minerals. This result suggests that hydrothermal systems produced significant quantities of ammonia in the prebiotic oceans.

Highlights

Tasks 1 and 2

- We have abiotically synthesized pyruvic acid under hydrothermal conditions; pyruvate is a key biomolecule for the emergence of biochemistry.

• We have identified a viable and robust geochemical ignition point for the prebiotic carbon fixation. Chemistry that is similar to that of the reductive citrate cycle occurs readily under hydrothermal conditions.

• We have demonstrated the viability of biochemical activity at pressures exceeding 10,000 atmospheres, experimental techniques for "direct" observations on microbial activity at extreme conditions, fluid phase-behavior at hydrothermal conditions, and *in situ* determination of organic reaction kinetics at hydrothermal conditions.

Task 3

We have synthesized vesicle-forming amphiphilic molecules from pyruvic acid under hydrothermal conditions. Furthermore, we observe striking similarities between these synthetic compounds and those extracted from the Murchison carbonaceous chondrite.

Task 4

• The common notion that formaldehyde/HCN chemistry led to amino acids on the primitive Earth is challenged by the demonstration that the early CO_2/N_2 atmosphere could not provide sensible quantities of those precursors. It can be shown specifically that the fayalite/magnetite/quartz redox buffer is insufficiently reducing to produce them, and that ammonium and formate are the more likely precursor candidates.

• This finding suggests a direct "test" of the endogenous route to life: Seeking aqueous ammonium formate conversions to product molecules that include the critical molecular features for life's precursors, branching, and difunctionality. A failure to find such product substances would strongly suggest that life's origins are exogenous.

• In accord with the model proposed by Orgel, our analysis of the kinetic data reported by Matsuno for production of peptides from amino acids in a hydrothermal setting suggests that the pertinent chemistry must take place on the reactor surfaces rather than in homogeneous solution. It is notable that the Matsuno study utilized unactivated amino acids.

Task 5

We find that ammonia is easily synthesized from nitrate under hydrothermal conditions in the presence of a wide variety of transition metal oxide and sulfide minerals.

Cross Team Collaborations

Tasks 1 and 2

In the course of this project, we have held extensive discussions and have initiated collaborations with Kenneth Nealson and Gene Macdonald (JPL Team) and Robert Minard (Pennsylvania State Team).

NASA Mission Involvement

Tasks 1 and 2

Procedures and experimentation developed at CIW for “direct” monitoring of biochemical activity at extreme conditions can form the basis of testing hypotheses for the feasibility of life in planetary environments. This work will help in constraining mission parameters for the search for life on other planetary bodies and can be used as testing ground in planning a mission aimed at finding “habitable zones” in planetary bodies. Data from these experiments will benefit planned missions to Mars and Europa.

Future Directions

Task 1

Over the next year we will continue exploring mineral-catalyzed reactions under hydrothermal conditions. In the course of our experiments we have observed that under some conditions the partial oxidation of saturated acids, e.g., methylsuccinic acid, to thiols is promoted in the presence of NiS. This is a remarkable result. Saturated compounds are notoriously stable, i.e., unreactive except under extremely oxidizing conditions. In extant biochemistry, considerable enzymatic hardware has been developed to handle critical RedOx reactions, e.g., the dehydrogenation of succinate to form fumarate. In order to establish that chemistry similar to that of anabolic metabolism arose naturally in the vicinity of hydrothermal vents, it is critical for us to derive the non-enzymatic pathway for alkane oxidation to olefins. We will be exploring a broad range of reactions centered on this important question. We will also begin to explore the role of tungsten and molybdenum sulfides. It may be significant that some extreme hyperthermophiles, such as *Pyrococcus furiousus*, require tungsten within the active center of the aldehyde oxidoreductase enzyme. The multiple valence states available to both W and Mo suggests that sulfide minerals containing these elements may provide ideal oxidants for the partial oxidation of saturated compounds.

In addition to these studies, now that we have refined our focus to a few key reactions, we will begin to utilize our high T and P flow reactor to begin to test the viability of promoting multiple reactions under conditions that mimic those of hydrothermal vents. One goal is to demonstrate the continuous synthesis of citrate and pyruvate from an initial solution saturated with propene and/or propane thiol. There are considerable analytical challenges associated with this goal.

Task 2

The goal for the forthcoming year is to expand further the study of viability of life at extreme conditions using the techniques developed for “direct” observations. This project will include studies simulating various Earth and other planetary subsurface environments for testing the viability of life in these extreme conditions. Our study is designed to facilitate experimental testing of hypotheses on the feasibility of life on Mars, Europa, Titan, and other planetary bodies. Combining such experiments with protein and amino acid analysis, further controls will be obtained to specify the factors and processes involved in adaptation of life at what are considered very extreme conditions for the viability of life.

Task 3

During the next year we will further characterize the suite of amphiphilic molecules produced in our synthesis experiments. Gas chromatography, mass spectrometry, UV spectroscopy, and laser-desorption time-of-flight mass spectrometry will be used for these analyses. We will then compare in more detail the synthetic products with those extracted from the Murchison meteorite.

Task 4

- The three essential features of the organic chemistry that led to life are routes that (i) include formation of C-C bonds, (ii) include formation of C-N bonds, and (iii) provide complexity (branching and polyfunctionality). With these tenets in mind we are devising studies to probe the vital question of whether life's origins were endogenous, or whether the critical, primitive organic compounds were introduced to the Earth through external sources.

- We have shown that, given a CO_2/N_2 primitive atmosphere, the well known formaldehyde/HCN route to amino acids is not feasible, and that ammonium and formate were the precursor candidates in the early oceans. A sensible route to the essential complex organic compounds is then Fischer-Tropsch Synthesis (FTS), which provides the first two crucial components, but not complexity.

- We submit, however, that the high temperatures necessary to bring the reaction to convenient rates in conventional commercial operation may mask the nature of the initial products through a subsequent overlay of stripping reactions. A probe into the initial products can be developed employing pressure in place of temperature to boost reaction rates, and accordingly we expect to conduct high-pressure studies of aqueous formate chemistry in the coming year. The work will be conducted in collaboration with colleagues at CIW employing high-pressure facilities both there and at the USGS in Menlo Park.

Task 5

Studies of the reduction of nitrate to ammonia in the presence of minerals will be expanded to investigate the synthesis and stability of amino acids under hydrothermal conditions, at various pH, both with and without minerals.

Roadmap Objectives

Project

Studies in Planetary Formation and Evolution

Senior Project Investigator(s):
S. Solomon, A. Boss, R. Butler, G. Wetherill

#8
Past and Present Life on Mars

#9
Life's Precursors & Habitats in the Outer Solar System

#11
Origin of Habitable Planets

#12
Effects of Climate & Geology on Habitability

Accomplishments

Task 1. Detection and Characterization of Extrasolar Planets

Our group is surveying the nearest 1,200 Sun-like stars (of spectral type F8-M5) with the precision Doppler technique at the Lick 3-m, Keck 10-m, and Anglo-Australian 3.9-m telescopes to search for extra solar planets. Over the past 6 years, these three surveys have led to the discovery of two-thirds of the known extrasolar planets. All 12 extrasolar planet discoveries published in peer-reviewed journals over the last year were found from these three surveys, including two new multiple planet systems. All of our Doppler surveys have achieved state of the art velocity precision of 3 m/s. These are the only active surveys sufficient to detect Solar System type planets (with masses similar to Jupiter and Saturn).

Beginning in early 2002 we will expand our program with the Magellan 6.5-m telescope at Las Campanas in Chile. By adding 800 stars with this telescope, we will complete a volume-limited survey of G dwarfs (solar type stars) out to 50 parsecs, K dwarfs out to 30 parsecs, and M dwarfs out to 10 parsecs. This first reconnaissance of all nearby dwarf stars will provide the target list for more intensive follow-up observations from ground and space-based interferometers, transit observations, reflected light searches, transit photometry, and ultimately direct imaging and spectroscopy with the NASA Terrestrial Planet Finder.

Task 2. Formation of Gas-Giant Planets

Two theories are competing to explain the formation of the gas-giant planets discovered in orbit around nearby stars. Core accretion, developed to explain our Solar System and currently the generally accepted mechanism, requires several million years or more to form a gas-giant planet in a protoplanetary disk. However, recent observations have shown that in most cases, protoplanetary disks lose their gas in several million years or less, implying that if core accretion is the only means of forming gas-giant planets, these planets should be rare. The ongoing census of extrasolar gas-giant planets strongly suggests otherwise. The alternative mechanism, disk instability, can form a gas-giant protoplanet rapidly, in about a thousand years. Last year we used three-dimensional hydrodynamical models to show that disk instability could form Jupiter-mass clumps even in a disk with a mass comparable to that inferred for the solar nebula and similar to that needed to form gas-giant planets by the core accretion mechanism. This year we have extended these models to include a complete thermodynamical description of the disk instability process, including a solution of the three-dimensional energy equation with radiative transfer in the diffusion approximation, and with detailed equations of state and dust grain opacities. Because the disk cooling times are comparable to the local orbital period, disk instabilities proceed in much the same manner as in the previous "locally isothermal'' models, though rising temperatures in the clumps do restrict their growth. The models imply that the disk instability mechanism could obviate the core accretion mechanism, in the solar nebula and elsewhere, possibly with important consequences for the formation of terrestrial planets in these systems.

Task 3. Formation of Earth-like Planets

A fundamental goal of astrobiological research is to understand the extent to which planets favorable to life are frequent or rare. Although it may be that chemical and physical processes different from those on Earth may provide habitable planets, Earth is today the only planet we know of that contains a living system of any kind. Therefore, understanding the frequency of Earth-like planets is an essential goal for this Institute. Our planetary system provides a unique body of firm facts concerning a system in which not only life, but cognitive life actually did occur. Furthermore, we have abundant knowledge of the present physical state of our Solar System. For example, we know for an absolute fact that our gas giant planets did not drift near the Sun, despite the observational data that clearly shows that this does occur in other planetary systems. At present, available instrumentation does not permit observation of Jupiter-size planets at the distance of Jupiter from their stars, but the time will soon come when this is possible. Before that time, it is essential that a body of theoretical work exists that can be compared with new observational data in order to provide the necessary balance between theory and observation. We are trying to contribute to this need by devising a model that can include both the observation of our Solar System and the presently observed extrasolar planets.

At this time most workers make use of a "standard model" for the formation of our Solar System, at least out to the distance of Saturn. According to this model, in the terrestrial planet region, planetesimals grow from ~10^{14}g bodies to ~10^{26}g bodies in about 10^5 years by a characteristic instability termed "runaway growth." This has the consequence that Mars-size "planetary embryos" form at ~1 AU in about 10^5 years and grow to Earth size in about 10^8 years even in the presence of giant planets that form after only ~10^7 years, according to this standard model. The problem is that if the limited observational data of extrasolar planets is typical of planetary systems in general, then the incoming giant plants should drift into the central star and also disturb the terrestrial planets.

As discussed last year, we developed a quantitative alternative to the standard model that is based on a new theory for giant planet formation. In this model, Jupiter grows on a very short timescale (~10^5 years). This work shows that in the presence of early-formed gas giant planets and nebular gas, Mars-size planetary embryos in the terrestrial planet region are formed despite the very strong gravitational perturbations by the giant planets. This is the consequence of synchronism in the orbital elements of the planetesimals, permitting sufficiently low relative velocities despite strong perturbations by the giant planets.

To a first approximation, this model also predicts that the asteroid belt will be very strongly depleted, as it actually is. However, in the extreme case of Jupiter growth before formation of any planetesimals at all, this goes too far, and growth of the asteroids approaching the size of those in the present asteroid belt will be precluded. However, disk evolution is not sufficiently well understood to rule out the likelihood of a delay of ~10^5 years in the development of the gravitational disk instabilities that led to the formation of Jupiter and Saturn. If this delay occurs, an asteroid belt with its largest bodies the size of Ceres can be formed. The detailed orbital and fragmentation evolution of such a system remains to be studied.

In a separate project, we have studied the formation of Jupiter and Saturn in the framework of the standard model, but including the effects of fragmentation and loss of planetesimals by the high-velocity perturbations caused by growth of the massive giant planet cores. Fragmentation reduces the quantity of material available to form a Jupiter core considerably. As a consequence, the usual choice of disk grain opacity makes it unlikely that the gravitational instability necessary to capture the gas mantle of Jupiter can form. However, if the grain opacity is reduced by a factor of ~100, a Jupiter mass planet can form within ~10^7 years. While not out of the question, recent work supports lower timescales than this for the formation of Sun-like stars. In addition, there remains the major problem of a planet in a gaseous environment avoiding drift into the inner Solar System on a timescale as long as 10^7 years. For these reasons, we believe that the alternative to the conventional model of planet formation should be considered seriously.

Task 4. Evolution of Planetary Water

One of the long-term goals of this astrobiology project is to assess the likelihood, timing, and physical and chemical environments of hydrothermal systems on Solar System objects other than Earth. Our efforts on this project during the past year have focused on Mars, primarily because of the influx of important new data from the Mars Global Surveyor (MGS) mission. The Tharsis region of Mars has long been recognized as a major center of magmatism and deformation that has been active throughout martian history as a source of heat and atmospheric volatiles. Recent work on defining the stages of deformation in the region has demonstrated that much of the tectonic activity was concentrated in the Noachian epoch of martian history, the period prior to the end of heavy bombardment of the inner solar system about 3.8 Ga ago. On the basis of the global topography and gravity anomaly fields determined by MGS, we have shown that much of the long-wavelength aspects of those fields can be explained as the response of the martian interior to loading by the volcanic materials of the Tharsis rise. Of particular importance to the history of martian water, we have also shown that the downhill directions of late Noachian valley networks on Mars appear to have been influenced by the long-wavelength slopes produced by this global response to Tharsis loading. There are two implications of this result. The first is that the Tharsis rise must have been substantially in place prior to the late Noachian, a conclusion indicating that the martian volcanic flux early in the planet's history was much higher than during any subsequent period. The second implication is that volatiles released from magmas erupted during the construction of Tharsis may have contributed substantially to the inventories of water and other species in the atmosphere-surface system, perhaps modifying climate and influencing the formation of valley networks.

Highlights

In the past year, members of our team

- Discovered 12 new extrasolar planets, including the second and third multiple-planet systems.

- Demonstrated that Jupiter-mass planets can form from instabilities in a protoplanetary disk of a mass similar to the solar nebula.

- Demonstrated that Mars-size planetary embryos can form in the inner solar system even in the presence of rapidly formed outer planets.

- Showed that the Tharsis rise on Mars was largely emplaced prior to the formation of late Noachian valley networks, and that the volcanic construction of Tharsis may have contributed significantly to the atmospheric inventory of water and other volatiles.

Field Trips

Observing schedules for R. Paul Butler and colleagues:

Date/Observatory/Observers

Jul 14	Keck	Marcy, Butler, Vogt, Noyes, Brown, Charbonneau
Jul 15-17	AAT	Tinney
Jul 28-31	Keck	Marcy, Butler
Aug 10-13	AAT	Tinney
Sep 4-9	Keck	Marcy, Butler
Oct 9-12	AAT	Tinney
Nov 1-3	Keck	Marcy, Butler, Vogt
Nov 7-10	AAT	Tinney
Dec 3-4	Keck	Marcy, Butler
Dec 18-21	Keck	Marcy, Butler
Jan 9-11	AAT	CGT
Mar 2-5	Keck	Marcy, Butler
Mar 12-14	Keck	Marcy, Butler
Mar 14-15	AAT	Jones
Apr 2-3	Keck	Marcy, Butler
Apr 7-8	Keck	Marcy, Butler
Apr 9-10	AAT	Penny
Apr 30	Keck	Vogt
May 3-8	AAT	Tinney, McCarthy

May 4-7	VLT2	Bedding, Butler
May 30-Jun 1	Keck	Marcy, Butler
Jun 3-4	Keck	Marcy, Butler

Cross Team Collaborations

Co-I Huntress (CIW) is a Collaborator on the newly selected JPL 2 Team. He is leading a multi-institutional group proposing a Discovery mission to Venus.

Co-I Butler (CIW) recently recruited a postdoctoral fellow (Chris McCarthy) from the UCLA Team to assist in planet-search observations.

CIW has also recently hired Alycia Weinberger (now on the UCLA Team) to our research staff. An infrared astronomer, Weinberger studies accretion disks and planet formation processes around young stars. She will join the CIW Team as a Co-Investigator in September.

PI Solomon (CIW) is collaborating with Bruce Jakosky (PI of the University of Colorado, Boulder Team) on the synthesis of the sequence of major planetary events in the history of Mars.

NASA Mission Involvement

Co-I Boss is a Co-I on NASA's approved FAME (Full-Sky Astrometric Mapping Explorer) mission, which will detect very low mass companions, including planets, around nearby stars. He is also on the Science Team for the Kepler mission, currently one of three missions being considered for final approval under NASA's Discovery Program. Kepler's photometric observations will permit the detection of numerous Earth-like planets in Earth-like orbits around stars in the disk of our Galaxy.

Co-I Butler states: "Our goal is to survey all nearby stars out to 50 parsecs in preparation for follow up by NASA missions including the Keck Interferometer, the Space Interferometry Mission, and the Terrestrial Planet Finder."

PI Solomon is a Co-Investigator on the Mars Orbiter Laser Altimeter (MOLA) experiment on Mars Global Surveyor (MGS) and the Principal Investigator on the MESSENGER (Mercury: Surface, Space Environment, Geochemistry, and Ranging) mission.

Future Directions

Co-I Boss will continue to calculate detailed three-dimensional hydrodynamic and radiative transfer models of the formation of gas giant planets by the disk instability mechanism. In particular, growing clumps will be modeled as virtual protoplanets (point masses undergoing Bondi accretion), allowing their orbital evolution to be followed much farther in time.

Co-I Butler will:

- Continue surveys of 1,200 nearby stars with Lick 3-m, Keck 10-m, and Anglo-Australian 3.9-m telescopes..

- Begin 900-star survey with Magellan 6.5-m telescope.
- Improve Doppler measurement precision from 3 m/s to 2 m/s.

PI Solomon plans a synthesis of the relative timing of important geological events in the early history of Mars, with a focus on implications for the history of surface and subsurface water.

Project

Studies of Organic Matter and Water in Meteorites

Senior Project Investigator(s):
C. Alexander, N. Boctor, G. Cody, E. Hauri

Roadmap Objectives

#1
Sources of Organics on Earth

#8
Past and Present Life on Mars

#9
Life's Precursors & Habitats in the Outer Solar System

#11
Origin of Habitable Planets

Accomplishments

A goal of astrobiology is to determine how common habitable planets are around other stars, a question whose answering requires a better understanding of planetary formation. We literally hold in our hands the clues to understanding this process, within meteorites. Chondrules, millimeter-sized spheres of melted rock in meteorites, carry a record of the conditions that prevailed in the Solar System during its earliest stages. That record has been undeciphered for lack of a compelling theory of what melted chondrules.

The organic material in primitive chondritic meteorites has also attracted considerable attention, not only because it retains a record of synthesis in the interstellar medium (ISM), but also because L-enantiomer excesses have been reported in meteoritic amino acids. If the meteorite organics are typical of the material accreted by the prebiotic Earth, these excesses may explain the homochirality of terrestrial life. The ISM origin of some or all the meteorite organics suggests the intriguing possibility that this or similar material is a source of complex prebiotic organics not just in our Solar System but in all solar systems. The amino acids, nucleic acids, and other soluble organics found in the meteorites probably formed by hydrolysis of the more abundant macromolecular material, whose structure and origin remains enigmatic. We are currently using a range of techniques to determine its structure. From this we hope to learn how it formed and how it would break down under various conditions to produce important, complex prebiotic molecules.

The martian meteorites are our only sample of another planet (not counting the Moon and asteroids). Early conditions on Mars may well have been conducive to the development of life. However, the surface of Mars is now an arid and inhospitable environment for life. The key to understanding how long conditions were conducive to life and whether life might still persist at depth on Mars is the evolution of water. Martian meteorites do contain water-bearing phases. The water in these phases is typically enriched in deuterium. Attempts have been made to gain insight into the water contents of the martian mantle using kaersutite, a TiO_2-rich amphibole, found in 7 of the well-studied martian meteorites. However, to date deriving the mantle water contents from the kaersutites has been frustrated by the uncertain origin of their low measured water contents. The current martian atmosphere is also enriched in deuterium as a result of the loss of water to space. The deuterium enrichments in the water-bearing minerals in the martian meteorites suggests that they contain water that at some time interacted with water from the martian atmosphere. If the deuterium-rich water in the oldest martian meteorite (ALH84001 - 4.0 Ga) reflects the composition of the ancient martian atmosphere, Mars had lost most of its water very early, leaving little time for life to evolve. However, there are processes associated with the intense shock most martian meteorites have experienced that may have produced the deuterium enrichments. We are trying to determine which of the two possible explanations for the deuterium enrichment is the correct one.

Task 1. Physics and Chemistry in the Early Solar Nebula

We have quantified the predictions of one leading theory for chondrule formation, melting by shock waves in the nebula gas. A numerical code models the thermal histories of chondrules passing through such shocks, including for the first time the transfer of radiation. It was discovered that chondrule-forming regions are optically thin, and that chondrules cool faster in chondrule-rich environments, the opposite of conventional wisdom. Also, it was shown that the gas, heated and compressed by the shock, keeps the chondrules warm for the hours inferred from their petrological textures. We were able to model the thermal histories of chondrules in great detail, lending great support to the shock wave model. A context for interpreting the chondrules record is anticipated. We have also been collaborating with other astrobiologists to determine the chemical effect of shock waves in the inner solar nebula, especially on interstellar nitrogen compounds. Shocks may be instrumental in breaking N_2 bonds to form CN bonds. These nitriles can then polymerize or participate in chemistry such as Strecker synthesis to form amino acids.

Task 2. Macromolecular Organic Matter in Carbonaceous Chondrites

We have completed a suite of complementary, double- and single-resonance solid state (^{1}H and ^{13}C) Nuclear Magnetic Resonance (NMR) experiments on a solvent extracted and demineralized sample of the Murchison meteorite organic macromolecule. These NMR data provide a consistent picture of a complex organic solid composed of a wide range of organic (aromatic and aliphatic) functional groups, including numerous oxygen-containing functional groups. The fraction of aromatic carbon (F_a) within the Murchison organic residue (constrained by three independent experiments) lies between 0.61 and 0.66. The close similarity in cross-polarized and single-pulse spectra suggests that both methods detect the same distribution of carbon. With the exception of interstellar diamond there is no evidence in the solid state NMR data for a significant abundance of large, laterally condensed aromatic molecules in the Murchison organic insoluble residue. Given the most optimistic estimation, such carbon would not exceed 10% and more likely is a fraction of this maximum estimate. The fraction of aromatic carbon directly bonded to hydrogen is low (~ 30 %), indicating that the aromatic molecules in the Murchison organic residue are highly substituted. The bulk hydrogen content, H/C, derived from NMR data, ranges from a low of 0.53 ± 0.06 to a high of 0.63 ± 0.06. The hydrogen content (H/C) determined via elemental analysis is 0.53. The range of oxygen-containing organic functionality in the Murchison is substantial. Depending on whether various oxygen-containing organic functional groups exist as free acids and hydroxyl or linked as esters and ethers results in a huge range in the estimated O/C, 0.22-0.37. The lowest values are more consistent with elemental analyses, requiring that oxygen-containing functional groups in the Murchison macromolecule are highly linked. The combined ^{1}H and ^{13}C NMR data reveal a high proportion of methine carbon requiring that carbon chains within the Murchison organic macromolecule are highly branched.

Task 3. Sources of Water for the Terrestrial Planets

Because of relatively high temperatures in the terrestrial planetary region of the solar nebula, it is thought that little water would have been incorporated into planetesimals that formed there. Another source of water is needed. At present the most likely sources seem to be water-bearing asteroids and icy bodies that formed in the region of Jupiter. The location of the snowline in the asteroid belt, the boundary between regions with or without ice condensation, has an important bearing on the availability of water-bearing objects to the forming terrestrial planets — the closer this snowline was to the Sun, the greater the availability. The most common primitive meteorites are the ordinary chondrites (OCs), which probably come from the inner part of the asteroid belt. Evidence for limited aqueous alteration has been found only in a few highly unequilibrated ordinary chondrites that are thought to have formed near the surface of their parent asteroids. This could be interpreted to mean that condensation of ice occurred only at the end of the formation of their parent asteroids, in which case these asteroids are not likely to be important sources of water. However, as part of a broader collaborative study of primitive chondrites, we have recently shown that aqueous alteration probably occurred throughout the OC parent bodies but has been largely obscured by subsequent heating (metamorphism). Thus it seems likely that the snowline was slightly inward of the location where the OC parent bodies formed, which is significantly closer to the Sun than is generally assumed.

Task 4. Sources of Extraterrestrial Water in Martian Meteorites

We continued our investigation of H isotope composition and the sources of water in martian meteorites. Last year we investigated four more martian meteorites, bringing the number of we have studied to date to 9. The meteorites we recently investigated are Shergotty, Zagami, Dar al Gani (DaG) 476, and Sayh al Uhaymir (SaU) 005. Shergotty and Zagami, being observed falls, may not have been as affected by terrestrial contamination as martian meteorite finds. DaG 476 and SaU 005 may possibly be less affected by weathering in the dry environments of the Sahara and Oman, where they were found, than Antarctic meteorites.

A significant result of our investigation is the discovery of a second occurrence of a post-stishovite high pressure form of silica in Zagami. This phase has been previously described in Shergotty. We discovered that this silica phase in both meteorites contains a hydrogen component with an extraterrestrial isotope signature. High δD values (3728 to 1960‰) were measured for the high-pressure silica in Zagami compared to 1975 to 1246‰ for the same phase in Shergotty. Impact melted feldspathic glass also is D enriched (δD 2532 to 579‰ for Zagami and 2239 to 687‰ for Shergotty.)

In SaU 005 and DaG 476, the only water-bearing phase was feldspathic glass formed by impact melting. The δD values for feldspathic glass (2391 to 743‰) in SaU 005 are on average higher than those of the same phase (2347 to 352‰) in DaG 476. The wide range in the δD values of feldspathic glass in both meteorites suggests that they show some contamination by terrestrial water, even though the meteorites resided in an arid environment before they were found.

The D enrichment in the feldspathic glass in the four meteorites may be attributed to incorporation of fractionated martian surface or underground water into their parent impact melts. The fractionation was possibly enhanced by devolatilization of hydrogen by impact. There are two possible sources of water in the post-stishovite silica phase. The precursor silica phase prior to shock was nominally anhydrous, containing trace amounts of water as hydrogen defects; D enrichment occurred when the precursor or its post-stishovite high-pressure phase exchanged H with a fractionated martian water reservoir. Alternatively, fractionated water was incorporated in the post-stishovite silica as it nucleated at high pressure during the impact event. The H isotope data are consistent with mixing of a martian high D component with a low D magmatic component or a terrestrial contaminant.

Task 5. Petrological Studies of Water in Martian Magmas

We are investigating the magmatic crystallization of kaersutite from a martian basalt in an effort to gain insight into the origin and low water contents of the kaersutites (martian meteorites). Ultimately, understanding the amount of magmatic water necessary for martian kaersutite formation provides insight into the history of water on Mars. The low measured water (and halogen) contents in the kaersutites might result from oxy-substitutions, which are potentially consistent with the Fe^{3+}/Fe^{2+} ratios and TiO_2 contents of the kaersutites [up to ~11 wt% TiO_2]. Other studies suggest that the low water contents were imprinted on the kaersutites after formation by processes such as dehydrogenation during ascent or impact shock devolatilization. Further complication arises from the fact that high-TiO_2 kaersutite has never been crystallized from a melt in the laboratory.

Experiments conducted on a water-bearing martian basalt in a piston cylinder apparatus suggest that high crystallization temperature and reducing oxygen fugacity encourage crystallization of high-TiO_2 amphiboles. Maintaining such conditions is experimentally challenging. The reducing nature of the experiments encourages water loss from starting material, which in turn leads to oxidation of the starting material. Additionally, the FeO-rich nature of martian basalts makes Fe loss to experimental capsules a particular concern. The experiments thus far have focused on identifying the best techniques to eliminate or minimize Fe loss, water loss, and oxidation of the experimental products. Saturating experiment capsules with the starting material prior to the experiment has proven successful in circumventing Fe loss difficulties. Progress has also been made in preventing the loss of water and the oxidation of the sample utilizing a double capsule technique, which reduces the gradient for H out of the starting material.

Task 6. Iron Isotope Measurements of Terrestrial Rocks and Meteorites

The identification of naturally occurring isotopic mass fractionation of the transition metals in chemical sediments has been cited as evidence for microbial utilization. These studies have prompted a search for similar variability in terrestrial rocks and meteorites in order to document both the baseline of abiotic isotope variability and to search for "isotopic biomarkers." Our work on Fe isotopes has been facilitated by recent advances in inductively coupled plasma mass spectrometry (ICP-MS). Iron isotopes are extracted from acid-dissolved residues of bulk meteorites and chondrules using anion exchange resin and measured using a VG Axiom ICP-MS. All of the bulk meteorite compositions, which include both chondrites and iron meteorites, are identical to the terrestrial basalt composition, confirming that planetary differentiation and core formation did not significantly mass fractionate Fe isotopes. The chondrules, on the other hand, all tend toward lighter compositions. This could reflect formation from an isotopically light starting material or under non-equilibrium conditions, which can enrich the light isotopes

Highlights

- The shock wave model for chondrule formation is beginning to receive renewed support as a result of Desch's contributed talk at the Lunar and Planetary Science Conference in March. This comes at a time when a crisis in chondrule formation had prompted many other researchers to seek less prosaic models such as solar flares.

- The stringent constraints provided by our suite of complementary NMR (nuclear magnetic resonance) experiments have brought us much closer to being able to construct a statistical model of the Murchison organic macromolecule.

- Our discovery of deuterium-rich structural water in the new high-pressure polymorph of silica provides additional evidence linking deuterium-rich water in martian meteorites and shock processes during impact cratering.

- The water-ice condensation line in the Solar Nebula seems to have been sunward of the ordinary chondrite formation region, which is closer to the Sun than is generally assumed.

• Improved ICP-MS (inductively coupled plasma source mass spectrometer) techniques have resulted in a precise method for measuring Fe isotopes in a wide variety of terrestrial and meteorite samples.

Cross Team Collaborations

The research described above under Tasks 4 and 5 constitutes CIW's connection to the new NAI Astromaterials Focus Group (formerly Martian Meteorites Focus Group), led by David McKay, PI of the JSC Team.

The research described above under Task 2 constitutes CIW's connection to a new effort to develop an "Extraterrestrial Macromolecular Characterization Focus Group." This Focus Group involves core members from CIW Team, Robert Minard of the Pennsylvania State Team, and Gene MacDonald of the JPL Team, as well as a number of other NAI members.

NASA Mission Involvement

• The study of martian meteorites will help guide the questions to be addressed and the types of samples to be returned by future missions to Mars.

• The Murchison macromolecule is probably the best available analogue for the 'refractory' organic matter in: (a) the interstellar grains to be collected by the STARDUST mission (collection of cometary material and interstellar dust samples) and (b) materials that will be encountered by future missions to comets. Examples of these missions are: CONTOUR (Comet Nucleus Tour); Deep Impact (comet impact to study the nature of inner cometary material as well as the crust); and Rosetta (a comet rendezvous for remote sensing investigations and carrying a probe to land on the comet's surface to perform *in situ* measurements.)

Future Directions

• We will continue to develop the shock wave model for chondrule formation, to show that it is consistent with other aspects such as the correlation between frequency of compound chondrules and chondrule texture, and the remnant magnetizations of chondrules and chondrites. Through parameter studies we also plan to determine the unique gas density, temperature, etc., in the formation regions of chondrules from the separate chondrite classes.

• We will be constructing models of the structure of the Murchison macromolecule. We will also extend our NMR studies to other primitive meteorites, such as Tagish Lake, Orgueil, and Allende.

• We will continue our survey of the H isotopic compositions of water-bearing phases in martian meteorites. In particular, we will concentrate on the analysis of melt inclusions. These inclusions are the most likely components of the meteorites to have retained martian mantle water. The isotopic composition of this mantle water can be used to constrain the total water budget of Mars.

- We will complete a survey of Fe isotope variations in iron meteorites, chondrites, chondrules, and calcium-aluminum-rich inclusions (CAIs), and we will begin a study of modern and ancient seafloor sediments and bacterial mats. In collaboration with Co-I David Emerson, we will also begin Fe isotope measurements of biogenic minerals from specific bacterial cultures and begin a series of Fe isotope fractionation experiments focusing on oxidation and reaction of Fe metal in aqueous solutions.

Theoretical Studies of Hydrothermal Synthesis Reactions

Project

Senior Project Investigator(s):
E. Shock and H. Morowitz

Roadmap Objectives

#1 Sources of Organics on Earth

#2 Origin of Life's Cellular Components

#3 Models for Life

#5 Linking Planetary & Biological Evolution

#7 Extremes of Life

#8 Past and Present Life on Mars

#9 Life's Precursors & Habitats in the Outer Solar System

Accomplishments

Task 1. Energetics of Hydrothermal Ecosystems

We have continued to apply thermodynamic calculations toward establishing the viability of both biological and abiotic reactions relevant to astrobiological issues. For example, we recently focused on determining the energetics of overall metabolic reactions of both thermophilic and hyperthermophilic prokaryotes. While we have determined the ΔG_R^o for critical metabolic reactions such as reduction-oxidation, hydrolysis, and disproportionation, we focused on the Knall-gas reaction, anaerobic sulfur and nitrate reduction, and autotrophic methanogenesis. We determined the overall Gibbs free energy as a function of temperature for a wide range of chemical compositions representative of near-surface and deep hydrothermal systems. To date we have calculated the values of ΔG_R^o as functions of temperature for 188 established metabolic redox reactions plus an additional 182 reactions that chemically link metabolic processes to the composition of natural waters. It is anticipated that these data will be useful in the design and optimization of culture media, the quantization of microbial energetics, and the placement of hyperthermophilic and thermophilic prokaryotes in the context of their geochemical and ecological environments.

We have also applied thermodynamic calculations to the analysis of the potential for the abiotic synthesis of aliphatic and aromatic hydrocarbons detected in martian meteorites, e.g., ALH84001. Our calculations show that polycyclic aromatic hydrocarbons (PAHs) and normal alkanes could form metastably from CO, CO_2, and H_2 below approximately 250-300°C during rapid cooling of trapped magmatic or impact-generated gases. Depending on the temperature, bulk composition, and oxidation-reduction condition, PAHs can form at low bulk H/C ratios, higher CO/CO_2 ratios, and higher temperatures than normal alkanes. We conclude that it is possible that all of the hydrocarbons detected within ALH84001 could have formed abiotically via a reaction of iron carbonate liberating CO during a thermal conversion to magnetite.

Task 2. Theoretical Foundation for Organic Synthesis Experiments

We have been seeking to explain the ubiquity and origins of intermediary metabolism. We note that the core of intermediary metabolism in autotrophic organisms lies the citric acid cycle. In a certain group of chemoautotrophs, the reductive citric acid cycle is an engine of synthesis, taking in CO_2 and synthesizing the molecules of the cycle. We have examined the chemistry of a model system of C, H, and O that starts with carbon dioxide and reductants and uses redox couples as the energy source. To inquire into the reaction networks that might emerge, we start with the largest available database of organic molecules, Beilstein on-line, and prune by a set of physical and chemical constraints applicable to the model system. From the 3.5 million entries in Beilstein we emerge with 153 molecules that contain all 11 members of the reductive citrate cycle.

A small number of selection rules generates a very constrained subset, suggesting that this is the type of reaction model that will prove useful in the study of biogenesis. The model indicates that the metabolism shown in the universal chart of pathways may be central to the origin of life, is emergent from organic chemistry, and may be unique.

Highlights

- We have established a global thermodynamic framework for metabolism in thermophilic and hyperthermophilic prokaryotes. These data will allow us to explore the theoretical range of life under extreme conditions.

- We have shown that it is likely that the hydrocarbons detected in the ALH84001 meteorite were formed abiotically through reactions of CO and H_2 in the presence of magnetite.

- We have shown that the citric acid cycle may be derived from physical and chemical selection rules, suggesting an emergent quality of the chemistry distinct to its environment. We conclude that the citric acid cycle may be inevitable to any emergent biochemistry.

Cross Team Collaborations

In the course of this work, we have held collaborative discussions with the abiotic organic synthesis group on the Arizona State University Team, as well as the abiotic organic synthesis project group at SUNY Stony Brook (part of the Pennsylvania State Team).

NASA Mission Involvement

The theoretical components in our project Tasks 1 and 2 provide fundamental information that will serve to guide missions to both Mars and Europa.

Future Directions

Task 1

We will continue to refine our thermodynamic analysis of extant metabolic chemistry and abiotic organic synthesis. We are applying our approach to experimental studies of hydrothermal systems whereby we can constrain the chemistry of natural fluids and derive accurate overall free energies of global systems, to establish the theoretical limits of life constrained via robust thermodynamic calculations.

Task 2

The next step in our analysis of the emergent biochemistry is to include the nitrogen. In particular we are interested in the key amination reactions that bifurcate off of the reductive citrate cycle. We postulate that initially reversible amination reactions may have served as a viable storage device for otherwise reactive alpha-ketoacids. Towards this end we must established how pyridoxal phosphate (PDP) emerged, as it is exclusively through PDP that transamination reactions occur, imparting the critical reversibility necessary for primordial biochemistry.

Education & Public Outreach

Interactive Educational Book/ Astrobiology: The Search for Water

An interactive educational book on astrobiology is aimed at middle and high school students. Volume 1 of this book is entitled, “Astrobiology: The Search for Water”. The print materials survey content in a substantial, yet conversational manner. The booklet has a unique and interactive dimension. Throughout the text there are embedded bar codes that can be scanned using the new CueCat technology developed by Digital Convergence. When an embedded bar code is scanned, it takes the reader/student/teacher to a web page with original source articles, more information, and perhaps an activity about the topic. These associated web pages in effect act like "filing cabinets" where the latest information can be stored about the topics found in the print material. For a rapidly changing field, this format allows us to print materials that will have a long and useful shelf life in the classroom. Booklets and bar-code readers are made available to classrooms free of charge upon request. Volume 1 is currently ready for distribution. Future titles in the series include Astrobiology: Discovering New Life at the Bottom of the Ocean and Astrobiology: Microbes.

Summer Intern Program

The Carnegie Summer Intern Program in Geoscience brings 12 - 15 college students, competitively selected, to the Carnegie campus every summer for an intensive 10-week experience in original scientific research. At a Symposium held at the end of the program, students report on the results of their research. Several interns have been lead authors of related articles later published in science journals.

Public Lectures

The following series of public presentations and lectures about astrobiology were given: Emergence and the Origin of Life - Smithsonian Baird Auditorium, Science, Religion and the Origin of Life - Retirement Institute, Astrobiology - West Virginia Science Teachers Association, Minerals and the Origin of Life - Potomac Geophysical Society.

Astrobiology: Discovering New Worlds of Life

A full color 24” X 36" educational poster about hydrothermal vents targets themes in astrobiology. The front of the poster consists of an original work of art by undersea artist Karen Jacobsen. The reverse side of the poster contains background text about hydrothermal vent life and the importance of these sites to the field of astrobiology. The poster discusses aspect of the research being conducted by Carnegie and other scientists. The poster will accompany a booklet entitled Astrobiology: Discovering New Worlds of Life. Both items will be distributed through NSTA, AAAS, and CIW.

Carnegie Academy for Science Education (CASE)

CASE is a five-week institute for elementary teachers that focuses on science content and pedagogy. Each of the five weeks is centered around a theme in the field of astrobiology. Themes include: microbes, mission designs and goals in astrobiology research, hydrothermal vent geo- and bio-diversity, the characteristics of life, and the potential for life on extra solar planets. Scientists teach and contribute to the work of classroom educators by highlighting scientific discoveries and leading activities.

Cue Cards

Cue Cards are content links to NAI and CIW astrobiology websites. The cards have an interactive dimension. The cards have bar codes that can be scanned using the new CueCat technology developed by Digital Convergence. When an embedded bar code is scanned, it will take the reader/student/teacher to a web page with original source articles, more information and perhaps an activity about the topic. These associated web pages in effect act like "filing cabinets" where the latest information can be stored about the topics found in the print material. For a rapidly changing field, this format allows students to access current materials that will have a long and useful shelf life in the classroom.

Magazine Publication: Life's Rocky Start

Robert Hazen wrote a feature article for the April 2001 volume of Scientific American on the chemical origin of life on earth.

Seminar Series at the Carnegie Institution of Washington

The Department of Terrestrial Magnetism and the Geophysical Laboratory each hold weekly scientific seminars. Speakers include visitors and members of the staff, with topics ranging from astrophysics and planetary sciences to geochemistry and seismology. The seminars are announced to the community with monthly schedules sent to 236 institutions and individuals in the Washington metropolitan area. These were 14 astrobiology related seminars this year.

Spectrum

Spectrum is a quarterly publication of the Carnegie Institution of Washington featuring articles about astrobiology. All articles are simultaneously published on the Institution's website. Titles from this year’s publications were: “Detecting Life Here and There”, “Jump-Starting the Biochemistry of Life?”, “Sniffing Out Proteins on Other Worlds”, “Signatures of Life” and “Butler, Boss and Planets”.

Visiting Investigators

Visiting Investigators are leading scientists from all over the world who conduct basic scientific research while in residence at the Carnegie Institution of Washington. The period of residence can range from several weeks to a year or more. While in residence, they are fully-integrated members of the Carnegie scientific community - delivering seminars, participating in symposia, and acting as advisors to Summer Research Interns.

Publications

Back to Table of Contents

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Back to Table of Contents

Year 3

Harvard University

NASA Astrobiology Institute

Cambridge, Massachusetts

Harvard University Team Members

Ariel D. Anbar
University of Rochester

Sam Bowring
Massachusetts Institute of Technology

Charles Kevin Boyce
Harvard University

Steve DiBenedetto
Massachusetts Institute of Technology

Katrina Edwards
Woods Hole Oceanographic Institute

Douglas H. Erwin
Smithsonian Institution

Brian Fehlau
Harvard University

David Fernández Remolar
Centro de Astrobiologia

Mick Follows
Massachusetts Institute of Technology

Stephanie Fuentes
Smithsonian Institution

S. Goldman
University of Rochester

John P. Grotzinger
Massachusetts Institute of Technology

Galen P. Halverson
Harvard University

John Hayes
Woods Hole Oceanographic Institute

Kai-Uwe Hinrichs
Woods Hole Oceanographic Institute

Paul F. Hoffman
Harvard University

Heinrich D. Holland
Harvard University

E. (Beth) Holman
University of Rochester

Andrew H. Knoll
Harvard University

Kathleen L. Londry
University of Manitoba

Adam C. Maloof
Harvard University

Charles Marshall
Harvard University

John Marshall
Massachusetts Institute of Technology

Mark W. Martin
Massachusetts Institute of Technology

Marylynn Musgrove
Harvard University

Nora Noffke
Harvard University

Ann Pearson
Woods Hole Oceanographic Institute

M. Polizzotto
University of Rochester

Kira Pratt
Woods Hole Oceanographic Institute

Jahan Ramezani
Massachusetts Institute of Technology

Aaron Reiter
Massachusetts Institute of Technology

Daniel P. Schrag
Harvard University

Stefan Schroeder
Massachusetts Institute of Technology

Alex Sessions
Woods Hole Oceanographic Institute/Indiana University

Yanan Shen
Harvard University

Diane C. Sheridan
Harvard University

Christian Sidor
Smithsonian Institution

Roger E. Summons
Australian Geological Survey Organization

Paradha Suntharalingam
Massachusetts Institute of Technology

Sean P. Sylva
Woods Hole Oceanographic Institute

Elisabeth Valiulis
Smithsonian Institution

Rong Zhang
Massachusetts Institute of Technology

Executive Summary

Accomplishments

The Harvard NAI team was constituted in 1998 as an interactive group of biogeochemists, paleontologists, sedimentary geologists, geochemists, and tectonic geologists assembled with the common goal of understanding the coevolution of life and environments in Earth history. The team originally proposed to focus multidisciplinary research on four critical intervals of planetary change: the early Archean (>3000 million years ago) when life began, the early Paleoproterozoic (2400-2200 Ma) when oxygen began to accumulate in the atmosphere and surface ocean, the terminal Proterozoic and Early Cambrian (750-525 Ma) when animal life radiated, and the Permo-Triassic boundary (251 Ma) when mass extinction removed some 90 percent of Earth's species diversity, permanently altering the course of evolution. Given reduced funding levels in years 1 and 2, the team chose to focus on the latter three intervals. As documented in the accompanying project reports, we have made substantial new contributions to all three areas in funding year 3. At the same time, increased funding and strong interest by colleagues at Harvard and MIT have enabled us to expand both our membership and intellectual purview in year 3. In addition to the three projects funded from the outset (and approached in new ways by our newest co-investigators), we have three new projects underway. John Hayes' research at the interface of microbiology and biogeochemistry has expanded as a result of incremental funding that enabled his group to undertake a substantial collaborative effort with the NAI Team at MBL. In association with the Spanish Center for Astrobiology, Andrew Knoll and Ariel Anbar have initiated research on Neogene iron formations in southern Spain that may illuminate both geobiological research on Earth's ancient iron formations and NASA's Mars lander mission slated for 2003 launch. Charles Marshall has undertaken new research on the molecular basis of evolution.

Research by the Harvard team is interdisciplinary, attracting increasing participation by scientists within the five member institutions (Harvard, MIT, WHOI, Rochester, Smithsonian Institution). We have also been successful in promoting cross-team collaborations — currently, individual research projects are underway with colleagues from the Carnegie, MBL, Ames, and JPL teams, as well as both the Spanish and Australian astrobiology centers. Moreover, our team participates actively in the EvoGenomics Focus Group (C. Marshall) and has taken a leadership position in the Mission to Early Earth Focus Group (A. Anbar). Team members are active in research on novel biosignatures and digital mapping technologies that can be applied to solar system research. Field research is underway in Svalbard, Namibia, Newfoundland, Morocco, northern Australia, and China, with new projects planned for year 4. We also teach actively at four universities, contributing to both the training of new professionals and the education of a broader university community. Other outreach efforts focus on lectures presented by team members in a wide variety of public and professional formats.

Research highlights for the past year include the following:

- Mo isotopes have been shown to be potential paleoredox probes in ancient sedimentary rocks.

- Deep waters in a mid-Proterozoic basin have been shown to be sulfidic, consistent with models suggesting that the modern oxic deep ocean only came to exist near the end of the Proterozoic Eon.

- Newly discovered microfossils show that in 1500 Ma marine basins, eukaryotic algae were most abundant and diverse in coastal environments; they expanded to become ecologically important throughout the oceans only toward the end of the Proterozoic Eon. Scarcity of critical nutrient metals in postulated sulfidic Proterozoic oceans may help to explain the observed ecological and stratigraphic distribution of eukaryotic algae in Proterozoic sedimentary rocks.

- Namacalathus, one of the earliest known biomineralizing animals, was reconstructed digitally using high-resolution serial sectioning. This permitted accurate assessment of the diversity, taxonomic relationships, and functional morphology of early skeleton-forming animals. Namacalathus and Cloudina were also discovered below and above a significant carbon-isotope excursion in Oman, interpreted as the Precambrian-Cambrian boundary biogeochemical event. This discovery indicates that these organisms did not go extinct at the boundary.

- A new model, based on carbon isotopic data, has been developed to explain the onset of Neoproterozoic Snowball Earth events.

- Elucidation of ecological and taphonomic processes that govern the formation and preservation of microbial mat signatures in ancient siliciclastic rocks provides new insights on a biosignature that is potentially applicable to Mars rover exploration.

- Combined field research in Newfoundland and laboratory analyses place new geochronological constraints on the timing and duration of late Neoproterozoic glaciation and its relationship to Ediacaran animal diversification.

- Field research in China and subsequent laboratory geochronology demonstrated that the end-Permian mass extinction was catastrophic, occurring in much less than 500 Ky.

- Oceanographic modeling has been used to investigate proposed mechanisms for the end-Permian mass extinction. New models clarify the conditions required to support a haline mode of circulation with sinking of brackish waters from the subtropics: haline modes are favored in climates that are warm, with weak pole-equator temperature gradients, enhanced hydrological cycles and supercontinents.

- A modification of standard electron microprobe technique allows micrometer-scale mapping of elements, including carbon, in fossils. This provides a non-destructive tool that will find use in microchemical analysis of samples returned from Mars and elsewhere.

- Team research has provided the first definitive and quantitative view of the sources of hydrogen used in biosyntheses of biomarker lipids. It thus provides vital information for the interpretation of hydrogen-isotopic compositions of organic compounds.

- Assembly and preliminary analysis of a new, comprehensive data base on the fossil record suggests that the large and continuing increase in diversity inferred for the past 250 million years could reflect geological and collecting biases as much as true evolutionary pattern.

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#12
Effects of Climate & Geology on Habitability

Project

Environmental Changes in the Context of Biological Evolution During Neoproterozoic on the Yangtze Platform, a Snowball Earth?

NAI Postdoctoral Fellow: Y. Shen
Advisor: A. Knoll

Accomplishments

How can the geochemical record provide evidence of redox evolution of Earth's atmosphere and oceans? Geochemical analysis has begun of the iron-sulfur chemistry and S-isotopic chemistry of sediments deposited along an environmental gradient in a mid-Proterozoic (ca. 1500 million year old) marine basin. The sample set extends from tidal flat and estuarine shales to deep-water mudstones deposited during maximum flooding of the basin. Preliminary analyses suggest that geochemical signatures change along the onshore-offshore environmental gradient in a way consistent with the hypothesis that mid-Proterozoic oceans were sulfidic below the surface mixed layer.

If verified by continuing analyses, this will establish the existence and long term persistence of an ocean more like the modern Black Sea than the Atlantic or Pacific. This has important consequences for primary production and biological evolution. The long term goal of this project is to establish geochemical profiles of onshore-offshore environmental gradient for two critical intervals of Earth history: (1) 2500 Myr ago, when Earth's transition to oxic surface environments was underway; (2) 1700-1500 Myr ago, when the transition to a fully oxic ocean had begun.

In other research, we will examine the geochemical records of deep water marine sediments associated with Neoproterozoic glaciation in southern China and shelf deposits associated with glaciation and iron deposition in northern China. These analyses should help to determine the stratigraphic relationship between Snowball glaciation and oceanic redox history.

Highlights

Fe/S/C shown to be useful redox probes of ancient ocean basins.

Field Trips

The field work related to this project has not yet started.

NASA Mission Involvement

This research contributes substantially to the identification of biological signatures in sedimentary rocks, a major goal of long term Mars exploration.

Future Directions

I will travel to China later this year to collect samples, and will conduct Fe/S/C analysis during the coming year.

Geobiology of Neogene Hematitic Sedimentary Rocks

Project

Senior Project Investigator(s):
A. Knoll

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

Accomplishments

This is a new subproject to study the geobiology of Neogene hematitic sedimentary rocks. Iron-rich rocks are widely distributed in sedimentary basins older than ca. 1850 million years. A number of these preserve microfossils and chemical signatures of ancient life, yet interpretation of these records has been difficult. At the same time, the Mars exploration community is likely to choose as a primary landing site for the 2003 Mars MER mission an area characterized by aqueously deposited hematite. Studies of Precambrian iron formations and martian hematite share a need for modern terrestrial analog ecosystems where biological and physical processes can be tied directly to paleobiological and geochemical patterns in deposited iron-rich sediments. The Rio Tinto drainage area of southern Spain offers just such an opportunity.

In collaboration with members of the Spanish Center for Astrobiology, we studying Neogene iron deposits associated with the Rio Tinto system. To date, we have begun petrological, paleontological and geochemical studies of sedimentary rock samples (including XRD and Mossbauer spectroscopy). Fieldwork will begin in September 2001. We have also obtained a promise from Steve Squyres, PI of the 2003 Mars MER rover, to analyze Rio Tinto samples using the instrument package that will fly to Mars. This will provide an illuminating study of what the rover can actually expect to "see" when it lands on Mars.

Field Trips

Fieldwork will begin in Spain in September 2001, in collaboration with Spanish colleagues.

Cross Team Collaborations

This project is a collaboration between the Harvard Team and the Spanish Center for Astrobiology, our NAI international partner. Richard Morris, an expert on iron mineralogy with the Johnson Space Center Team, will also participate.

NASA Mission Involvement

Our research area ties directly into the NASA 2003 MER (Mars Exploration Rover) lander mission, particularly regarding the hematite landing site.

Through Knoll's involvement and testing studies for the 2003 MER mission, we will be able to analyze samples from the Rio Tinto system with Mars Athena rover instruments.

Future Directions

- We will expand collaborative research with the Centro de AstrobiologÍa (Spain) on Neogene iron deposits in southern Spain to provide an actualistic framework for renewed paleobiological study of Precambrian iron formations as well as a potentially informative analogue for hematite deposits on Mars.

- We will initiate new paleobiological research on Precambrian iron formations and related Paleoproterozoic rocks.

The Planetary Context of Biological Evolution: Molecular and Isotopic Approaches to Microbial Ecology and Biochemistry

Project

Senior Project Investigator(s):
J. Hayes

Roadmap Objectives

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#13
Extrasolar Biomarkers

Accomplishments

The Harvard team has been working to develop techniques for the analysis of ^{13}C in sub-nanomole quantities of nucleic acids. Thus far, we have demonstrated accurate analyses of sub-nanomole quantities of carbon in samples of nucleic acids and have gained experience with the isolation of native RNA from laboratory cultures and natural samples. Our goal is to use probe-capture techniques for the isolation of specific nucleic acids. At present, we can obtain high purity (i. e., capture of RNA from only a single species) or reasonable yield, but not both at the same time. We are continuing to pursue techniques with high genomic resolution and expect eventual success.

We have collaborated with NASA Ames in studies of the fractionation of hydrogen isotopes by methanotrophic bacteria. These organisms provide an ideal entry point to the study of unicellular systems because H can flow from only two possible sources, the C-H bonds of methane or the O-H bonds of water. Interactions with the Ames group also led to the identification of a series of cultures of sulfate-reducing bacteria. We have completed hydrogen-isotopic analyses of the lipids from these same samples. We have also worked with members of the Ames group in investigations of carbon-isotopic biogeochemical studies of hydrothermal vent microbial communities. Hayes has completed a major review entitled “Fractionation of the isotopes of carbon and hydrogen in biosynthetic processes.”

We are beginning to investigate how methane functioned in Earth’s early atmosphere, using detailed studies of how methanogenesis occurs in modern sediments, and working to calculate the rates of microbial reactions. We are also developing the ability to measure calcium isotopes with high precision to investigate biomineralization in Proterozoic sediments.

Highlights

Work by Sessions and Jahnke has provided the first definitive and quantitative view of the sources of hydrogen used in biosyntheses of biomarker lipids. It thus provides vital information for the interpretation of hydrogen-isotopic compositions of organic compounds.

Cross Team Collaborations

The Woods Hole portion of the Harvard Team is involved in collaborations with these astrobiology research teams: 1) Ames Research Center; 2) Marine Biological Laboratory (MBL); and 3) University of Rhode Island and the Woods Hole Oceanographic Institution (URI-WHOI).

Work with the Ames Team involves hydrogen-isotopic studies of microbial cultures prepared by Linda Jahnke and coworkers, as well as structural and carbon-isotopic analyses of biomarker lipids from microbial communities in natural hot springs. Both of these projects were described in the progress-report section of this document.

Isotopic studies of nucleic acids, also described in the progress report, were planned jointly by John Hayes (Harvard Team) and Mitch Sogin (PI of the MBL Team). Our work thus far has drawn on Dr. Sogin's advice, but it has been based entirely at WHOI. Later phases of the work will utilize the sequencing facilities and other capabilities of the MBL Team.

Collaborations with members of the URI-WHOI group have been ongoing and, up to now, informal. In the future, we plan to use our RNA-isotopic techniques to assist Dr. Andreas Teske's studies of seafloor hydrothermal communities. Additionally, we plan to use techniques for compound-specific isotopic analyses of carbon and hydrogen to assist the URI-WHOI team in studies of sub-seafloor microbial communities.

NASA Mission Involvement

Our research asks geobiological questions about the Earth. However, techniques developed by Hayes, Schrag, and their collaborators may find application in molecular and isotopic investigations of extraterrestrial samples.

Future Directions

- Continuing development of techniques for the isotopic analysis of rRNA from microbial communities, to include improved techniques for the isolation of RNA, and the definition of sample-size requirements.

- With K. Londry, explore the nitrogenase-catalyzed hydrogenation of acetylene as a means of determining the hydrogen isotopic composition of NADPH in bacterial cells.

- With Deborah Kelley, University of Washington, analyze biomarker lipids newly available from the "Lost City" hydrothermal field, recently discovered at 30°N on the Mid-Atlantic Ridge.

The Planetary Context of Biological Evolution: Neoproterozoic-Cambrian Environment Change and Evolution

Project

Senior Project Investigator(s):
A. Knoll, J. Grotzinger, P. Hoffman, D. Schrag, S. Bowring, D. Ervin, C. Marshall

Accomplishments

Our team effort on Neoproterozoic-Cambrian environmental change and evolution has enjoyed the broadest participation of Harvard team members, for good reason. The Proterozoic-Cambrian transition witnesses remarkable changes in tectonics, climate, atmospheric composition, and especially life. This is the interval during which animal life — and hence the prospect of intelligence — radiated on Earth. Harvard team researchers are studying the paleontology, geochronology, tectonics, and environmental changes of this interval, with an eye to integrated change in the Earth system.

In conjunction with Jin Yugan from the Nanjing Institute of Palaeobiology, China, we have begun geochronological work on a series of volcanic ashes just above glacial deposits and as high as the lower middle Cambrian in an effort to constrain the ages of the spectacular fossil embryos found in terminal Proterozoic phosphorites from China, the age of Chinese glacial deposits, and the tempo of the Cambrian explosion. Continuing work on ash beds associated with Neoproterozoic glacials in Newfoundland has been problematic, but data indicate that the glacial rocks are ca 580 Ma and that the duration of their deposition is less than 10 Ma. Importantly, spectacular Ediacaran fossils first occur at 575 Ma, lending support to the idea that large animals appeared soon after the last glacial.

In 1998 Seilacher shocked the geological community by publishing a paper in which he described ca 1.0 Ga metazoan trace fossils from India. We initiated a project in conjunction with Jan Veizer at Ottawa University in Canada to investigate these findings. Rhyolitic volcanic rocks that belong to the Deonar Porcellanite Formation and which occur just below the unit containing trace fossils yield a conventional U—Pb zircon age of 1631 +/- 5 Ma. These results indicate that the Kajrahat limestone is of Paleoproterozoic age. The biological interpretation of Seilcher's bedding features remains problematic.

We have obtained an age for the Precambrian/Cambrian boundary in Oman of ca 542 Ma, defined by a negative excursion in carbon isotopes. We have also collected many ashes from drill cores to further document the tempo of changes in carbon and Sr isotopic signatures. This result raises questions regarding the biological significance of the Cambrian/Precambrian boundary as distinctive

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#12
Effects of Climate & Geology on Habitability

#14
Ecosystem Response to Rapid Environmental Change

Neoproterozoic small-shelly fossils occur both above and below the boundary. In addition, a suite of "ashes" from within the glacial deposits exposed in Oman as well as a non-deformed granite that is unconformably overlain by the glacial deposits have been collected. Geochronological data indicate that the granite age is 727 Ma, and that of the overlying glacials, 710 Ma, which may be the first robust age for the "Sturtian" glacial deposits.

In an attempt to fully understand the resolving power of U-Pb geochronology, we initiated a project to date one of the best know Ar-Ar standards, the Fish Canyon Tuff. This fully explores all sources of errors involved in U-Pb geochronology and allows us to have confidence in determining U-Pb dates in the age range of 2-550 Ma at the 20-500K year level.

We have investigated new findings from molecular development, with an eye for their application to understanding early animal evolution, concluding that developmental data have been overinterpreted, with ancestral bilaterians less morphologically complex than claimed.

The project in northern Namibia currently focuses on events directly preceding low-altitude glaciation. A decline in the $d^{13}C$ of seawater proxies of 10 per mil in ~0.5 million years is observed in virtually all sections of the Otavi Group of northern Namibia preceding the Ghaub glaciation. An analogous isotopic change is observed before the Elatina glaciation in South Australia.

We have completed the final year for our project in northeast Svalbard, which will be a major part of Halverson's doctoral thesis at Harvard. We have discovered a sequence having the lithologic and isotopic signatures of "cap" carbonates (purportedly diagnostic of Snowball Earth deglaciations) in the lower Akademikerbreen Group, predating any known glacial event in Svalbard. This indicates either that "cap" carbonates are not unique to glacial events, or that a third, previously unrecognized Snowball event occurred. Preliminary paleomagnetic measurements indicate that the sequence retains a primary remnant magnetization. The data imply that the "phantom" glaciation occurred at low paleolatitude.

Dan Schrag continues to explore models of coupled environmental and biological change at the end of the Proterozoic Eon. This work has been collaborative with four team members: one manuscript is in preparation with Schrag, Bowring and Hoffman as co-authors, and another manuscript is in preparation with Schrag, Erwin and Knoll as co-authors. In addition, postdoc MaryLynn Musgrove is developing the ability to measure calcium isotopes with high precision to investigate biomineralization in Proterozoic sediments.

Highlights

• Completed the accurate digital reconstruction of Namacalathus, a terminal Proterozoic calcified metazoan, using high-resolution serial sectioning. This reconstruction enabled accurate assessment of taxonomic diversity as well as functional morphology of early calcified animals remains.

• Discovery of Namacalathus and Cloudina below and above a significant carbon-isotope excursion in Oman, interpreted as the Precambrian-Cambrian boundary biogeochemical event. This discovery indicates that these organisms did not go extinct at the boundary.

• Development of a counterintuitive model, based on carbon isotopic data, that provides a potential trigger mechanism for Neoproterozoic "Snowball Earth" events.

• Elucidation of the ecological and taphonomic processes that govern the formation and preservation of microbial mat biosignatures in ancient siliciclastic rocks, providing new insights on a biosignature that is potentially applicable to Mars rover exploration.

• New geochronologic constraints on the timing and duration of late Neoproterozoic glaciation in Newfoundland and its relationship to Ediacaran animal diversification

Field Trips

Oman, January 4-17, 2001.

To visit two localities where terminal Neoproterozoic rocks are exposed. In both localities, we searched for ash-beds and other rocks that could constrain the age and duration of glacial deposits. Participants: Grotzinger, Schroeder, Bowring.

Kunming, China, March 23 - April 1, 2001.

The purpose of this trip was to look for and collect ashbeds through the transition from the terminal Neoproterozoic to the Cambrian. Participants: Bowring and Erwin.

St. John's, Newfoundland, June 9 - 12, 2001.

The purpose of this trip was to collect ashbeds and to look for fossils below, within, and above the Neoproterozoic Gaskiers glacial deposits exposed in Newfoundland. Jahan Ramezani, MIT.

Namibia, July 31 - August 15, 2001.

Stratigraphy and paleobiology of the terminal Proterozoic Nama Group. Participants: Grotiznger, Bowirng, Erwin, Knoll, Noffke, plus James Hagadorn and Dianne Newman (Caltech).

Namibia, Morroco, Svalbard, 2000-2001

Namibia (1 month), Morocco (3 months), Svalbard (0.5 months)
This research is designed to characterize and test models for Snowball glaciation and early animal evolution. Participants: Hoffman, Halversen, Maloof.

Cross Team Collaborations

Co-I Grotzinger initiated collaborative research with Roger Summons, currently part of the Australian Astrobiology Centre. R. Summons will soon join the faculty at MIT (and, hence, the Harvard Team).

NASA Mission Involvement

The digital mapping techniques under development by Grotzinger will be applicable to any serious investigation of martian geology undertaken in Mars lander expeditions in 2007 and beyond. Such Mars lander and rover missions are being planned to perform scientific studies of Mars and demonstrate technology for accurate landing and hazard avoidance in traveling to difficult -to-reach sites.

Elucidation of microbial mat biosignatures in siliciclastic rocks, investigated by Noffke and Knoll, provides insights into one of the only sedimentary biosignatures that can be observed by the 2003 MER mission (Mars Exploration Rover) and not be mimicked easily by physical processes.

Understanding the physical environmental conditions of Earth during, before, and after Snowball glaciation is also immediately relevant to projects on astrobiological characterization of extrasolar planets, currently under development by NASA.

Future Directions

- Bowring will continue to analyze large suites of ashes from southern China and Newfoundland. The goal is to complete a coordinated field and laboratory geochemical study of volcanic events that can calibrate the Proterozoic-Cambrian time scale of Earth system evolution.

- Hoffman and his students will continue their field projects, emphasizing new projects in southern Africa and Australia. The goal is to constrain the timing, mechanisms, and consequences of Neoproterozoic ice ages, including specific tests of the snowball Earth model.

- Grotzinger will continue examination of subsurface cores in Oman for paleontologic information on Cambrian ecosystem expansion. The project will be in collaboration with Roger Summons who will collect for organic biomarker analysis aimed at elucidating the taxonomic diversity and paleoecology of microbial communities.

- Knoll will initiate a project with postdoctoral fellow Jochen Brocks and Roger Summons to evaluate how molecular biomarker signatures change across the Proterozoic-Cambrian transition in China and Siberia. Along with the Oman work, this project has the goal of characterizing the whole-ecosystem changes associated with the Cambrian explosion of animal life.

- Schrag will continue to develop and test models of Snowball glaciation (and deglaciation) and will continue geochemical work designed to provide new insights into the origin and geochemical consequences of skeleton biomineralization.

- Erwin will continue to evaluate the relative importance of ecological and developmental components of the late Neoproterozoic metazoan radiation by focusing on the origin of the bilateria, using a variety of field and theoretical approaches.

The Planetary Context of Biological Evolution: Permo-Triassic Mass Extinction and its Consequences

Project

Senior Project Investigator(s):
S. Bowring, D. Erwin, J. Marshall, A. Knoll

Roadmap Objectives

#5 Linking Planetary & Biological Evolution

#12 Effects of Climate & Geology on Habitability

#14 Ecosystem Response to Rapid Environmental Change

Accomplishments

We have completed a lengthy review of the nature of the end-Permian mass extinction, including an evaluation of proposed extinction mechanisms. We seek to understand the causes and biological consequences of the great Permo-Triassic mass extinction, 251 million years ago. We continue to make progress along the lines outlined in our proposal. The additional of John Marshall as a Co-I has substantially improved our ability to frame and test hypotheses by oceanographic modeling.

We have been working on possible physical/biogeochemical mechanisms that may have been players in the end-Permian mass extinction. This has involved developing circulation models of the paleo-ocean and using them to drive biogeochemical models. Their focus has been the end Permian, but the insights and tools that have been developed are relevant to other warm periods of climate. This work goes a long way toward spelling out precisely what conditions are required to support a haline mode with sinking of brackish waters from the subtropics, a recurring scenario in the geological literature. Haline modes are favored in climates that are warm, with weak pole-equator temperature gradients, enhanced hydrological cycles and supercontinents.

Our recent work has sharply constrained the timing of end-Permian mass extinction. The event took place in a time interval shorter than (and perhaps much shorter than) 200,000 years. This result provides firm new constraints on the types of planetary or extraplanetary processes that could have caused this great extinction. Complementary research has clarified the nature of biological recovery following the extinction. It was the nature of the recovery and not just the particulars of the extinction that set the subsequent course of evolution on Earth.

In collaborative research, we modified standard electron microprobe techniques to make micrometer-scale elemental maps, including carbon, of silicified plants.

These provide direct tests of taphonomic hypotheses regarding silica permineralization and demonstrate high-resolution, non-destructive microchemical assays of the type that will be critical in the analysis of samples returned from Mars. We have also used nuclear magnetic resonance (NMR) and soft X-ray techniques to examine the distribution of lignified tissues in early plants. This research shows that the anatomical modeling of tracheids was decoupled in time from lignification; more generally it paves the way toward physiological studies of ancient plants.

Highlights

- End Permian mass extinction was catastrophic, evidently occurring in much less than 500 Ky.

- Oceanographic modeling spells out the conditions which are required to support a haline mode of circulation with sinking of brackish waters from the subtropics, a recurring scenario in discussion of Permo-Triassic mass extinction. Haline modes are favored in climates that are warm, with weak pole-equator temperature gradients, enhanced hydrological cycles and supercontinents.

- Modified electron microprobe techniques allow micron-scale mapping of elements, including carbon in fossils. This provides a non-destructive tool that will find use in microanalysis of samples returned from Mars or elsewhere.

Cross Team Collaborations

Collaborative research in this project is with the Carnegie Geophysical Lab (CIW Team). NAI members involved in this project include K. Boyce, A. Knoll, R. Hazen, G. Cody, and M. Fogel. The principal aim of this research is to understand aspects of physiological evolution in ancient organisms.

Our project research concentrates on silicified vascular plants because of their anatomical richness and the close spatial relationships between biochemical constituents and anatomy. To date, we have used modified electron microprobe techniques to make micron-scale elemental maps (including carbon) of silicified plants, providing direct tests of taphonomic hypotheses regarding silica permineralization.

We have also used NMR and soft x-ray techniques to examine the distribution of lignified tissues in early plants. This research shows that the anatomical modeling of tracheids was decoupled in time from lignification. Lignitic tissues may have first evolved in cortical tissues as protection against bacteria. Later, the relevant genes were expressed during development of conducting tissues.

NASA Mission Involvement

Our research project has no specific NASA mission involvement. However, the non-destructive elemental mapping techniques discussed in this report will find application in microchemical analysis of future samples returned from Mars or elsewhere.

Future Directions

- Erwin's primary goal is continued development of processed-based models of post-extinction biotic recovery patterns which are testable using geologic and paleontologic data.

- In collaboration with Erwin, Bowring will continue to collect and analyze volcanic ash beds that will allow improved calibration of Early Triassic time (the interval of biological recovery).

- As an NRC Fellow working at Harvard, Boyce will continue his research on plant development and evolution, including continuing paleophysiological research with members of the Carnegie team.

- Marshall's team will continue to develop oceanographic models able to constrain thinking about end-Permian events.

The Planetary Context of Biological Evolution: The Proterozoic Oxidation of the Earth's Surface

Project

Senior Project Investigator(s):
A. Anbar, H. Holland, A. Knoll, P. Hoffman, D. Schrag

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#12
Effects of Climate & Geology on Habitability

Accomplishments

The Harvard team's research focuses on major transitions in the evolution of life on Earth. Currently we have six active subprojects that address this theme:

- The oxidation of the Earth's surface, especially events that took place 2400-2200 million years ago and, again, near the end of the Proterozoic Eon

- Intense glaciation, biogeochemical change and the radiation of animals 600-525 million years ago

- Mass extinction at the Permo-Triassic boundary 250 million years ago

- Isotopic characterization of molecular biosignatures, with the goal of understanding the functional as well as systematic relationships of microorganisms in natural ecosystems

- Geobiology of sedimentary iron deposits, with goals that include understanding terrestrial analogs of the aqueous hematite terrain likely to be chosen as a principal landing site in the Mars MER 2003 mission

- EvoGenomics: collaborative focus group research on molecular phylogeny

Our results include the following:

In a reconnaissance study, Mo isotopic compositions were characterized in Devonian black shales, Black Sea sediments, Pacific and Atlantic ferromanganese nodules, continental molybdenites, and seawater. The largest variation, between Mo in ferromanganese and carbonaceous sediments, suggest Mo isotope fractionation related to environmental redox conditions. The isotopic composition of Mo in seawater is similar to that of carbonaceous sediments, consistent with high efficiency Mo scavenging in sulfidic environments. The data suggest that fractionation occurs during scavenging of Mo onto ferromanganese particles. Isotopic mass balance indicates that the observations are consistent with the proportions of Mo removal to oxic and sulfidic marine sediments today. If so, the Mo isotopes in seawater may reflect changes in the relative proportions of oxic to sulfidic seafloor, and the isotopic composition of Mo in black shales may constitute a paleoredox record.

Geochemical study of mid-Proterozoic black shales deposited during maximum flooding of the McArthur Basin, northern Australia, indicates sulfidic deep water, as proposed in 1998 by Canfield. Both ca. 1730 and 1636 Ma shales display features of Fe and S chemistry that match patterns found in the deep Black Sea today. S-isotopes also suggest substantial sulfate depletion in these waters, consistent with an oceanic sulfate inventory much lower than today's.

Canfield proposed that the deep sea was sulfidic, rather than oxic, from 1850 Ma (when Banded Iron Formations (BIF) stopped forming) until at least 1200-1000 Ma ago. This period of time is also characterized by unusual stability in the carbonate carbon isotope record, by unexplained delay in the diversification of eukaryotes, and by unusual onshore-offshore decrease in diversity of fossils of eukaryotic origin. We propose that these observations are related by the extreme redox sensitivity of Fe and Mo- both metals critical to the biological N cycle.

We have discovered abundant and well-preserved microfossil assemblages in the siliciclastic Roper Group, northern Australia. The Roper Group is well characterized in terms of age (1492+/-3 SHRIMP age on zircon in a tuff low in the succession) and sequence architecture. It is also fossiliferous throughout, enabling us to relate fossil assemblages to facies. Eukaryotic fossils decrease in abundance and diversity from marginal marine to basinal shales, but increase in equability. This ecological pattern is predicted by the trace element model. Process-bearing acritarchs provide the first evidence for modern cytoskeletal architecture in 1500 Ma protists.

Highlights

- Mo isotopes shown to be potential paleoredox probes

- Deep water in a mid-Proterozoic basin is shown to be sulfidic, consistent with models suggesting that the modern oxic deep ocean only came to exist near the end of the Proterozoic Eon.

- Scarcity of critical nutrient metals in postulated sulfidic Proterozoic oceans may help to explain the observed ecological and stratigraphic distribution of eukaryotic algae in Proterozoic sedimentary rocks.

Cross Team Collaborations

Our primary collaborative effort in Year 3 was planning the "Mission to Early Earth" (MtEE) field excursion to West Australia, an outgrowth of the MtEE Focus Group. This excursion will involve approx. 12 participants from at least 6 NAI Teams, as well as a participant from the Centro de Astrobiologĺa (Spain) and from the Australian Centre for Astrobiology. Four of the participants are graduate students.

Year 3 also saw publication of a collaborative manuscript on the bombardment environment of the early Earth in Journal of Geophysical Research-Planets, which involved S. Mojzsis (CUB Team) and K. Zahnle (ARC Team).

Research to evaluate the utility of Fe isotopes for biosignature investigations continued our collaboration with the JPL Team. This interactive work in Year 3 led to submission of two new manuscripts (to Earth and Planetary Science Letters and to Chemical Geology), plus publication of a review article in Eos. Anbar has begun a new collaboration with Sue Brantley (PSU Team) on Fe isotope effects.

Knoll's research on the geobiology of Proterozoic basins in Australia is conducted jointly with Malcolm Walter of the Australian Astrobiology Centre.

NASA Mission Involvement

A.H. Knoll is a member of the Science team for the 2003 MER (Mars Exploration Rover) mission to Mars.

More generally, research by the Harvard team contributes substantially to the identification of biological signatures in sedimentary rocks, a major goal of long term Mars exploration. This project contributes studies of trace metals and microfossils that help shape debate about biomarkers that can be applied to solar system exploration. Isotopic fractionation of metals by microorganisms holds promise as a biosignature applicable to extraterrestrial materials. Research on Mo complements ongoing research on Fe.

Our research also aims to provide sharper constraints on the atmospheric chemistry of the early Earth, which is highly relevant to biodetection research on extrasolar planets. One of the proposed search strategies for extraterrestrial life is to look for evidence of atmospheric oxygen, using spectroscopic observations of extra solar planets. It is generally accepted that the Earth's atmosphere was largely devoid of free oxygen for the first half of its history. Hence, such a search strategy requires a mechanistic understanding of the evolution of atmospheric oxygen. Our research project is aimed squarely at this issue.

Future Directions

• Anbar will continue research on Mo isotopes, initiating analyses of Precambrian and younger sediments as well as laboratory experiments designed to understand the processes that fractionate Mo isotopes in nature.

• Anbar will also study the concentrations of Mo, Re and other redox-sensitive metals in Precambrian black shales to complement isotopic insights (a delayed project from prior years). This will include studies of the Roper Group — see collaborative projects in the next bullet.

• NRC Fellow Yanan Shen will conduct studies of Fe/S/C systematics and S isotopes in shales of the ca. 1500 Roper Group. New associate Jochen Brocks will study biomarker distribution in Roper, Neoproterozoic and Cambrian carbonaceous shales, searching for biomarker signatures of eukaryotic clades that can augment Knoll's continuing stratigraphic and paleoecological studies of the fossil record.

• Holland will continue geochemical analyses of Archean black shales and paleosols, with the goal of placing firmer limits on oxygen concentrations prior to the great oxygen event of 2400-2200 Ma.

EvoGenomics (Collaborative Focus Group Research)

Project

Roadmap Objectives

#4
Genomic Clues to Evolution

Senior Project Investigator(s):
C. Marshall

Accomplishments

This year we began a new subproject on molecular evolution and phylogeny as part of the NAI Focus Group in EvoGenomics.

(a) Veracity of molecular clocks: work has continued on a new method for accounting for the stratigraphic ranges of species not preserved in the fossil record, with special application to primate origins. The goal is to provide a better understanding of the relationship between molecular clock and paleontological estimates of evolutionary divergence times. This is collaborative work with scientists at the University of Southern California and in Zurich.

(b) De-convolving the biological from geological signals in Phanerozoic marine diversity studies: We have just published a large study that posits the steady increase in biodiversity in the marine realm after the end-Permian mass extinction may be a sampling artifact, the result of the increase in fossiliferous rock towards the Recent. We will continue to develop the database and analysis tools to under take this work.

(c) Quantitative analysis of evolutionary patterns in local stratigraphic sections: work continues on how to best estimate the position of mass extinction horizons in local sections, using both frequentist and Bayesian methods.

(d) Evolutionary genomics: The NAI Evolutionary Genomics Focus Group has made good progress towards identifying key metazoans that should be the focus of some relatively intensive shot-gun sequencing efforts. The object will be to provide data for molecular clocks, as well as to search for key developmental genes.

Highlights

- Assembly and preliminary analysis of a comprehensive data base on the fossil record. Early results suggest that the large and continuing increase in diversity inferred for the past 250 million years could reflect geological and collecting biases as much as true evolutionary pattern.

Future Directions

- To continue analytical research in paleontology and stratigraphy, with the goal of understanding better how fossils reflect evolutionary history.

- To build collaborative research with the NAI EvoGenomics Focus Group.

Education & Public Outreach

Research and Conference Presentations

Harvard team members presented 26 lectures at universities, national and international meetings, including plenary lectures and major meetings by A. Knoll, J. Hayes, D. Erwin, and P. Hoffman.

Curriculum Evaluation

A.H. Knoll served on the external Visiting Committee for three programs evaluating curriculum. All programs seek to build excellence in geobiology/astrobiology.

Earth's Early Environments

Earth's Early Environment was presentation to 4th grade students at the American-British Academy in Muscat, Sultanate of Oman given by J. Grotzinger.

Snowball Earth lecture

Dan Schrag spent an evening discussing the Snowball Earth hypothesis, and how extreme environmental fluctuations affect biological evolution with a public audience of 300 people in Santa Fe, New Mexico.

Teaching Astrobiology to 5th graders

Members of the Harvard team gave a series of four talks on the climate of early Earth to 5th grade students in Oakland, California.

Publications

Back to Table of Contents

The Planetary Context of Biological Evolution: Molecular and Isotopic Approaches to Microbial Ecology and Biogeochemistry

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Back to Table of Contents

Jet Propulsion Laboratory (1)

NASA Astrobiology Institute

Pasadena, California

Principal Investigator

Kenneth Nealson

Jet Propulsion Laboratory (1) Team Members

Thomas J. Ahrens
California Institute Of Technology

Ariel D. Anbar
University of Rochester

G. Arnold
University of Rochester

Jill Banfield
University of Wisconsin Madison

William W. Barker
University of Wisconsin Madison

J. Barling
University of Rochester

Brian L. Beard
University of Wisconsin Madison

Rohit Bhartia
Jet Propulsion Laboratory

Christopher Boxe
California Institute of Technology

Karen Brinton
California Institute of Technology

Jeffrey Brownson
University of Wisconsin Madison

Clara Chan
University of Wisconsin Madison

Pamela G. Conrad
Jet Propulsion Laboratory

Marilyn L. Fogel
Carnegie Institution of Washington

Lisa Gaddis
USGS Flagstaff

S. Goldman
University of Rochester

Satish Gupta
California Institute of Technology

James W. Hagadorn
California Institute of Technology

Robert M. Hazen
Carnegie Institution Of Washington

E. (Beth) Holman
University of Rochester

William Hug
Photonics Inc.

Clark M. Johnson
University of Wisconsin Madison

Jeff Johnson
USGS Flagstaff

Vadym Kapinus
California Institute of Technology

Joseph L. Kirschvink
California Institute of Technology

James Lyons
University of California Los Angeles

Francis MacDonald
California Institute of Technology

Gene D. McDonald
Jet Propulsion Laboratory

John Moreau
University of Wisconsin Madison

Kenneth H. Nealson
Jet Propulsion Laboratory

Maria Nesterova
University of Wisconsin Madison

William Newman
University of California Los Angeles

John D. O'Keefe
California Institute of Technology

Jeffrey Plescia
USGS Flagstaff

M. Polizzotto
University of Rochester

Radu Popa
Jet Propulsion Laboratory

Thom Rahn
California Institute Of Technology

J. Roe
University of Rochester

James H. Scott
Carnegie Institution of Washington

Andy Shen
California Institute of Technology

Anne Skatvold
University of Wisconsin Madison

Joseph Skulan
University of Wisconsin Madison

Andrew Steele
University of Portsmouth

Sarah Stewart
California Institute of Technology

Michael Storrie-Lombardi
Jet Propulsion Laboratory

Henry J. Sun
Jet Propulsion Laboratory

Yohey Suzuki
University of Wisconsin Madison

Kenneth Tanaka
USGS Flagstaff

Alexandre Tsapin
Jet Propulsion Laboratory

Benjamin Weiss
California Institute of Technology

Susan Welch
University of Wisconsin Madison

G. Wortman
University of Rochester

Yuk Yung
California Institute of Technology

Executive Summary

Accomplishments

The JPL/Caltech node of the NAI has slowly evolved its scientific focus to be on the definition and detection of biosignatures that might be used to find evidence for extant or past life on Earth and in extraterrestrial environments, or within samples returned from such extraterrestrial sites. This evolution has occurred both in response to the members of our group expressing these interests, as well as the reviewers' suggestions from the initial CAN application. The group includes members from a variety of research environments, including JPL, Caltech GPS (Geological and Planetary Sciences), University of Wisconsin (Geology), University of Rochester (Chemistry), USGS in Flagstaff, and the Carnegie Institution of Washington. The goals can be generally divided into three groups:

1. Stable isotopes of biosignatures:
    - a. nitrogen isotopes – Blake, Caltech
    - b. carbon and sulfur isotopes – Fogel and Rummel, Carnegie Inst. of Washington
    - c. iron isotopes – Beard and Johnson, Wisconsin
    - d. transition metal isotopes – Anbar, Univ. of Rochester

2. Reflectance spectroscopic biosignatures
    - a. large scale signatures in water bodies – USGS group
    - b. reflectance signatures in soils and rocks – Conrad, JPL

3. Life detection methodology – biosignature definition and measurement
    - a. definition of non-earthcentric biosignatures – Conrad & Nealson, JPL
    - b. UV fluorescence and Raman methods – Storrie-Lombardi et al., JPL
    - c. X-ray CT scanning methods – Tsapin, JPL
    - d. software development for life detection decision making and complexity analysis
    - e. study of extreme environments as analogues for extraterrestrial sites – Nealson, JPL

4. Metal oxide/organism interactions as biosignatures
    - a. magnetite as a biosignature – Kirschvink, Caltech
    - b. iron oxidizing bacteria, and low pH metal alteration – Banfield, Wisconsin
    - c. iron and manganese oxidizing and reducing bacteria – Nealson, JPL

The research focus is on microbiology, geomicrobiology, and the interactions between microbes and metals of different types. The work on stable isotopes has resulted in a number of advances in our thinking, especially in the area of stable isotopes of iron, but the work with sulfur, carbon, and nitrogen has also been very good. The work on iron isotopes is spearheaded by two groups, the Johnson/Beard group at Wisconsin, and the Anbar group at Rochester. While there is some disagreement between these two groups, both have published several articles on various aspects of iron fractionation, and the NAI effort in this area can be regarded as world-class. The acquisition of the NAI facility mass spectrometer at U. of Wisconsin was a significant event, and has greatly aided in data availability by requiring smaller samples, speeding up the analysis time, and increasing the precision of the measurements.

One part of the biosignatures work, headed up by Gene McDonald (JPL), has been using amino acid ratios as indicators of life. This work has revealed two new and potentially very useful facts. First, amino acids (and their racemic signatures) are preserved for millions of years inside of Antarctic rocks where there are populations of endolithic bacteria near the surface. This allows one to not only estimate the amount of time that the rock has been populated, but would in principle allow one to identify a rock that had been, but was no longer populated by endolithic bacteria. Second, using amino acid racemization analyses, it has been possible to estimate the temperature history of some Arctic permafrost samples, something that has been very difficult to accomplish in the past.

The project on life detection methodology at JPL has emerged as one of importance. This project, led by Drs. Michael Storrie-Lombardi and Alexandre (Sasha) Tsapin, focuses on the use of two new technologies for the detection of potential life-containing strata. These two approaches (deep UV (224 nm) fluorescence, and CT X-ray scanning) have both yielded exceptional results, and are being prepared for publication now. Both of these projects have led to submission of instrument proposals to the Astrobiology Science and Technology Instrument Development program for flight instrument development.

Of particular note has been the work of Dr. J. Kirschvink, who has taken a strong stand on the issue of magnetite as a biosignature, and has published several abstracts and one paper strongly defending the idea that magnetite in the ALH84001 meteorite is an indicator of past life on Mars. This admittedly controversial stance has added an element of dynamics and tension to our already dynamic and tense NAI group!

Roadmap Objectives

#1
Sources of Organics on Earth

#2
Origin of Life's Cellular Components

#6
Microbial Ecology

#9
Life's Precursors & Habitats in the Outer Solar System

Project

A New Molecular Recognition Instrument for Astrobiology Applications

Senior Project Investigator(s):
M. Fogel

Accomplishments

Protein chip-based molecular recognition technology is being developed as a method for detecting life on Earth and other solar system bodies. The Ciphergen Biosystems ProteinChip Reader (PBSII) is dependent on the surface chemistry of small chips to selectively capture femtomole amounts of complex mixtures of organic molecules for molecular weight determination by time of flight mass spectrometry. By using different chips, experiments can be designed to recognize a complex combination of molecular markers by selecting a series of chip surface chemistries to pull apart complex mixtures.

Proteomics: We determined the protein profile of *Shewanella putrefascens*, a microorganism that is commonly isolated from geochemical gradient zones in which dissolved oxygen levels range from fully oxic to completely anoxic. Replicate analyses of the chip detected 62 peptides or proteins from 754 to 237,954 daltons, some of which corresponded to molecular weights of protein identified from the genome. The instrument detected proteins in the lower pico-mole to higher femtomole range.

Geochemical Studies: On Earth, as well as Europa, the bulk of organic carbon molecules in the world's oceans are found dissolved in seawater. However, the chemical composition of dissolved organic matter is complex and poorly under-stood, making it a good proxy to what might exist on Europa or Mars. The distri-bution of organic molecules was determined to see whether large proteins or microbial cell wall fragments could be detected from the Pacific Ocean (100 m depth) and from a riverine-derived filtrate.

Highlights

- Compared three different types of organic molecules, including microbial pro-teins, biologically-produced but chemically modified microbial molecules, and abiotically synthesized organic matter

- Molecular weight ranges of the three groups segregated: biological material (up to 250,000 Daltons), geochemical molecules (up to 6,000 Daltons), and abiotic molecules (up to 1,000 Daltons

Field Trips

Belize, August and November, 2000.

Microbial mats. With Biocomplexity project including Matthew Wooller (CIW-GL) and Samantha Joye (Univ. of Georgia)

NASA Mission Involvement

In this research project, we would like to develop the scientific portions of this instrument and its capabilities. We would then eventually adapt this instrument for space flight to carry out *in situ* analysis of martian soils.

Future Directions

- Expand list of microbes investigated with the technology, to include the Archaea
- Increase the number of collaborators participating in the project
- Become more familiar with the use of antibody binding chips

Astrobiology Funding to the US Geological Survey

Project

Senior Project Investigator(s):
J. Plescia

Accomplishments

The primary objective of last year's work was to examine hyperthermophile communities in Yellowstone National Park and tufa deposits in the Mono and Searles Lake Basins to determine whether these different communities exhibited unique spectral signatures in the visible to near-infrared wavelengths (350-2500 nm). We focused the YNP work on Mushroom Spring, Octopus Spring and Nymph Creek. In those areas we found that each of the major communities displays spectral absorptions unique to each community. For example, *Cyanidium caldarium* from Nymph Creek has a green peak at 542 nm and an otherwise flat visible spectra. The red edge occurs at 710 nm and there is an absorption centered at 878 nm. Downstream, Chlorella-like organisms occur. These organisms have spectral characteristic very different from the Cyanidium-like community at the spring source. Phormidium from Octopus Spring has a complex spectra with visible maxima at 590 and 648 nm, chlorophyll a absorption at 675 nm, and absorptions at 740 (bacteriochlorophyll c), 798, and 874 nm. The *in situ* spectra show the same absorptions as the extracted *Synechococcus* chlorophyll a absorption, red edge at 765 nm and absorptions at 716, 798 and 877 nm. The red color of the *Chloroflexus* is subdued as it lies below the *Synechococcus*. These differences are due not only to the various compounds but also the manner in which they occur in the organisms. Differences in the spectral character of sinter were also observed, with sinter around pools having hyperthermophile organisms exhibiting absorptions due to organic compounds. Data for tufa from Mono Lake and Searles Lake show that both thermal emission and visible-near infrared variations in the spectral character of tufa occur, which can be attributed to differences not only in the mineralogy but also to differences in other attributes such as the amount of absorbed water.

Highlights

- Spectral data can be used to remotely discriminate and identify hyperthermophilic organisms.

- Carbonates deposited in extreme alkaline environments display unique spectral signatures.

Field Trips

Yellowstone National Park, October 2000.

Fieldwork was conducted at Yellowstone National Park in October 2000 by J. Plescia and J. Johnson (USGS)

Mono and Searles Lake, California, Summer 2000.

Fieldwork was conducted at Mono and Searles Lake, CA, in the summer of 2000 by J. Plescia and J. Johnson.

NASA Mission Involvement

NAI-funded investigators in the U.S. Geological Survey primarily focus on research to examine hyperthermophile communities in Yellowstone National Park (Wyoming) and tufa deposits in the Mono and Searles Lake Basins (California).

Research is aimed at understanding whether extremophile habitats can be characterized with remotely-acquired spectral data and whether the presence of fossilized organisms can be recognized with remotely-sensed data. An understanding of the spectral and spatial aspects of such data is needed to recognize particular extremophile environments using remotely sensed data This work will contribute to the planning and interpretation of such data acquired from surface or orbital platforms for martian sites.

Future Directions

- Obtain additional spectral data on hyperthermophilic organisms. Our objective is to determine if the same organisms in different springs have the same signatures; also collect data for additional communities.

- Obtain spectral data on various age sinter. Objective is to determine if any of the spectral features observed in the hyperthermophilic communities are preserved in the sinter.

Coevolution of Earth and Mars

Project

Senior Project Investigator(s):
Y. Yung

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

#13
Extrasolar Biomarkers

Accomplishments

Previously suggested gases for terraforming Mars include the chloro fluorocarbons and SF6. Recent, detailed calculations for C2F6 and the CFC's have led to optimism about terraforming. Some means other than greenhouse gases must be used for the initial warming, as no known gas mixture in a small total column amount will absorb strongly at current martian pressure (600 Pa). We introduced some new candidate gases, and we framed our inquiry by asking what mixture of trace gases could sustain an earthlike temperature if Mars were somehow endowed with an earthlike atmospheric composition and earthlike surface pressure. We suggested that a 70K greenhouse effect might be maintainable with as little as 5×10^{22} m^{-2} column amount of a mixture of "designer" greenhouse molecules. This molecular column corresponds to about 240 ppbv in Earth's atmosphere.

Highlights

A more speculative corollary is that advanced extrasolar civilizations, if they exist, may have already engineered planetary environments in zones we would consider inhospitably cold. Therefore, searches for extraterrestrial intelligence, which now mainly seek radio waves, should perhaps include looking for spectra of manufacturable molecules such as those mentioned here.

Cross Team Collaborations

We have had fruitful exchange with Chris McKay with the ARC Team.

NASA Mission Involvement

Our research will have an impact on Mars Sample Return (MSR) programs by focusing on what material we should look for on Mars. These important Mars materials are those essential for life and terraforming.

Future Directions

- We will carry out detailed modeling that will improve upon the greenhouse model of Mars to test out new ideas and new molecules capable of producing a supergreenhouse effect on Mars.

Roadmap Objectives

#2
Origin of Life's Cellular Components

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#9
Life's Precursors & Habitats in the Outer Solar System

#10
Natural Migration of Life

#11
Origin of Habitable Planets

#12
Effects of Climate & Geology on Habitability

#14
Ecosystem Response to Rapid Environmental Change

#17
Planetary Protection

- We also propose to model the sensitivity of biogenic gases such as CH_4 and N_2O as an indication of the martian biosphere. For example, if we can measure CH_4 and N_2O to 0.14 and 0.015 ppbv, then a simple argument shows that we can set useful limits like Mars source/Earth source < 10^{-8} and 10^{-4}, respectively.

Project

Coevolution of Life and Planets

Senior Project Investigator(s):
J. Kirschvink

Accomplishments

We have shown that magnetism in martian meteorite ALH84001 is at least 4 Gy old, demonstrating that Mars had a geodynamo within 500 million years of its formation. We have provided strong evidence that seven characteristics of the magnetites in ALH84001 are actually only known elsewhere among magnetites produced by magnetotactic bacteria. We have characterized the climatic and biological changes resulting from the Snowball Earth episodes: the Paleoproterozoic Snowball Earth may have been the chief stimulus that led to the evolution of oxygenic photosynthesis and the rise of atmospheric oxygen. And we have expanded our understanding of the terminal Neoproterozoic history of southwestern North America and the origin of bilaterians.

Highlights

- Evidence for an ancient martian magnetic field: our new magnetic studies of martian meteorite ALH84001 demonstrated that Mars had a geodynamo by 4 Ga.

- Magnetofossils in ALH84001: The magnetites in ALH84001 have seven characteristics which collectively are only known in the magnetite produced by terrestrial magnetotactic bacteria.

- A low temperature transfer of ALH84001 from Mars to Earth: our magnetic studies of ALH84001 demonstrate the meteorite was not heat-sterilized during its journey from the martian surface to our laboratory.

- The Paleoproterozoic Snowball Earth and the rise of atmospheric oxygen: This glacial interval may be the chief climactic stimulus that led to the arise of oxygenic photosynthesis and the evolution of the eukaryotic cell.

Cross Team Collaborations

Working with the Bowring group at MIT (Harvard Team), we reported a critical age constraint on the origin of the first bilaterian fossils (Martin et al.).

NASA Mission Involvement

Understanding life during Snowball Earth conditions has clear implications for detecting life on Europa and Ganymede. Work on characterization of bacterial magnetofossils may lead to detection criteria for identifying them in samples returned from Mars.

Future Directions

- To see whether Hadean zircons from Australia record evidence of an ancient terrestrial field.
- To read the history of lunar magnetism and impacts from impact-produced glass spherules in the Apollo collection.
- To further strengthen the evidence for the inertial interchange true-polar wander episode at the Precambrian-Cambrian boundary with more pole positions.
- To further strengthen the evidence for the Paleoproterozoic wander Snowball Earth episode with more pole positions, magnetostratigraphy, and chemostratigraphy across the proposed glacial interval.
- To determine whether magnetofossil-like magnetites can be produced from shock-heated carbonate.

Development of Life Detection Methodology and Technology

Project

Senior Project Investigator(s):
K. Nealson

Accomplishments

This project has become one of the major activities of the Astrobiology Program at JPL/Caltech, with many members of the JPL laboratory being involved in the definition of non-earthcentric biosignatures and in the development of methods to measure these biosignatures. It is closely connected to the conceptual issue of the co-evolution of Earth and life on one hand, and the search for life outside the Earth on the other. The program consists of four parts: 1) the definition of life and its biosignatures; 2) the development of methods for measurement of these biosignatures;

Roadmap Objectives

#3
Models for Life

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#17
Planetary Protection

3) the fabrication of instruments and software to enable these measurements in the field and eventually in flight, and 4) the use of extreme environments on Earth as terrestrial analogs of life – both for testing our life detection methods, and for learning more about the limits of life that should be considered. Finally, there is a genomics component to the work, as one of the major organisms of interest has now been completely sequenced, and genome expression and analysis has become possible. This latter program is jointly funded by NAI and the Department of Energy microbial program.

In the definition of life, we have published two articles on the subject of non-earth-centric life detection, defining the elements that should be measured, and delineating a preliminary strategy for the measurements. In addition, an internal document describing this strategy and how it can be applied to missions is under preparation.

With regard to the development of methods for measurements, two major projects have emerged. One of these involves the development of UV laser induced fluorescence methods for the detection of organic carbon signatures, while the other involves the development of a desktop computerized tomographic (CT) X-ray unit for the detection of life within endolithic environments. This approach has been very successful as well, with current resolution of endolithic populations now possible at the few micrometer level.

With regard to instrument fabrication, the UV fluorescence apparatus is now to the size of a backpack unit, while the CT instrumentation is a small desktop unit, soon to be made smaller. In both cases considerable software development has been necessary to adapt the instruments to their new scientific roles. Finally, the work in extreme environments has yielded a range of results, from the isolation of several new extremophilic microorganisms to the genome sequencing and analysis (both molecular analysis and expression analysis) of *Shewanella oneidensis* MR-1. This work has revealed that major metabolic innovation in MR-1 has occurred by gene duplication and adaptation within the organism, and has begun to reveal the major modes of regulation under aerobic and anaerobic conditions.

Highlights

- Use of the UV fluorescence methodology for the detection of single cells on surfaces. This new approach should allow very sensitive detection of organic carbon on surfaces and be valuable for *in situ* life detection.

- Ability to see single cells and fossils inside rocks (in three dimensions) using non-invasive CT (computerized tomographic) scanning methods

- The realization that MR-1 has had major parts of its evolutionary innovation from gene duplication and the beginning of the analysis of major pathways of anaerobic evolution

Field Trips

Mojave Desert & Mono Lake.

A major part of the work in this project is in one way or another linked to field work. For the most part, we have focused on local sites while developing the instrumentation and methodology. We have had a series of field trips to the Mojave Desert, and to the area around Mono Lake, CA, involving many members of the laboratory.

Cross Team Collaborations

For this particular project, interaction with other NAI team members has been minimal, as it is a very specific project with goals that are very focused.

That being said, several members of other teams have been to visit the lab and expressed an interest in working with us in the future when the instrumentation is ready for field work.

NASA Mission Involvement

This project interfaces very closely with upcoming 2007 and 2009 explorer/ rover missions to Mars, in the sense that it supplies both a scientific rationale for non-earthcentric *in situ* life detection, plus a definition of some approaches and instrumentation that may be valuable for these missions.

In addition, the mineralogical/organismal research approach is one that interfaces very well with the Odyssey 2001 mission that is now on its way to Mars. Dr. Nealson is a Co-I on this mission, and at least two of the JPL/Caltech NAI group will be involved as scientists to carry out data analysis.

Future Directions

- Field testing of the UV fluorescence method for life detection

- Laboratory resolution of single cells inside endolithic environments using the CT (computerized tomographic) scanning approach

- Continuation of the extremophilic environment analyses as sites for testing the non-earthcentric methods

- Continuation of the molecular analyses of *Shewanella oneidensis* with regard to the evolution of metabolism

- Definition of metal ligands as major biosignatures

- Definition of the metal/microbe interface and its changes as a biosignature

Roadmap Objectives

#3
Models for Life

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

#11
Origin of Habitable Planets

#13
Extrasolar Biomarkers

Project

Fe Isotope Biosignatures Group

Senior Project Investigator(s):
C. Johnson and B. Beard

Accomplishments

Our major efforts this year focused on a) bringing new instrumentation on line for ultra-high precision Fe isotope measurements on very small samples, b) continuing Fe isotope studies of natural samples that may have a biologic origin, and c) experimental determination of Fe isotope fractionation in minerals and fluids. A next-generation multi-collector ICP-MS was installed in a new clean lab, in part funded by NAI, and this instrument produces an unprecedented level of precision (+/- 0.05 per mil) for Fe isotope analysis on very small samples (~ 100 ng). Using our new instrumentation, we completed a survey of late Archean Banded Iron Formations, where up to 3 per mil variation in $^{56}Fe/^{54}Fe$ ratios were found, making it clear that Fe isotope variations can be preserved in the ancient sedimentary record. Work on determining Fe isotope fractionation in the lab led to the discovery of large (2.7 per mil) isotopic fractionation between Fe(III) and Fe(II) in dilute aqueous solution. Work on the hematite-Fe(III) system showed that although kinetic isotope effects can occur during rapid precipitation of hematite from solution, equilibrium isotope fractionation in this system appears to be minimal. In summary, our results demonstrated, for the first time, that Fe isotope anomalies appear to be restricted to low-temperature, aqueous environments, providing a powerful biosignature for detecting conditions on a planet that were favorable for life. In terms of accomplishments related to our original goals, installation and development of our new instrumentation progressed much faster than we had anticipated. The Fe isotope variations we documented in Banded Iron Formations were much larger than we originally thought, as were the isotopic fractionations among aqueous species. These discoveries have clearly influenced our plans for the coming year.

Highlights

- Have found that ion isotope variations appear to occur only in low-temperature sedimentary environments in the presence of water.

- In contrast to the remarkable isotopic homogeneity of the vast majority of continental crust and mantle, significant (up to 5 per mil) variations in Fe isotope compositions ($^{56}Fe/^{54}Fe$) in nature are found only in modern and ancient chemical sediments (Banded Iron Formations and Fe-Mn nodules) and modern environments where bacterially-mediated cycling of Fe occurs. In laboratory experiments, significant Fe isotope fractionation occurs between Fe(III) and Fe(II) in solution, as well as metabolic processing of Fe by Fe-reducing bacteria. These results provide a robust biosignature for conditions that are necessary for life, and further study is aimed at distinguishing biologically from abiologically processed Fe in such environments.

Field Trips

Australia and Africa.

Our fieldwork has been minimal to date; the majority of our samples have come from prior well-constrained collections from Australia and Africa from Profs. Kase Kelin (Univ. of New Mexico) and Nik Buekes (Rand Afrikaans Univ.), respectively.

Cross Team Collaborations

- Within the JPL Team

Work on Fe-reducing bacteria in the lab, Fe speciation experiments, and measurement of samples from Antarctica

- With the Pennsylvania State University Team

Planned work on ancient paleosols

- With the ASU and ARC Teams

Planned work on microbial mats

NASA Mission Involvement

We have been working with NASA/JPL scientists to explore the possibility that a miniature mass spectrometer may be developed that is capable of being launched on a lander mission for *in situ* Fe isotope analysis on other planetary bodies. The precision of the analyses must be quite high, likely requiring reference to an internal standard. Also, new methods of sample introduction must be developed (such as reduction to Fe carbonyl).

So far, we have shown that reductive processing of Fe in rocks and minerals does not fractionate Fe isotopes, which is an essential first step toward developing *in situ* analysis. Parallel work by JPL scientists is focusing on ion counting methods that will meet the required precision.

Future Directions

We have four targets for the next year:

- <u>Modern sites of active bacterial processing of Fe</u>: Our preliminary look at modern ground-water systems has shown the largest Fe isotope range yet measured, about 5 per mil, reflecting complex redox cycling of Fe by bacteria. This work should be completed in the coming year and will also include parallel experiments on Fe-reducing bacteria that build on our earlier work.

- <u>Sources for Fe in global geochemical cycles</u>: Our new, ultra-high-precision Fe isotope analytical methods have allowed us to constrain the Fe isotope composition of "igneous" Fe on the Earth and Moon to be homogeneous to within 0.05 per mil, suggesting that detrital sources of Fe to the modern or ancient oceans will also be homogenous. However, this must be confirmed through analyses of dissolved and suspended loads in river systems. In addition, a major Fe flux to the oceans may be hydrothermal vents such as “black smokers”, and these will also be studied.

- Banded Iron Formations: Based on our preliminary work, this is a very fruitful avenue for research, and we will make a major effort in this for the coming year. In addition to expanding our analysis program to a wider sample base, we will focus on obtaining pure mineral separates of very small samples.

- Meteorites: We know very little about the Fe isotope variability in meteorites, and this is essential to constrain before Fe isotope geochemistry may be applied to other planetary bodies. In addition to work on chondrite and iron meteorites, we will also work on SNC meteorites.

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#7
Extremes of Life

#8
Past and Present Life on Mars

Project

Fractionation of Transition Metal Isotopes

Senior Project Investigator(s):
A. Anbar

Accomplishments

The focus of this research is to determine if fractionations of transition metal isotopes can be used as biosignatures. We are also interested in the possible use of such fractionations to study changes in metal geochemical cycling, which could provide insight into environmental change and/or biochemical evolution. The latter applications are pursued jointly with the Harvard Team.

Following our demonstration of Fe isotope fractionation by nonbiological processes, we conducted studies to elucidate the fractionation mechanism in our experiments. We found strong evidence of an equilibrium isotope effect and no evidence of a kinetic isotope effect, although the expression of equilibrium fractionation can be kinetically inhibited under certain conditions. We also extended our work to natural samples in which biological influences are unlikely. In particular, we examined the isotopic composition of Fe in hydrothermal fluids from the deep-sea Juan de Fuca ridge. These 350°C fluids, sampled *in situ*, are sterile. We observed small (<1‰) differences in isotopic composition between fluid and mineral Fe which we attribute to nonbiological fractionation. This is good evidence that chemical processes such as identified in the laboratory are also important in nature.

We initiated a collaboration with the PSU Team into Fe isotope fractionation during leaching from minerals in the presence and absence of microbes and of metal chelating ligands. Preliminary results suggest fractionation during biogenic leaching associated with chelation. This suggests that paleosols could preserve biosignature information.

We explored fractionation between biogenic siderite and magnetite as well as between non-biogenic mineral phases.

Finally, we have begun to explore metal isotope fractionation from a theoretical standpoint, to be able to predict the magnitudes of equilibrium effects.

Highlights

- Fe isotopes can be fractionated by equilibrium chemistry at room temperatures.

- Small Fe isotope variations are seen in high-temperature hydrothermal fluids, suggesting that non-biological fractionation processes can be important in natural settings.

- Fe isotope fractionation during mineral leaching suggests a route for future biosignature research.

Cross Team Collaborations

Our primary collaborative effort in Year 3 was planning the “Mission to Early Earth” (MtEE) field excursion to Western Australia, an outgrowth of the MtEE Focus Group. This excursion will involve approximately 12 participants from at least 6 NAI Teams, as well as a participant from the Centro de Astrobiología (Spain) and from the Australian Centre for Astrobiology. Four of the participants are graduate students.

Continued development of Mo isotope measurements as a potential paleoredox tool led to submission of a new manuscript to Earth and Planetary Science Letters in Year 3.

We are in the early stages of developing a collaboration with Laurie Leshin (ASU Team) to look at Fe isotope effects in secondary minerals formed at low temperatures in meteorites.

Finally, we have begun a new collaboration with Sue Brantley (PSU Team) on Fe isotope effects during leaching.

NASA Mission Involvement

There is a need for an improved “arsenal” of biosignature tools to search for evidence of life in materials brought back to Earth in future sample return missions. Metal isotopes may be one such important biosignature tool.

Future Directions

- Continue exploration of biogenic Fe isotope effects in controlled laboratory experiments, particularly fractionation during leaching, with an eye toward biosignature applications

- Continue exploration of non-biological Fe isotope effects in controlled experiments to establish a baseline against which putative biogenic effects can be compared, with special emphasis on chemistry of hydrothermal environments

- Begin exploration of biogenic effects for other metal isotopes (e.g., Mo, Cu)

- Continue modeling efforts to predict magnitudes of equilibrium effects

Roadmap Objectives

Project

Impact Frustration and Subsequent Generation of Biologically Tenable Climates on Earth and Mars

Senior Project Investigator(s):
T. Ahrens

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

#11
Origin of Habitable Planets

Accomplishments

Mars is a water rich planet. Impact vaporization and melting studies of Mars subsurface H_2O ice and computer simulation of crater ejecta demonstrate that most of the subsurface material sampled by visible impact craters appears to have contained some 5-30% by volume of water ice, indicating that the water mass on Mars is at least 3.9 $\times 10^{-4}$ times the mass of the planet. This compares to 2.8 $\times 10^{-4}$ in the case of the Earth.

Our analysis of the global acidification that occurred at the K/T boundary indicates that ~1 $\times 10^{17}$ equivalents of strong acid was deposited on the continents. We conclude that North America was preferably leached with greater densities of acid from prompt fallout of H_2SO_4 aerosol.

Shock temperatures measured in crystal calcite in the 95-160 GPa range yield radiative temperatures of 3300° to 5400° K. Calculations indicate that the temperatures are 400° to 1350° K lower than predictions indicating massive bond breakage under pressure. Calcite therefore decomposes upon unloading from only 18 GPa.

Highlights

Have demonstrated that most of the subsurface material sampled by visible impact craters appears to have contained some 5-30% by volume of water ice, indicating that the water mass on Mars is at least 3.9 x10^{-4} times the mass of the planet.

NASA Mission Involvement

The present analysis of Mars craters supports the Mars Global Surveyor Mission. This mission was launched in 1996, entering Mars orbit in September 1997 to study the red planet.

Future Directions

- Complete comparison of theoretical and observed ejecta blankets on Mars and refine water estimates
- Conduct speciation of impact vapor studies for minerals

Laboratory/Observational Studies of Biogenic Greenhouse Gases on the Early Earth

Project

Senior Project Investigator(s):
G. Blake

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

#12
Effects of Climate & Geology on Habitability

Accomplishments

Our research centers on isotopic studies of biogenic greenhouse gases as evidence for life on the large scale, appropriate for remote sensing approaches and on global biogeochemical investigations. Our early work in this area was concerned with the early and present biogeochemical evolution of the Earth, but over the last year we have focused strongly on developing new tools for the exploration of Mars and other extraterrestrial environments. Our particular tracer of emphasis is stable isotope fractionation, and we seek to understand whether or not this signature can be reliably detected in extraterrestrial solar system bodies and exoplanetary systems. We have now characterized in detail the fractionation induced by the photolysis of N_2O first predicted by Prof. Yung using a combination of non-linear spectroscopic light sources and mass spectrometry or FTIR spectroscopy.

Neither of these approaches is compatible with the stringent space and weight requirements of *in situ* planetary exploration strategies. We are therefore developing new technologies that should enable the *in situ* measurement of important radiatively and biogenically active gases such as carbon dioxide, carbon monoxide, water, methane, nitrous oxide, and hydrogen sulfide to very high precision in order to examine the atmospheric dynamics and potential biogeochemistry on Mars, Titan, and other solar system bodies. Specifically, we are using laser diodes and sensors to image infrared laser induced fluorescence (IR-LIF). The support from NAI was used to conduct pivotal modeling tests of this approach, which has resulted in a successful proposal to the NASA PIDDP program . PIDDP now provides the larger funding base to actually fabricate these new tools through a three-year program to combine microchip lasers with state-of-the-art HgCdTe detectors to investigate the sensitivity of IR-LIF under realistic planetary conditions, to optimize the optical pumping and filtering schemes for important species, and to apply the spectrometer to the non-destructive measurement of stable isotopes in a variety of test samples. These studies form the necessary precursors to the development of compact, lightweight stable isotope/trace gas sensors for future planetary missions. We will continue to work on modeling of the potential biological activity on Mars and the early Earth, and on the isotopic signatures of various model biospheres. This research is groundbreaking, and it provides the necessary context for IR-LIF to be used as a robust diagnostic of extant or extinct (through its sampling of gases trapped in martian soils and ice caps) biota on and underneath planetary surfaces.

Cross Team Collaborations

A goal for next year is to participate in the analysis of the stable isotope fractionation of nitrous oxide with both the Yung group (JPL Team) and the group of Prof. Paul Wennberg, Professor of Planetary Science and Atmospheric Chemistry at Caltech (JPL Team).

NASA Mission Involvement

Our prototyping and technique development work is aimed at future participation in the Mars exploration program, specifically landers or other *in situ* probes. The development time frame is such that deployment will not be possible until after the 2007 MER (Mars Explorer Rover) launch opportunity.

Future Directions

Our research this past year has resulted in initial measurements of the stable isotopes of water. This coming year our goals are to:

- Determine the optimum laser pumping and detection wavelengths for carbon dioxide and methane

- Construct a miniaturized brassboard instrument with dedicated lasers and detectors for water, carbon dioxide, and methane. We also plan to make atmospheric measurements in the Los Angeles basin using this instrument.

• Participate in the analysis of the stable isotope fractionation of nitrous oxide with the Yung group and with the group of Paul Wennberg, Professor of Planetary Science and Atmospheric Chemistry at Caltech

Mineralogical Biosignatures Formed At and Near the Cell-Solution Interface

Project

Senior Project Investigator(s):
J. Banfield and W. Barker

Roadmap Objectives

#13
Extrasolar Biomarkers

Accomplishments

The primary goal of this project is to learn how microbial metabolism and metal ion-polymer interactions lead to production of novel assemblages interpretable as biosignatures. The first focus of our research has been development of laboratory biomineralization systems that simulate reactions at hydrophobic-hydrophilic interfaces. Our experimental design involves assembly of functionalized surfaces at boundaries between organic and aqueous solutions and their mineralization by iron oxyhydroxide, iron silicate, and iron phosphate minerals. The goals of this work are to develop an understanding of how organized polymers template nanocrystal particle growth. We have conducted experiments using alginic acids that resulted in extensive mineralization of oriented assemblies of nanoparticles, simulating biosignatures associated with neutrophilic iron-oxidizing bacteria. Using DADMAC, a spiral-shaped polymer that carries positively charged N-containing groups, we have induced mineralization of tubes and produced biomimetic iron silicate phases.

The second goal involves study of the structure, microstructure, form, and organization of nanoparticles of metal sulfides and oxides formed extracellularly as the result of enzyme-mediated redox reactions. We have documented extremely small (<2nm particles) generated through bacterial iron oxidation and uranium and sulfate reduction. Under some conditions, the <2nm particles aggregate to form larger crystals that assemble into novel morphologies predicted using the Debye-Smoluchowski equation. The oriented aggregation-based crystal growth pathway also leads to a generation of biomaterials containing point, line, and planar (twin) defects. Understanding of the evolution of these highly reactive nanomaterials over time, a topic to be studied in the future, is critical to evaluation of the fate of biosignature. NASA funding has allowed incorporation of a biosignature component in the studies of several students whose primary support is provided by other projects with other goals. Progress over the past 12 months has exceeded expectation.

Highlights

- Two papers were published in *Science* in 2000. The first demonstrated, for the first time, that iron oxyhydroxide nanoparticles produced by neutrophilic iron-oxidizing bacteria coarsen via oriented aggregation and that this pathway leads to microstructural characteristics that have important implications for subsequent reactivity. The second paper documents the formation of micron-scale, spherical aggregates of nanophase ZnS particles formed by sulfate-reducing bacteria in groundwater.

- An additional paper is in preparation on particle size and mobility of biologically induced uraninite (UO_2), and flocculation-assembly-based growth of nanophase uraninite.

Field Trips

Tennyson, Wisconsin.

Fieldwork has been carried out at Tennyson, WI. Many sampling trips have been conducted in order to obtain materials for this, and related projects. The team of researchers is affiliated with UW Madison.

Cross Team Collaborations

A collaboration with Dr. David Emerson represents a link to the CIW Team. Discussions with other Carnegie team members preliminary to isotope analysis have not yet led to a collaborative experiment.

NASA Mission Involvement

This project is directly relevant to the search for evidence of current or past life on Mars.

Most of the materials sampled on Mars or returned to Earth for analysis will be minerals. Study of the nature of mineralogical biosignatures and their interpretation is still in its infancy. Mineralogical biosignatures represent an important frontier for NASA.

Future Directions

- Experimental analysis of Fe-silicate and Al-silicate biomineralization

- Determination of relationships between biomineral phase and polymer chemistry

- Evaluation of the diversity of microbes associated with Fe biomineralization at neutral pH and of their biosignatures

- Experimental studies of the fate of nanocrystalline biomineralization products

- Publication of recent results

Organic Molecules as Biosignatures

Project

Roadmap Objectives

Senior Project Investigator(s):
G. McDonald

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

Accomplishments

We have continued our study of the temperature and environmental history of Siberian permafrost core samples, from which viable bacteria apparently several million years in age have been isolated. Our data clearly show that the rate of aspartic acid racemization below approximately 5 m, or about 20,000 years in age, has been consistent with the measured temperature of the permafrost. This indicates that there has been little or no metabolic activity in the organisms below 5 m. In contrast, the organisms present from the surface down to 5 m have clearly warmed sufficiently to metabolize D-amino acids and thus partially reset the racemization "clock". This study demonstrates the usefulness of amino acid racemization as an *in situ* indicator of metabolic activity in cold environments.

We are also still studying the fate of organic material produced by cryptoendolithic organisms colonizing Antarctic dry valley sandstone. Soluble organics, including amino acids, are transported into the interior of the rock by small flows of snowmelt. The amino acids in the rock interior show significant amounts of racemization, suggesting that they have been in the interior for long periods of time. We are currently comparing the amount of organic carbon lost by the cryptoendolithic community into the rock interior with the amount used in the production and secretion of iron-mobilizing agents. This will allow us to more closely constrain and better understand the metabolic cost that these organisms must pay in order to survive in the dry valley environment.

We have also used experimentally determined rate constants for aspartic acid racemization in Siberian permafrost sediments, combined with temperature vs. latitude data from Mars Global Surveyor, to calculate the range of latitudes on Mars over which a biological chiral amino acid signature would survive throughout the planet's history. We find that such a signature should be detectable in samples collected at latitudes poleward of approximately 30º and from depths of at least 1-2 meters.

Highlights

We are developing the use of amino acid racemization as an *in situ* indicator of metabolic activity in microbial communities from cold environments. These data are allowing us to begin to place limits on the maximum survival time of metabolically inactive microorganisms.

Field Trips

Siberia.

Siberian permafrost samples were collected by David Gilichinsky and colleagues from the Russian Academy of Sciences.

Antarctica.

Antarctic sandstone samples were obtained courtesy of E. Imre Friedmann, from Florida State University and NASA Ames Research Center.

Cross Team Collaborations

James Scott and Marilyn Fogel of the Carnegie Institution of Washington (CIW) Team have determined stable carbon isotope ratios for amino acids isolated from the Antarctic cryptoendolithic communities described above. These data are still being evaluated in the context of our overall project.

NASA Mission Involvement

Permafrost and cryptoendolithic microbial communities are both analogs of possible past or present biota on Mars.

The lessons we learn about the interactions of these organisms (from both their mineral matrices and the chemical traces they leave behind) are relevant for Mars exploration. This background is critical for design of *in situ* astrobiology instrumentation for Mars, as well as in planning for the analysis of returned samples.

Future Directions

- Measure extent of aspartic acid racemization in Siberian permafrost samples from deeper levels (>40 kyr).
- Extend racemization studies to other ancient biogeological samples.
- Analyze additional colonized Antarctic sandstone samples at finer vertical resolution.

Education & Public Outreach

Lectures

Gene McDonald gave a talk entitled, "Astrobiology", to the Continuing Learning Experience program at California State University at Fullerton.
Presentation on astrobiology by Gene McDonald to Jack R. Howard of the Science Reporting Institute at Caltech in Pasadena, California.
Ken Nealson, presented "Searching for Life in the Universe: Lessons from the Earth" at the Capital Science Lectures, Washington, DC., and at the American Astronomical Society annual meeting, Pasadena California.

Science Fair Judging

Michael Storrie-Lombardi, Pamela Conrad, Alexandre Tsapin, Rohit Bhartia of JPL were judges for the Alhambra High School science fair.

Skymobile

Skymobile is a mobile interactive exhibit about planetary exploration and the search for life in the universe that travels to elementary schools. Members of the JPL team served as advisors to the staff of Los Angeles County Natural History Museum on the Skymobile education project.

Publications

Back to Table of Contents

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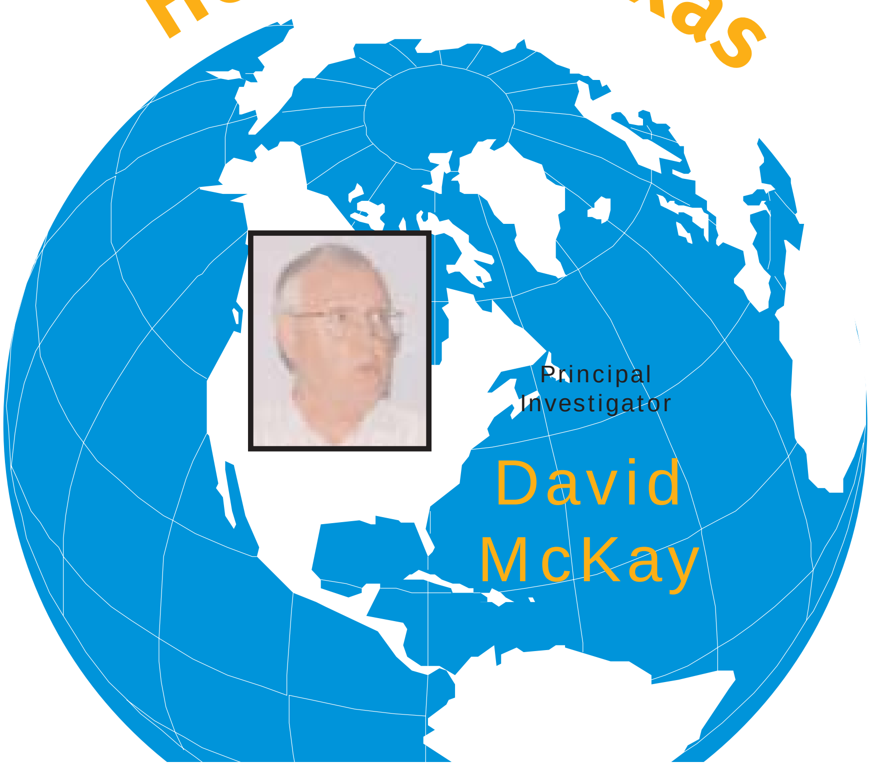
Back to Table of Contents

Year 3

Johnson Space Center

NASA Astrobiology Institute

Houston, Texas



Johnson Space Center Team Members

Carlton C. Allen
Johnson Space Center

Recep Avci
Montana State University

Dennis A. Bazylinski
Iowa State University

Mary Sue Bell
Johnson Space Center

Carlos Briones Llorente
Centro de Astrobiologia

Monica Byrne
Wellesley College

Simon J. Clemett
Lockheed Martin

George Flynn
SUNY, Plattsburgh

George E. Fox
University of Houston

Everett K. Gibson
Johnson Space Center

Larry Hersman
Los Alamos National Laboratory

Nancy Hinman
University of Montana

Richard Hoover
Marshall Space Flight Center

Thomas Kieft
New Mexico Institute of Mining and Technology

John Lisle
Lockheed Martin

Teresa Longazo
Lockheed Martin

David S. McKay
Johnson Space Center

Tom Moore
University of Montana

Penny A. Morris-Smith
University of Houston

James Papike
University of New Mexico

Victor Parro Garcia
Centro de Astrobiologia

Juan Perez Mercader
Centro de Astrobiologia

Seth Pincus
Louisiana State University

Luke Probst
Bemidji State University

Chris S. Romanek
Savannah River Ecology lab

Rachel Schelble
University of New Mexico

Craig Schwandt
Johnson Space Center

Mary Schweitzer
Montana State University

Gordon Southam
The University of Western Ontario

M. N. Spilde
University of New Mexico

Verena Starke
Philipps-University Marburg

Andrew Steele
University of Portsmouth

Kathie Thomas-Keprta
Lockheed Martin

Jan Toporski
Portsmouth University (UK)

Allan H. Treiman
Lunar And Planetary Institute

H. Vali
McGill University

Norman Wainwright
Marine Biological Laboratory

David Warmflash
Johnson Space Center

Susan J. Wentworth
Lockheed Martin

Frances Westall
Lunar And Planetary Institute

Executive Summary

Accomplishments

The JSC Astrobiology Institute Team is very diverse and has about as many non-JSC members as it does JSC members. Most of the JSC team consists of contractor scientists, student interns, an NAI postdoctoral fellow, and scientists from nearby institutions. Our team represents a variety of disciplines including geology, mineralogy, microbial physiology, geochemistry, and planetology.

The uniting theme for the JSC team is sample and material analysis to provide characterization data on terrestrial samples, astromaterials, and experimental samples. The types of data sought are usually features related to microbial life. Such features include morphology of living microbes and their surroundings including biofilms, as well as the morphology of fossilized forms of these living biota. In addition to morphology, chemistry and mineralogy of microbial produced features and fossilized microbes are of great interest to us. We typically use probe instruments Scanning Electron Microscope (SEM), Transmission Electron Microscope (TEM), electron microprobes, Time-of-flight Secondary Ion Mass Spectrometer (TOF-SIMs), double laser beam mass spectrometers) to acquire information on living or dead microbial life. The overall goal is to relate chemistry, mineralogy, atomic structure, morphology, spectral interactions, and other properties for specific types of features so that multiple data can be used to characterize or fingerprint life in all its forms (including ancient fossils and bioassisted mineral precipitates). But we also strive to understand the processes that create the characteristics of life, the processes that alter or fossilize life, and the processes that produce and preserve interaction between life forms and rocks or minerals. Fingerprints of life are valuable for themselves, but they may also contain the history of that life and its environment over time.

The ability to identify the presence of life or the former presence of life has been taken for granted in many types of terrestrial samples. However, as we look more and more at astromaterials from beyond Earth (and also take a fresh look at archean samples on Earth), the scientific community has realized that it is not always obvious or easy to determine whether life was or is present in the rocks, minerals, soils, and fluids of Earth or another planet.

Consequently, our overall goal is to develop better techniques for detecting and understanding life. It will be absolutely necessary to do this for returned Mars samples, but these techniques should first be well tested on terrestrial samples, and should also be applied to other types of samples as well: meteorites, cosmic dust, cometary dust, and samples from various satellites and small bodies. Because of the extreme interest in the possibility of life elsewhere, we must develop strong criteria for detecting it and certifying it. That is the current overarching goal of the JSC team. That may also become the core objective for all of NASA in the coming decade or two.

Highlights

Science Projects

We have subdivided our work into seven projects: Below are listed the projects with several highlights from the past year's activities.

(1) New Technique Development and Application

- Hopanes, a reliable established class of chemical biomarker, have been detected for the first time by TOF-SIMS analysis of fossils.

- Direct identification of a variety of organic compounds was made on fossil bacteria at the micrometer scale.

- Use of multiple fluorescent probes may provide a wealth of data on wild bacteria that cannot be cultured.

- New analyses of Nakhla Mars meteorite by double laser spectrometry shows the presence of complex organics.

(2) Mars Immunoassay Life Detection Instrument (MILDI) Development

- New technique using antibodies and state-of-the-art array approach that may be used to search for life on Mars on robotic missions.

- This Mars instrument design uses the same principle as a home pregnancy test.

- The instrument will be developed in time for the 2007 Mars lander mission.

(3) Hematite on Mars: Possible Biogenic Origin

- Hematite is present in a variety of sedimentary rock types formed at low temperatures.

- Iron oxides including hematite are known to fossilize and preserve microbes and biofilms.

- Hematite is present as a secondary alteration mineral formed on Mars in some of the Mars meteorites.

- Based on terrestrial examples, the martian hematite site identified by the Thermal Emission Spectrometer (TES) on Mars Global Surveyor (MGS) is an excellent candidate site for a search for life.

(4) Cold Regions Microbial Ecology and Biomarkers

- A variety of microbes and viruses thrive in permanently ice-covered Antarctic lakes, even though the ice filters out more than 98% of the light.

- “Hot” (2°C) springs emerging from the upper surface of some Canadian Arctic glaciers contain microbial life.

(5) Carbonate Globules in Igneous Rocks

- Iron-rich carbonate globules in Mars meteorite ALH84001 appear to have counterparts in igneous rocks on Earth.

- Carbonate globules in igneous rocks may be a potential recorders of the history of the hydrosphere, the atmosphere, and possibly life on the Earth and Mars.

- Carbonate globules in igneous rocks are extremely complex and are often associated with secondary silica-rich phases—preliminary evidence suggests that this silica phase forms at relatively low temperatures and is not a melt glass.

(6) Mineral Biomarkers

- Tiny magnetite in ALH84001 is confirmed as indistinguishable from magnetite made by bacteria.

- Magnetite in ALH84001 strongly supports the early life on Mars hypothesis.

- Manganese minerals formed in caves with the help of bacteria may provide possible biomarkers for use on Mars.

(7) Morphologic Biomarkers and Microbial Ecology

- New morphologies were found in Mars meteorites which may be the fossilized remains of microbes (but more supporting data is needed).

- Some morphologic biomarkers may be distinctive enough to use without supporting chemistry and mineralogy, but many require multiple types of supporting data to be accepted as true biomarkers.

- 2.1 Ga old iron-rich rocks on Earth have a variety of features showing complex microbial morphologies which may be related to the preserved mineralogy.

- Hydrogen peroxide in Yellowstone hot springs is produced by UV sunlight and removed by microbes. This may have implications for Mars where it has been proposed that UV light produces hydrogen peroxide which has destroyed organic compounds and life near the surface. Perhaps martian microbes also learned to destroy excessive levels of hydrogen peroxide in near-surface moisture.

- Unusual phosphate microfossils were found in Mongolian rocks.

(8) Rock Varnish

- Desert varnish on rocks from southwestern Arizona showed evidence for microbial life.

• Some rocks in Viking and Pathfinder imaging appear to have a smooth shiny surface with properties similar to terrestrial rock varnish.

• Varnish on Mars rocks may be a location for either fossil life, extant life, or both and this varnish should be a target for future missions and sample return.

Additional details on each of these projects can be found in the attached report or the published papers and abstracts.

Cross Team Collaboration

We have put together two research teams from Historically Black and Hispanic Universities and have helped them get independent funding through proposals. We now have an Astrobiology Group at New Mexico Highlands University, which is a subunit of the JSC Astrobiology Institute. In addition, a member of our local team (Penny Morris-Smith) has been awarded a significant grant to incorporate minority academic people from both Texas Southern University and the University of Houston (downtown campus) into some of our projects. They serve as interns, visiting faculty, and graduate students. We will also work with the Houston Museum of Natural History on this project. In summary, we have added three universities to our active group, all specifically in astrobiology. We have raised more than half a million dollars from other sources to support these efforts.

Currently, we also have a Memorandum of Understanding (MOU) in negotiation to work with a group at the Geological Survey of Canada specifically on astrobiology topics, generally focused on biomarkers. We anticipate this MOU will be signed this calendar year.

As indicated in our current science project reports, we are now actively working with a number of universities. This was documented also in previous progress reports. These collaborations have proven to be valuable. They have provided our team with considerable expertise (which we originally lacked), specifically in microbiology, genomics, microbial ecology, plus field and laboratory procedures for sampling, culturing, preserving, and working with microbes. We now have a microbial ecologist/physiologist on our immediate JSC Team (John Lisle), so that we are better able to interact with other NAI teams, which mainly consist of biologists.

We have established a working relationship with the Centro de Astrobiologĺa (Madrid, Spain) for work on the MILDI (Mars Immunoassay Life Detection Instrument) project. They are helping develop the array technology, and we are working on the antibody-antigen reactions. We will both participate in final design and in the test program. They will be partners on any flight hardware that we are successful in getting onto a mission.

We plan to establish a strong and active partnership with Carnegie Institution of Washington when Andrew Steele relocates there and gets his research program going.

Overall, we believe we have made considerable progress this year. We look forward to more collaboration with other NAI Members in the coming year.

Roadmap Objectives

#6
Microbial Ecology

#8
Past and Present Life on Mars

Project

Biogenic Mineral Weathering

Senior Project Investigator(s):
S. Wentworth

Accomplishments

This project now has three parts. First is documentation of weathering features on terrestrial minerals from various environments. Next is an experimental program which produces abiologic weathering on mineral surfaces, produces biologically assisted weathering, and compares the two. The biologically assisted weathering experiments also have a sterile control group. The third part of this project is to document likely and potential weathering features on Mars meteorite samples and to compare them to the data from the terrestrially weathered samples and the experimental samples. For the Mars meteorite samples, care must be taken to try to separate possible terrestrial weathering from martian weathering. This can be done to some extent using the fusion crust as a stratigraphic time marker separating terrestrial effects from Mars effects. Other techniques for doing this may include other microstratigraphy features, and hydrogen and carbon isotopic analysis. The overall goal is to understand mineral weathering features related to microbial activity, and to separate them from strictly abiotic weathering features.

Highlights

- Complex textures in Martian meteorites suggest water-based weathering.

- Biogenic weathering may produce microetch textures not formed in abiogenic weathering; if confirmed, these textures may make a useful biomarker for Mars samples.

Field Trips

No specific expeditions are necessary, but a variety of samples from various environments will help with the database.

Cross Team Collaborations

We currently have no collaborative research on this topic with other NAI team members. However, we welcome the participation of interested scientists or students.

NASA Mission Involvement

This project mainly applies to Mars meteorites and will be most useful when Mars samples are returned by the 2011 Mars robotic mission. It will help us identify the effects of weathering by either ancient or extant microbes.

It is conceivable that robotic *in situ* analysis instruments (such as AFM (audio-frequency modulation) or SEM (scanning electron microscopy)) on the Mars 2007 lander or 2007 Scout missions will have enough resolution to detect fine-scale weathering textures and document enough to compare to our database of weathering textures.

Future Directions

- Continue with our experimental project to compare biogenic weathering, sterile controls, and strictly abiogenic weathering.
- Develop new morphologic criteria, if any, which are distinctive for biogenic weathering textures.
- Catalog existing morphologic features of each type of weathering.
- Determine chemical changes on the micrometer and submicrometer level associated with each type of weathering.
- Catalog weathering features in ALH84001, Nakhla, and other Mars meteorites.

Carbonate Globules in Igneous Rocks

Project

Senior Project Investigator(s):
D. McKay

Roadmap Objectives

#6
Microbial Ecology

#8
Past and Present Life on Mars

#12
Effects of Climate & Geology on Habitability

#17
Planetary Protection

Accomplishments

This project was inspired by the carbonate globules in martian meteorite ALH84001, which is an igneous rock in which carbonates have formed on Mars in cracks and holes within the rock. These carbonates have generated two major controversies: (1) did the carbonates form at high temperatures thereby excluding any possible biological role in their formation? (2) if the carbonates formed at low temperatures, did biology play any part?

We have begun detailed studies of carbonate globules in Columbia River Basalt (CRB) samples from Spokane, WA, and in basaltic rocks and ultrabasic igneous inclusions from Spitzbergen, Norway. Both samples contain Fe-carbonate (siderite) as well as other compositions of carbonate. In both samples the carbonates are closely associated with silica-rich veins and thin layers having somewhat different chemical compositions from layer to layer. Microstratigraphic relationships prove that some, and possibly all, of the silica in the CRB samples formed after the carbonate was crystallized. The lack of alteration or decomposition of the siderite adjacent carbonate shows that the silica was emplaced at relatively low temperatures, likely as an aqueous deposit of a gel.The close association of the silica gel with the carbonate globules suggests that the carbonate globules also formed at low temperatures, at least in the CRB samples. Some of both the CRB globules and the Spitzbergan globules are radially zoned in texture and composition, showing some similarity to the ALH84001 carbonate globules.

An unexpected result for the first two examples studied is the close association of carbonate globules with silica-rich glass or gel-like microlayers or veins. The glass or gel-like silica shows a complex surface morphology. Work is underway to search for organic particulates or residue in these two terrestrial samples and to use the results to influence additional work on the ALH84001 samples. Even without organic residue or microfossil morphology, the presence of secondary carbonates in igneous rocks has preserved a history of likely hydrothermal or low temperature alteration which adds a new dimension to the preserved volcanic history of the rocks; the rocks have recorded information about the hydrosphere and atmosphere. A preliminary literature search indicates that secondary carbonate globules in igneous rocks on Earth may be much more common than realized. For example, carbonates are often found in vesicles in suboceanic basalts. Carbonates readily form in cavities in desert surface rocks, including at least one documented meteorite.

With one example already from Mars and with multiple terrestrial examples, the possibility exists that many igneous rocks on Mars (mainly basalts and andesites) may contain secondary carbonate globules formed at relatively low temperatures. Such carbonates may record a history of near surface aqueous activity on Mars. The carbonates may also be potential locations of fossils or even extant life. Consequently they constitute a possible new environment for life in the solar system, particularly for bodies that are dominated by igneous rocks.

Highlights

- Iron-rich carbonate globules in Mars meteorite ALH84001 appear to have counterparts in igneous rocks on Earth.

- Carbonate globules in igneous rocks may be a potential recorders of the history of the hydrosphere, the atmosphere, and possibly life on the Earth and Mars.

- Carbonate globules in igneous rocks are extremely complex and are often associated with secondary silica-rich phases—preliminary evidence suggests that this silica phase forms at relatively low temperatures and is not a melt glass.

Field Trips

A field trip to several volcanic terrains would be desirable. Possible sites include the Columbia River basalt flows, the Craters of the Moon, Idaho, the basalt flows of south central Arizona, and the preserved basalt flows of Knippa, Texas. The objective is to find and document additional carbonate globules, determine the relationship of globules to the flow stratigraphy, the ground water table, and the weathering profile. In addition, we intend to acquire selected suboceanic basalts containing carbonate globules.

Cross Team Collaborations

We propose to work with the University of Washington Team and the Arizona State Team on this research project.

NASA Mission Involvement

Carbonate globules in igneous rocks have possible value in the search for life on Mars. Awareness of this value may influence the 2003 MER (Mars Exploration Rover) mission. Igneous terrains cannot be ruled out as possible clues to Mars water history and the history of possible life. The possibility that secondary carbonate globules have formed in typical basalts or andesites would make igneous terrains fair game in the search for life.

It is too late to influence instrument selection for the Mars 2003 mission. However, consideration of carbonate globules in igneous rocks might influence the final site selection, the operations, and the type of data collected for the 2003 Mars mission. Data collection might include the search for globules in basalts, as well as spectral data collection from globules, etc. In addition, the possibility of carbonate globules may influence the site selection and instrument choice for the Mars 2007 mission and for the types of rocks collected on the Mars 2011 sample return. Clearly, additional studies of carbonate globules in igneous rocks may contribute to future science requirements for Mars missions.

This project is key to understanding possible early life on Mars and therefore influences the design of all Mars robotic missions. These include the 2003 Mars site selection for rovers, the Mars 2007 *in situ* lander mission, and the Mars 2011 sample return mission. In addition, our research project will influence the 2007 Mars Scout missions. The presence of magnetotactic bacteria on early Mars has great implications for site selection, sample collection strategy, instrument selection for *in situ* analysis, and confirming early oceans.

Future Directions

- Determine whether CRB (Columbia River Basalt) and Spitzbergen globules contain any evidence for biogenic processes contemporaneous with the formation of the carbonates and the low-temperature silica.

- Search for additional terrestrial samples containing carbonate globules in igneous rocks.

- Relate this work to ALH84001 and Nakhla Mars meteorites, both of which contain secondary carbonate features.

- Search for and identify organic compounds in igneous rock carbonates.

Roadmap Objectives

Project

Cold Regions: Microbial Ecology and Biomarkers

Senior Project Investigator(s):
J. Lisle

#7
Extremes of Life

#8
Past and Present Life on Mars

Accomplishments

This project investigates microbial ecology and the production of life in cold regions. Progress includes the documentation and description of microbial life in perennially ice covered lakes in McMurdo Dry Valleys of Antarctica. These lakes were found to have active and diverse microbial ecosystems The lakes are highly stratified in regard to supersaturation of gases (O_2, NO_2, H_2S), contain strong salinity gradients, and a 20 degree range of temperatures from top to bottom. The ice cover reduces the flux of light by more than 98%. In spite of these harsh conditions, bacteria, lysogenic bacteria, and viruses are present in all of the samples and are similar in abundance to that found in open ocean water of more temperate conditions.

In a related study, water emerging from springs in glaciers in the Canadian Arctic has been identified as containing both biofilms and bacterial cells. Deposits of yellow and white minerals are formed on the surface as the water evaporates. These deposits include gypsum and sulfur. The water is no more than 1-2°C and the microbes may be growing in the plumbing system of the glacier interior. Such occurrences in terrestrial glaciers may be mirrored on Mars during periods when the polar ice caps are melting or retreating.

In yet another facet of this project, we are studying psychrophilic microorganisms collected from the Fox permafrost tunnel in Alaska, piedmont glaciers and tidewater glaciers in Alaska, and the Vostok core in Antarctica. Samples include a variety of fungi, bacteria, and cyanobacteria, some of which have been cultured.

Highlights

- A variety of microbes and viruses thrive in permanently ice-covered Antarctic lakes, even though the ice filters out more than 98% of the light.

- "Hot" (2°C) springs emerging from the upper surface of some Canadian Arctic glaciers contain microbial life.

Field Trips

Antarctica, ongoing fieldwork

This project is supported by ongoing fieldwork in Antarctica. John Lisle has been going there for several years and plans on yearly field expeditions.

Cross Team Collaborations

We currently do not have extensive NAI cross team collaboration. However, we would welcome additional members in our cold regions studies. We are currently collaborating with Montana State University researchers, who collaborate with the ARC Team on their Early Microbial Systems project.

NASA Mission Involvement

The purpose of this project is to examine cold region life on Earth and use the information to help us choose the optimum sites for searching for life on Mars.

Mars contains ice at the poles with glacier-like features, and many morphologic features on Mars have been interpreted as glacial deposits and modifications of the landscape. Consequently, knowing where to search for life relative to glacial geology and morphology, either past or present, may help with site selection on the Mars 2003 mission, the 2007 lander mission, and the 2007 Scout missions. In addition, Mars sample collection in 2011 will be highly influenced by likely spots for extant or past life.

Cold region locations on Earth will give us insight into the best places for exploring Mars. For example, if Mars had lakes, as most geologists believe, the frozen-over lakes may have behaved similarly to the Earth analogs in Antarctica. We would therefore know how to locate them on Mars, how to sample them, and what to look for in terms of sediments, biofilms, fossilized microbes, and fossilized viruses.

Future Directions

- Continue study of microbes and viruses in Antarctic permanently ice-covered lakes.

- Continue collection and study of microorganisms from Vostock core, Antarctica.

- Characterize the microbial populations from these sites.

- Sample the lake floor for microbial mats and deposits of microbes and viruses.

- Continue analyzing products of spring eruptions in Arctic glaciers.

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#8
Past and Present Life on Mars

#14
Ecosystem Response to Rapid Environmental Change

Project

Hematite on Mars: Possible Biogenic Origin

Senior Project Investigator(s):
C. Allen

Accomplishments

Our approach to the investigation of hematite on Mars consists of three parts. First, we are evaluating literature data on the occurrence of low temperature, aqueous deposited hematite. Such deposits include some banded iron formations (BIFs), as well as some other iron-rich deposits in the geologic record. Next, we are undertaking a laboratory study of samples containing iron oxides to document the hematite occurrences, the chemistry of the rock and its oxides, and the presence or absence of morphologic biomarkers such as fossil microbes, filaments, or biofilm. Finally, we are apply the results to an evaluation of hematite sites on Mars as identified by the Mars Global Surveyor (MGS). This project is also related to our Rock Varnish project which is identifying iron and manganese oxides and hydroxides associated with alteration zones or coatings on rocks.

Highlights

- Hematite is present in a variety of sedimentary rock types formed at low temperatures.

- Iron oxides including hematite are known to fossilize and preserve microbes and biofilms.

- The 2.1 Ga Gunflint Formation has clear evidence for preserved microbes associated with the precipitated iron oxides.

- Hematite is present as a secondary alteration mineral formed on Mars in some of the Mars meteorites.

- Based on terrestrial examples, the Martian hematite site identified by the Thermal Emission Spectrometer (TES) on Mars Global Surveyor is an excellent candidate site for a search for life.

Field Trips

A field trip to iron-rich sediments such as BIFs is desirable.

Cross Team Collaborations

We currently have no specific NAI cross-team collaborations for this project, but we would welcome the participation of other researchers.

NASA Mission Involvement

Understanding the occurrence of hematite in various terrestrial sediments and low temperature deposits will provide the basis for targeting Mars robotic missions to hematite sites already identified by TES (Thermal Emission Spectrometry).

The Mars 2003 landers are excellent examples of missions that may be targeted to one or more hematite sites. As we understand the morphology, chemistry, and key characteristics of terrestrial hematite occurrences associated with biogenic activity, we will be more intelligently able to design instruments, procedures, and operations to be aimed at detecting evidence for past life on Mars.

Consequently, the work of this project will feed directly into Mars mission site selection and operational planning for 2003. Depending on the Mars 2003 mission results, the Mars 2007 lander site selection, analysis instrument selection, and operation planning may also be influenced by the results of this project. Similarly, the Mars 2011 sample return mission may be directed at a site containing iron oxides with the expectation that biology may be preserved in such deposits.

Future Directions

- Analyze and describe additional samples of terrestrial rocks that have formed hematite at relatively low temperatures.

- Determine whether biological activity played a role in the hematite formation.

- Provide new data and advise on the hematite sites as possible landing sites for the 2003 rovers.

Project

Immunoassay Life Detection Test

NAI Postdoctoral Fellow: D. Warmflash
Advisor: D. McKay

Roadmap Objectives

#8
Past and Present Life on Mars

Accomplishments

This project focuses on the development of an instrument that will employ immunoassay to detect a wide range of chemical compounds in extraterrestrial environments. The goal for the current funded period of the team as a whole is to demonstrate by the middle of 2002 that the concept will work in a prototype instrument. We will need to show not only that immunoassay is highly sensitive and specific for the detection of various bio-organic molecules but also that it is useful for determining whether a sample of extraterrestrial material (such as the regolith on Mars) contains these compounds. So far, we have collected various researchers and investigated technologies and determined that it is possible to build a small array instrument that can detect several thousand compounds using immunoassay. We have formed important collaborations, created a list of potential antibodies - some of which are in our possession, others that we need to buy or acquire through collaborations – that we now need to evaluate individually to determine usefulness in the instrument. I have participated in seeking out these antibodies and have put together a literature survey that focuses specifically on those antibodies against DNA, RNA, individual nucleotides and related compounds.

Since my NAI/NRC funding began in January 29, 2001, I have done the following: I have helped to form collaborations and strategies for instrument development with the researchers in Spain, listed above as well as collaboration with Prof. Ronald Nezlin of the Weizmann Institute in Israel. As part of my participation in the general strategy for the project, I met with Andrew Steele, Mary Schweitzer and Seth Pincus at Montana State University in April. Connected with the international collaborations, I was in Spain with Andrew Steele and Mary Schweitzer in February and in Israel with Mary Schweitzer in late February/early March. I have also attended three conferences: the Lunar and Planetary Science Conference in March, the NAI General Meeting in Washington in April and the Aerospace Medical Association scientific meeting in May. In general, the period from the beginning of my funding to the present time has served to allow me to seek out potential collaborators necessary for our project and sort through relevant literature in order to begin laboratory testing of antibody assays. I am currently in the process of determining materials and reagents that I will need to begin conducting laboratory work in conjunction with George Fox and I expect that materials will be ready in July.

Highlights

- Together with collaborators Drs. Andrew Steele and Mary Schweitzer, I formed a collaboration with Dr. Juan Perez Mercader and his team at the Centro de Astrobiología (CAB) in Spain. The group in Spain has array technology that is essential to the development of our instrument.

- I have completed a literature survey focussing on anti-DNA and anti-RNA antibodies and related antibodies that includes listings of antibodies that we can potentially use as part of assays in our instrument.

Cross Team Collaborations

We are working on this project in collaboration with investigators at the Centro de AstrobiologÍa (Madrid, Spain), an NAI international partner.

Additionally, we collaborate with Andrew Steele, who will begin a position at the Carnegie Institution of Washington Geophysical Laboratory (CIW Team) in August, 2001.

NASA Mission Involvement

This project P.I., David McKay, is proposing a version of our instrument called the Mars Immunoassay Detector (MID) as part of the 2007 Mars Scout mission that will land a scientific probe on Mars.

Future Directions

- I will begin by testing a commercially available antibody and, if possible, begin with one that is potentially useful to the search for life on Mars. Therefore, it is possible that I will test an antibody that recognizes the molecule cyclic adenosine monophosphate (cAMP). My plan is to establish baseline binding curves for an immunoassay for cAMP antigen.

- I also plan to study this antigen in combination with simulated Mars regolith that Carl Allen of JSC has made available to researchers. From this I will determine if it is possible to detect the cAMP in the simulated Martian regolith using the same antibody, or to extract it from the regolith and detect it in the extract and see how the detection from regolith compares to the detection of known quantities of pure antigen.

MILDI Development

Project

Senior Project Investigator(s):
D. McKay

Roadmap Objectives

#1
Sources of Organics on Earth

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#17
Planetary Protection

Accomplishments

MILDI stands for Mars Immunoassay Life Detection Instrument. The purpose of the MILDI project is to develop detection and quantification methods for a large number of organic compounds, many of which may be associated with life. The methods will be built into a small instrument designed for robotic missions to Mars, Titan, and other NASA targets.

The NASA Fundamental Biology Program has supported early development, but the project is transitioning over to an Astrobiology Institute project. Other acronyms used by us for the same general concept include MIDI (Mars Immunoassay Detection Instrument), PASO, Protein Array Sensor for Organics, and one or two others. The general principal is that using antibodies developed to react with either a specific or a class basis can detect organic compounds. The reaction triggers a fluorescence dye, and UV or near UV laser light can be used to trigger the fluorescence, the glow can be detected by a small Charge Coupled Device (CCD) camera, and the brightness can be calibrated to the abundance of the target antigen. An array can be made by borrowing current DNA array technology so that a thousand or more different antibodies can be deposited at indexed locations on a small chip. Up to 10,000 different test sites on a single 2-3cm chip have already been demonstrated for this technology. Soil, rock, or water containing unknown compounds can be extracted and the concentrated extract deposited on the chip. Examples of sought compounds include a large variety of proteins, polysaccharides, amino acids, Polycyclic Aromatic Hydrocarbons (PAHs), and hopnoids (for searching for remains of fossil life). The technique has potential picomole sensitivity for many compounds. Activation of the fluorescence component or dye associated with the antibody is automatic and readout can proceed after an appropriate time interval. This technique has been demonstrated on a number of antibodies and is the basis of clinical detection for many types of tissue, bacteria, viruses, and organic compounds. The antibody-antigen technique is used in the drugstore over-the-counter pregnancy test. It is very reliable and very cheap. A large number of normal fluorescence stains or dyes can also be included in the array in addition to the antibodies.

Progress during this past year includes assembling and coordinating the team, enlisting an experienced project manager, identifying several dozen initial appropriate antibodies, acquiring some of the antibodies, outlining a testing protocol, and engineering the instrument design. The instrument concept has already been included on three of the Mars Scout technology development proposals. While none of the three were awarded development seed money, we anticipate that MILDI will be included on actual Scout proposals when that opportunity is available. We also intend to propose it for the 2007 Mars lander mission.

The basic concept and instrument design is also adaptable to laboratory use for Mars sample return testing for organics and for life. It may also have spinoff use in clinical laboratories, home health care, and biohazard detection.

Highlights

- An instrument no larger than a coffee cup is under development which has the ability to detect up to 10,000 different organic compounds; potentially for use in the detection of life on Mars. The instrument uses a technique, based on antibody reactions, which is the same principle as the home pregnancy test.

- We initiated collaborations with the Centro de Astrobiologĺa (Spain) for the development of this instrument.

Field Trips

When the prototype instrument is built, we plan to take it on field trips to a number of locations to field test it. One of the most severe tests will be on soils in the Atacama Desert. Field tests in Antarctica may also be undertaken.

Cross Team Collaborations

We plan to work closely with the Carnegie Institution of Washington Team to continue development and testing for the MILDI (Mars Immunoassay Life Detection Instrument) project.

In our formal work collaboration arrangement with the Centro de Astrobiologí́a (Madrid, Spain), they are doing design work and array development technology with us for the MILDI.

NASA Mission Involvement

As noted above, this instrument concept has already been included in three different Mars Scout development proposals. We will continue to propose it for actual Mars Scout missions planned to take place in 2007.

In addition, we will propose our MILDI instrument for the Mars 2007 lander and the Mars 2011 lander/sample return mission. We also plan to include it in planning for the construction of a Mars sample return facility on Earth.

Future Directions

- Begin test program using antibodies currently available.
- Acquire or prepare additional antibodies.
- Develop extraction methods suitable for robotic missions.
- Continue engineering design and build prototype.

Mineral Biomarkers

Project

Senior Project Investigator(s):
K. Thomas-Keprta

Roadmap Objectives

#6
Microbial Ecology

#8
Past and Present Life on Mars

#17
Planetary Protection

Accomplishments

This past year we made major progress in characterizing biogenic magnetite made by magnetotactic bacteria MV-1. This magnetite has an unusual non-equilibrium morphology: truncated hexa-octahedral. It also has a very pure composition consisting of only Fe and O at least down to the few hundred ppm level. The size distribution of this magnetite, when corrected for geometry effects of non-perpendicular projection, shows a peculiar truncated non-normal distribution. The sizes are truncated before they exceed the single-domain size field. It then turned out that martian meteorite ALH84001 has essentially identical magnetite comprising a subpopulation that constitutes about 25% of all the magnetites in the carbonate globules.

Extensive literature search revealed no other magnetite, either natural or synthetic, that matched the unusual properties of this magnetite. Experimental work by a team at JSC has produced single domain magnetite by thermal decomposition of siderite, but this magnetite does not match more than one or two of the six characteristic properties of the MV-1 magnetite or the ALH84001 subpopulation magnetite. Experimental work by Chris Romanak and colleagues at the Savannah River Ecology Laboratory shows that both magnetite and siderite can be produced at relatively low temperatures from aqueous solutions under CO_2 partial pressure equal to that on Mars.

Recent work used off-axis electron holography in the transmission electron microscope (TEM) to characterize the magnetic microstructure of magnetotactic bacteria MV-1 and MS-1. One result was the unexpected finding that superparamagnetic magnetite in chains, while too small to retain stable magnetic moments if isolated, are influenced by the single domain magnetite nearby so that they behave like single domain magnetite. This explains how the ends of magnetic chains in magnetotactic bacteria, while often too small to have stable moments, can be influenced by the rest of the chain and can therefore contribute to the overall magnetic moment of the chain. Studies of submicrometer greigite (iron sulfide) show that its crystallization can be biologically induced or biologically mediated, and it is possible to tell the difference from the size distribution. Greigite may provide a suitable biomarker if it can be well characterized.

Studies of Mn precipitates in caves show that Mn minerals are almost certainly formed by the action of Mn-oxidizing microbes. Diagenesis of amorphous and nanocrystalline Mn oxides and hydroxides as well as biofilms form the mineral todorokite. The possibility that Mn minerals may make excellent stable mineral biomarkers can be applied to potential Mars locations and samples.

Highlights

- Tiny magnetite in ALH84001 was confirmed as indistinguishable from magnetite made by bacteria.
- Magnetite in ALH84001 strongly supports early life on Mars hypothesis.
- Manganese minerals formed in caves with the help of bacteria may provide possible biomarkers for use on Mars.

Field Trips

We anticipate no special fieldwork. Members of our team occasionally collect magnetotactic bacteria from various bodies of water throughout the world. Part of our team studying Mn minerals formed in caves may have additional expeditions to the Lechuguilla Cave, NM.

Cross Team Collaborations

We need specialized instruments such as ion probes, which are available at other NAI team institutions such as Carnegie or Arizona State. This may lead to collaborative interactions.

With the ARC Team, we are starting to collaborate by proposing a joint workshop to examine the whole question of magnetotactic bacteria on Mars. Issues regarding magnetotactic bacteria on Mars are: 1) evidence for and against their existence; 2) implications for Mars geology and early life; and 3) possible changes in Mars mission planning.

NASA Mission Involvement

This project is key to understanding possible early life on Mars and therefore influences the design of all Mars robotic missions. These include the 2003 Mars site selection for rovers, the Mars 2007 *in situ* lander mission, and the Mars 2011 sample return mission. In addition, it will influence the 2007 Mars Scout missions. The presence of magnetotactic bacteria on early Mars has great implication for site selection, sample collection strategy, *in situ* analysis instrument selection, and confirming early oceans.

Based on the studies reported here (that some Mn oxides are possible excellent biomarkers), we have new information on what to look for with the various Mars missions. Relevant missions include the 2003 Mars Exploration Rover (MER), the 2007 lander mission, possible 2007 Scout missions, and samples to be selected for return in 2011. If the Mn minerals found on Mars can be characterized well enough, they may tell us that microbes played a part in their formation.

Future Directions

- Acquire additional data on magnetites in ALH84001.

- Synthesize magnetite using procedure of Golden et al. (2001) and compare to ALH84001.

- Analyze matrix of ALH84001 magnetite for other phases.

- Additional analysis of Mn minerals to better determine their value as mineral biomarkers.

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#17
Planetary Protection

Project

Morphologic Biomarkers and Microbial Ecology

Senior Project Investigator(s):
D. McKay

Accomplishments

The purpose of this project is to study a large variety of terrestrial samples to document the micromorphology of microbial biota, both extant life and fossilized life. The goals are to (1) determine, characterize, and document new biomarkers, and (2) relate microbial types, variations, and related features such as biofilms and mineral precipitates to environment, including acidity, water temperature, water chemistry, and changes in these over hours, days, seasons, years, and geologic time. The application is to provide new potential biomarkers for Mars samples and other astromaterials, and to contribute to our understanding of the effects of environmental parameters on microbial life and their resulting biomarkers. This project considers both extant life and fossilized life.

During the past year we have studied fossil microbes in the Gunflint formation, a complex variety of extant microbes and biofilm forming stromatolite structures in Storr's Lake in the Bahamas, a variety of biofilms from several locations, and daily and seasonal variations of microbes in a hot spring in Yellowstone. We have found (along with many others) that the Gunflint formation contains abundant evidence for microbes and we are trying to determine whether the various minerals are bioprecipitated, bioassisted, or coincidentally coexistent with microbes. In addition, we have documented possible fossilized microbial structures in the Mars meteorites Nakhla and Shergotty. Studies are underway to determine whether these structures are associated with organic compounds, have martian deuterium isotopic ratios, and have carbon isotopic ratios characteristic of martian materials. We found that the hot springs studied in Yellowstone show daily variations in hydrogen peroxide content related to UV solar flux and also to breakdown of hydrogen peroxide by microbial activity. We have also shown significant seasonal and longer term variations of sediments in a Yellowstone hot springs which may be related to different sources of water, water chemistry variation, and the resultant change in the mix of bacteria species.

With our Russian colleagues we have documented possibly the best phosphate microfossils ever found from a deposit of Cambrian phosphorites from Mongolia, and have published a photographic atlas of many of them. Phosphate microfossils are bacteria that are mineralized mainly by Ca-phosphate. They provide an important control for comparison to carbonate-replaced microbes, iron oxide-replaced microbes, or silica-replaced microbes.

Highlights

- New morphologies have been were found in Mars meteorites which may be the fossilized remains of microbes; more supporting data are is needed for confirmation.

- Some morphologic biomarkers may be distinctive enough to use without supporting chemistry and mineralogy, but many require multiple types of supporting data to be accepted as true biomarkers.

- 2.1 Ga old iron-rich rocks on Earth have a variety of features showing complex microbial morphologies which may be related to the preserved mineralogy.

- Hydrogen peroxide in Yellowstone hot springs is produced by UV sunlight and removed by microbes. This may have implications for Mars where it has been proposed that UV light produces hydrogen peroxide which has destroyed organic compounds and life near the surface. Perhaps martian microbes also evolved mechanisms to destroy excessive levels of hydrogen peroxide in near-surface moisture.

- Unusual phosphate microfossils were found in Mongolian rocks.

Field Trips

No particular field expeditions are contemplated, but additional key samples from a variety of terrains will be welcome from NAI Members on field trips for other purposes.

Cross Team Collaborations

We are starting a major collaboration in this research area with the Carnegie Institution of Washington.

In this interactive work, we identify possible morphological biomarkers, then we use Carnegie instruments to acquire key chemical and isotopic data. Finally, our teams work together on the ecological interpretations of the biomarker data.

NASA Mission Involvement

The documentation of reliable biomarkers in the form of distinctive morphologies may have extremely important application to Mars missions. Particularly important will be images of rocks and soils returned by the Mars 2003 landers and rovers.

Closeup cameras will be sufficient to see some of the larger microbial features known from Earth such as stromatolites, clusters of microbes, biofilm, filaments, and even larger individual microbes. It is therefore important to document the morphology of these features in typical terrestrial situations to provide a useful database for the missions.

In addition, the 2007 Mars lander mission may have chemical analysis experiments that can provide supporting data. This situation would combine morphology with chemistry, and even mineralogy could be used to search for and possibly find reliable biomarkers.

Finally, when Mars samples are returned by the 2011 Mars mission, the identification of fossilized microbial life will be highly dependent on having reliable morphological clues for microbial features such as individual cells, colonies, filaments, and biofilms. Even if replaced by minerals, some morphological evidence may remain as shown by the phosphorite fossils.

Future Directions

- Analyze biomorph features in Nakhla and ALH84001 for organic compounds and deuterium isotopic composition.

- Determine whether proposed a Mn oxide biomarker found in a New Mexico cave can be applied to other occurrences and whether it is duplicated by inorganic oxidation of Mn.

- Add to the database of morphologic properties of extant bacteria and associated biofilm and mineral precipitates from the Bahaman location, from Dead Sea samples, and from additional Archean samples.

- Begin to establish some universal morphologic features of extant microbes, of fossilized microbes and of their associated products, all of which can serve as morphological biomarkers.

- Begin analysis of a variety of salt and evaporate deposits from the Dead Sea and other locations.

Roadmap Objectives

Project

New Technique Development and Application

Senior Project Investigator(s):
S. Clemett and A. Steele

#1
Sources of Organics on Earth

#6
Microbial Ecology

#8
Past and Present Life on Mars

#17
Planetary Protection

Accomplishments

This project is important because it is pushing the state-of-the-art for analysis and detection of key organic compounds in terrestrial and astromaterials. The project consists of several distinct parts. First is the development and construction of a new and improved double laser system based on the original Stanford design. We have produced a new design which will increase sensitivity and increase spatial resolution from 20-30 micrometer down to <5 micrometer and possibly to 1 micrometer. We have already acquired appropriate lasers. We have submitted a proposal to a NASA announcement for instrument development to acquire funds to build a new double laser probe. While waiting for additional funding, we are reducing and publishing additional data on ALH84001 and Nakhla, two of the Mars meteorites which have been found by other groups to contain indigenous martian organic compounds. Only by probe-based mapping (laser probe, ion probe, etc.) will it be possible to relate specific compounds to the phases which carry them. The double laser mass spectrometer system has the potential to detect organic compounds down in the attomole range. Although it was originally claimed that the polycyclic aromatic hydrocarbons (PAHs) in ALH84001 were Antarctic contamination, we have now demonstrated that ALH84001 contains PAHs in concentrations and locations consistent with indigenous martian organics. The second part of this project is the application of time-of-flight-secondary ion mass spectrometers (TOF-SIMS) to the difficult task of detecting and quantifying very small amounts of organic compounds on the surface of rocks, minerals, and fossils. We are currently using an instrument at Montana State University in collaboration with Ricep Avec, the scientist in charge of the laboratory. The advantage of this technique is that it can have rather high spatial resolution, on the order of 1 micrometer or less. Analysis maps can be made which correlate exactly with SEM maps made in the same instrument so that the exact location of each organic specie can be mapped onto a morphologic or BSE image. Identified compounds include alkenes, alkanes, aromatic and polycyclic aromatic hydrocarbons. Less certain identification includes alkyl pyrroles and pyridyl. In addition to detecting the compounds, we have begun to develop quantitative techniques based on standards. In addition, we have documented the location of organic compounds in the samples and shown their spatial distributions using SEM images as a mapping base. Detection of hopanoids, known to be a very stable family of organic biomarkers, has also been demonstrated by this technique using standards of various concentrations. New results from both of these techniques have been published by our group or are in press.

Another part of this project is the development and testing of new combinations of fluorescent probes as applied to live or nonfossilized cells and biofilms. This part of the project is managed by John Lisle. When properly used in the appropriate combinations, such probes can provide the following information on single cells, cell colonies, and complex biofilms: (1) physiological status, (2) specific metabolic activities, (3) gene expression, and (4) total cell densities. We are setting up a new laboratory at JSC to pursue these techniques.

A new and very sensitive method to detect microbial traces in rocks is being developed by Norman Wainwright. This technique uses Limulus Anti-lipopolysaccharide factor (LALF) and Prophenoloxidase to detect lipopolysaccharide (LPS), glucan, and peptidoglycan down to subpicogram levels. Current efforts involve staining the reacted sample with fixed LALF with an antibody which binds to the reacted LALF and a gold-labeled protein and then detecting the stained material with optical and electron microscopes.

Larry Hersman and his team at Los Alamos are continuing to develop sensitive techniques for detecting microbe fossils. Current results show Atomic Force Microscope images of fossils as well as X-ray Photoelectron Spectroscopy analysis of fossil surfaces.

Highlights

- New analyses of Nakhla Mars meteorite by double laser mass spectrometer show complex organics.

- Detection of hopanes by TOF-SIMS (Time-of-Flight-Secondary Ion Mass Spectrometers) and beginning of standardization

- Identification made of organic compounds in 25 million year old fossilized bacteria—the first successful effort to combine morphologic and chemical biomarkers on fossilized bacteria.

- Use of fluorescent probes to acquire multiple types of information from single microbial cells or small colonies is being developed as an alternate to culturing for use as a life detection technique.

Cross Team Collaborations

Andrew Steele, the PI for the TOF-SIMS part of this project, is moving to the Carnegie Institution of Washington Team in the fall of 2001. We intend to continue working closely with him and with others at Carnegie who might contribute to this project.

The current plan is for Carnegie to acquire a new state-of-the-art TOF-SIMS, which we would then use on this project. The objective would be to develop and refine techniques then apply them to terrestrial rocks, minerals, fossils, as well as to astromaterials. We will continue to work with our Montana State University collaborators. We will build and install a double laser system with the JSC Team. However, we will continue to work with our collaborators at Stanford University.

We welcome other NAI members to participate in both the instrument development and the actual analysis of terrestrial rocks, minerals, biology features, and fossils.

NASA Mission Involvement

This project is directly related to sample return of Mars samples. We are developing the next generation of analytical instruments and techniques that will be critical in analyzing samples returned from the 2011 robotic Mars sample return mission.

Because of the inherently small amount of sample to be returned from Mars, probe techniques will be absolutely required to understand the location, phase carriers, and textural relationships of indigenous organic compounds in the Mars samples. Such techniques require long lead times to develop and test, and that will be done over the next 10 years. The techniques being developed here may also be necessary to help evaluate the returned Mars samples during preliminary examination for any potential hazard to terrestrial life.

Future Directions

- Construct a new double laser mass spectrometer at JSC (dependent on additional funding).

- Acquire and set up a new TOF-SIMS at Carnegie Institution of Washington (Andrew Steele).

- Perform analysis of new terrestrial and astromaterial samples using existing instruments, and new instruments as they come on line.

- Address the question of what organics exist in Mars meteorites, which ones are indigenous, and how did they get there.

- Refine the detection of hopanes and measure the minimum detection limit.

- Analyze possible organic compounds in terrestrial carbonate globules.

- Build a database on compounds, detection limits, and calibrated abundance.

- Set up a new JSC laboratory for bacteria culturing characterization with fluorescent probes and testing on appropriate wild microbes, microbial consortia, and biofilms.

- Continue refining the LALF (Limulus Anti-LPS Factor) detection method for cell components.

- Continue investigating XPS and AFM as tools for detecting microbial fossils.

Roadmap Objectives

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#17
Planetary Protection

Project

Rock Varnish and Microbes

Senior Project Investigator(s):
C. Allen

Accomplishments

It has been proposed that rocks on Mars might be sometimes covered with a coating which may be equivalent of desert varnish found on many rocks on Earth. It has further been proposed that this desert varnish on Mars may have developed with the assistance of microbial activity and may even contain extant life. To prepare for more detailed study of varnish on martian rocks, either by robotic investigation or on returned samples, we have undertaken a study of rock varnish from terrestrial locations. Initial work has been with varnish from southwestern Arizona. Samples are thin sectioned, examined with petrographic microscope, selected areas are microtomed for Transmission Electron Microscope (TEM) imaging and analysis, and samples are analyzed by Scanning Electron Microscope (SEM) and electron microprobe. Results on the Arizona samples show clear evidence of microbial activity. The varnish contains biomorphs, biofilms, and carbon-rich areas. The relationship between microbial activity and the precipitation of the Fe and Mn phases, which comprise the varnish, is being investigated.

Highlights

- Desert varnish on rocks from southwestern Arizona shows evidence for microbial life.

- Some rocks in Viking and Pathfinder imaging appear to have a smooth shiny surface with properties similar to terrestrial rock varnish.

- Varnish on Mars rocks may be a location for either fossil life, extant life, or both and this varnish should be a target for future missions and sample return.

Field Trips

No dedicated fieldwork has occurred as part of this project. However, we solicit documented samples of rock varnish from various terrestrial environments including permafrost areas, Antarctica, dry hot deserts, and other regions. Fossil rock varnish, from old conglomerates for example, is another potential source of useful samples. We may put together a field trip this coming year to investigate the range in populations of rocks with varnish from a given area, and also to investigate their alteration and weathering and the effects of burial.

Cross Team Collaborations

As this is a relatively new project, we have no collaborations in place with other NAI members or teams. However, we would welcome collaboration in the areas of field documentation and sample collection, as well as in analysis techniques.

NASA Mission Involvement

This project is directly focused on the possibility that rock varnish on Mars may be an environment for past life or even extant life. Results of this project will feed directly into operations planning for the Mars Athena mission, which is now the 2003 MER (Mars Exploration Rover) mission. In addition, depending on the results and conclusions of our rock varnish project, the Mars 2007 lander mission should be designed for sampling and analysis of rock varnish, both for organic compounds and for extant life. Tools and procedures must be designed to sample rocks with apparent varnish, and instruments must be included to analyze the varnish for possible biosignatures.

The first robotic Mars sample return mission (possibly 2011) must be designed to sample and return typical rock varnish material from a variety of rock types. Such material may have a high potential for containing biosignatures of fossil or extant life.

Future Directions

In the coming year we will characterize the phases which make up the varnish on the Arizona rocks and compare them to rocks from other localities including desert regions, permafrost regions, and Antarctica.

In each case we will document the extent of microbial activity, if any.

We will also use 16S RNA techniques to attempt to identify the microbial population.

Education & Public Outreach

Distance Learning Events

Johnson Space Center has an advanced lab for distance learning events where scientists share their research and career backgrounds with students and educators in schools that otherwise would not be able to visit our site or attend a presentation. Most presentations are about fifty minutes long and involve slide or PowerPoint presentations and rock samples or other demonstrations.

Educator Workshops

The education team of scientists and educators deliver a variety of hands-on workshops for educators that focus on exploring the solar system. Each of these workshops includes a segment that focuses on searching for life in the solar system. A sampler of workshops are presented so the participants get a large number of activities over a short amount of time. This year 15 workshops were given that reached over 360 teachers.

Informal Education Presentations

Johnson Space Center informal presentations involve hands-on activities for young people that are suitable for a non-classroom setting. Informal settings offer an environment where students may attempt activities that they would not otherwise have time or space to accomplish. The venues vary from after school programs for students to casual weekend family events. This year there were more than 6 informal events ranging from Girl Scouts activities to YMCA programs to latchkey after school programs.

JSC Astrobiology Classroom Activities Development

A team of six educators and all of the Johnson Space Center Astrobiology science team have worked on the development of a series of activities that highlight aspects of research conducted in laboratories. They also have developed and tested several activities that help students to understand some of the difficulties of working with very small samples. Other activities allow students to experiment with yeast's range of tolerance for salt and temperature. These activities will be available on the Johnson Space Center Astrobiology web site and on CD.

JSC Astrobiology Display Events

The JSC Astrobiology team displays and explains a portable exhibit highlighting the research projects of our team at a variety of public events. Highlights from this year include Johnson Space Center Open House and Inspection Day.

JSC Astrobiology Education CD and Website development

A team of educators, scientists, and a graphic artist are working to develop a JSC Astrobiology Education CD containing activities for educators, resources, links, and astrobiology background information. It is designed to be user-friendly and will open and operate like a web site. The same material and format will be used for the education page of the JSC Astrobiology web site.

Microbes! Astrobiology exhibit and education module

Preparation of a 1000 sq ft NASA Astrobiology supplement to the national traveling exhibit "Microbes! Invisible Invaders & Amazing Allies" was done by the astrobiology outreach teams from Johnson Space Center, Marine Biological Laboratory and NASA Astrobiology Institute in collaboration with Space Center Houston. The exhibit was on public display at Space Center Houston from February thru May of 2001. The team also prepared an education module for the exhibit to focus on the basic science and NASA supplement. The Johnson Space Center team presented two teacher workshops at Space Center Houston using the module. Each included hands-on activities and presentations by NASA astrobiologists. Johnson Space Center has continued to follow the Microbes exhibit to other venues and offer the supplement and education module, as well as to work with the Planetary Protection Office to make the supplement a permanent addition to the exhibit.

NAI All Hands Conference Education Posters

The first poster and display materials showcased the astrobiology activities developed by the Johnson Space Center education and science team. The second poster highlighted the Mirobes! exhibit and workshop.

Public Presentations

Johnson Space Center scientists engage the public through more than 17 presentations in a variety of venues around the world. Highlights from this year included the: Planetarium Director's Meeting at JSC, Grant Institute of Geology, University of Edinburgh, Scotland and the Houston Museum of Natural History, Houston, Texas. This talk reached over 450 students, 40 teachers, 50 university facuilty and 400 members of the general public.

ScienceTeam Media Contacts

Events for television, print, and electronic formats.
Media and Other Interviews

Undergraduate Interns

Astrobiology supports a variety of research projects by undergraduate students. Interns conduct research in our laboratory facilities through a mentoring process that involves our entire science team. Students are identified through various programs, matched with one or more scientists who have proposed a research project that can be completed in a ten to twelve week time frame or a project that contributes to on going research. The interns interact with our entire team through meetings where progress on all the JSC team research projects is discussed. The students present the results of their research to the wider community through seminars here at JSC. Many projects are also submitted as abstracts for presentations at professional meetings. We have established a longer research relationship with some of the interns through extended funding and mentoring.

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Back to Table of Contents

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Back to Table of Contents

Marine Biological Laboratory

NASA Astrobiology Institute

Woods Hole, Massachusetts

Principal Investigator

Mitchell Sogin

Marine Biological Laboratory Team Members

Linda A. Amaral Zettler
Marine Biological Laboratory

Ricardo Amils
Centro de Astrobiologia

Mike Atkins
Woods Hole Oceanographic Institute

David J. Beaudoin
Marine Biological Laboratory

Simone Boer
Woods Hole Oceanographic Institute

David A. Caron
University of Southern California

Allen G. Collins
University of California Berkeley

Michael P. Cummings
Marine Biological Laboratory

Alvin de Vera Gomez
Woods Hole Oceanographic Institute

Mark R. Dennett
Woods Hole Oceanographic Institute

Virginia P. Edgcomb
Marine Biological Laboratory

Rebecca J. Gast
Woods Hole Oceanographic Institute

Felipe Gomez
Centro de Astrobiologia

Kai-Uwe Hinrichs
Woods Hole Oceanographic Institute

Barbara Inzina
Marine Biological Laboratory

Holger W. Jannasch
Woods Hole Oceanographic Institute

Brendan G. Keenan
Marine Biological Laboratory

David T. Kysela
Marine Biological Laboratory

Ping Liang
Marine Biological Laboratory

Pauline B. Lim
Marine Biological Laboratory

Andrew McArthur
Marine Biological Laboratory

Tom McCormack
Marine Biological Laboratory

Laura A. McInerney
Marine Biological Laboratory

Monica Medina
Marine Biological Laboratory

Mark Messerli
Marine Biological Laboratory

Stephen Molyneaux
Woods Hole Oceanographic Institute

Dawn M. Moran
Woods Hole Oceanographic Institute

Laila A. Nahum
Marine Biological Laboratory

Thomas A. Nerad
American Type Culture Collection

Tara Nihill
Marine Biological Laboratory

Lorraine C. Olendzenski
Marine Biological Laboratory

David J. Patterson
Marine Biological Laboratory

David Remsen
Marine Biological Laboratory

Monica Riley
Marine Biological Laboratory

Andrew Roger
Dalhousie University

Mak Saito
Princeton University

Margrethe H. Serres
Marine Biological Laboratory

Jeffrey D. Silberman
University of California Los Angeles

Alastair G.B. Simpson
Marine Biological Laboratory

Peter J. Smith
Marine Biological Laboratory

Mitchell L. Sogin
Marine Biological Laboratory

Andreas P. Teske
Woods Hole Oceanographic Institute

Carl Wirsen
Woods Hole Oceanographic Institute

Executive Summary

Accomplishments

The evolution and survival of Earth's biota would not be possible without single-cell organisms. For nearly 90% of this planet's history, microbes were the only kind of life. These organisms dominate every corner of our biosphere, and they are likely to be the only life-forms that might be encountered in other parts of our solar system if not the entire cosmos. There is only sparse information about the true diversity of microorganisms and how they orchestrate and drive key biogeochemical cycles that shape our ever-changing planet. We are only now beginning to appreciate the tremendous diversity of microorganisms, which dwarfs animal/plant conventions. A more complete understanding of microbial diversity and evolution, descriptions of ecosystem-wide patterns of gene expression, and detailed analyses of biogeochemistry would provide a new foundation for interpreting paleontological and geological studies that describe Earth's early history. In addition to contributing towards the design of life-detection experiments, information about microbial evolution and their limits of habitability provides guidance for seeking extraterrestrial life.

The MBL Astrobiology team employs molecular techniques to explore how relatively simple organisms and their genomes (compared to those of metazoans) evolved into more complex forms. We are united by questions that parallel the evolutionary biology goals in the Astrobiology Roadmap: Where did we come from? How did we get here? and Where are we going? We seek to understand how multi-member gene families and entire genomes have evolved, how evolution of genotype is related to changes in phenotype, how processes other than simple mutation influence evolution of life on earth e.g. endosymbiosis, how microbial lineages have adapted to extreme environments and how eukaryotes originated and evolved in to complex, multicellular forms. Our investigations span the levels of individual genes, genomes, cells, populations, communities and entire ecosystems.

An important goal for projects that investigate the evolution of biological complexity is to develop an understanding of protist diversity and a robust phylogeny for all eukaryotes. To address these questions we have taken advantage of the remarkable conservation of both small subunit (SSU) and large subunit (LSU) ribosomal RNA sequences to infer eukaryotic phylogenies. This work addresses two important questions. 1) What is the identity of early diverging eukaryotic lineages? and 2) What is the identity of protist groups that were ancestral to the metazoans? The rRNA analyses are complemented by studies of other genes including alpha-tubulins, beta-tubulins and fumerases. One of our guiding hypotheses is based upon SSU rRNA trees, which show the earliest diverging eukaryotes lack mitochondria and peroxisomes. In nearly all examples, early diverging eukaryotic lineages in rRNA trees are represented by organisms of medical relevance.

Since it is unlikely that clinicians would have discovered the origins of eukaryotes, an important goal of the MBL astrobiology program is to search for novel eukaryote diversity in rarely studied environments, some of which may resemble conditions that could have existed millions/billions of years ago on other solar system bodies. We use molecular techniques to explore protist diversity in extreme environments including sedimentary cores from the Guaymas hydrothermal vent environment, the iron-rich, acidic environment of the Rio Tinto in southwestern Spain, and in marine and freshwater anoxic sediments. Rio Tinto is of particular interest because in many ways it may represent an analogous environment to early martian conditions when liquid water might have been present. In support of these investigations and as part of a very significant expansion of our outreach program, we have also developed a new website titled **micro*scope** (http://www.mbl.edu/microscope). The site provides an image-rich means to access and identify microbial diversity. The following are highlights from these investigations.

- Analysis of rRNAs show that the absence of organelles occurs in both deep branching and more recently diverged eukaryotic lineages. Such losses have occurred independently several times.

- Based upon comparisons of both rRNAs and tubulins, the excavate taxa including diplomonads and other species that possess similar ultrastructure features are not a monophyletic assemblage.

- Phylogenies based on alpha-, beta-, and combined alpha- plus beta-tubulin genes place the excavate taxon Carpediemonas as a close relative of diplomonads (and with SSUrRNA, retortamonads), to the exclusion of all other eukaryotes. This novel relationship is recovered across phylogenetic methods and various taxon-deletion experiments and under different SSU rRNA alignments.

- Phylogenetic analysis of combined data sets from both small subunit and large subunit ribosomal RNA sequences show that Bilateria are not more closely related to the Ctenophora than to the Cnidaria. The Bilateria most likely are the sister group to the Silicea.

- The molecular survey of eukaryotic microbial diversity in a hydrothermal vent environment shows that these anoxic sediments and the overlying seawater harbor a mixture of genetically diverse protists. Many sequence isolates represent novel protists, including early branching eukaryotic lineages or extended diversity within described taxa.

- At least two mechanisms, with overlapping consequences, account for the eukaryotic community structure of this environment; adaptation to warm anoxic environments, and wide distribution of aerophilic protists in marine environments, some of which may migrate into and survive in the sediment, while others e.g. phototrophs, are simply deposited by sedimentary processes.

• In contrast to most environments, eukaryotic microorganisms rather than prokaryotes dominate parts of Rio Tinto.

• **Micro*scope**, a comprehensive website for microbial diversity, was released for public use in May, 2001.

• Our investigations of eukaryotic microbial diversity in the hydrothermal sediments of the Guaymas Basin (Gulf of California, Mexico) are complemented by ^{13}C isotopic analysis of archaeal and bacterial lipids and molecular diversity surveys based upon comparisons of 16S rRNAs. This allows us to correlate community composition with modes of nutrition. A major lineage of uncultured euryarchaeota (ANME-1 including ANME-1b), and uncultured members (ANME-2) of the Methanosarcinales, the acetoclastic and methyl-disproportionating methanogens dominate this ecosystem. There are also associated bacterial populations that are predominantly made up of gamma, delta and epsilon-Proteobacteria, green non-sulfur bacteria, and the uncultured candidate subdivision OP11.

• We have identified a methanotrophic microbial consortium at a hydrothermal vent site that utilizes geothermal and biogenic methane.

• In contrast to oxygen-respiring, sulfide-oxidizing hydrothermal vent microbes that are commonly the basis of the hydrothermal vent food chain, the archaeal/bacterial consortium in Guaymas thrives without oxygen and requires only sulfate as oxidant.

The projects on photosymbioses and studies of genetic changes to phenotypic are progressing. Both of these efforts required field trips to obtain samples for analysis. Gast has isolated several cDNA clones that may be important in formation of photosymbiotic algal relationships but more experiments are required before reporting definitive results. Cummings' study of opsin evolution has made substantial progress in both the analytical areas and data acquisition. Sequences are in progress or have been completed for 35 opsin cDNA clones and RNA samples have been prepared for more than 60 species. In anticipation of a large data base of opsin clones, Cummings has made progress in the implementation of computationally demanding methods of sequence analysis for linking phenotypic change with genotypic evolution. He has completed the largest and most comprehensive study of spectral tuning in vertebrate color vision. Furthermore this work has demonstrated the successful application of novel analytical techniques related to those that have been applied to space shuttle landing to genotype-phenotype relationships. Highlights include:

• Few of the hundreds of variable amino acid positions in vertebrate opsin have an influence on wavelength of light absorbed.

• Several positions appear to be equivalent, or indistinguishable, because they co-vary, with regard to their influence on spectral tuning.

• Most of the variance in spectral tuning can be explained by two or three amino acid changes.

Riley's "Evolution of Protein" group has been very active over the course of the past year with the publication of six manuscripts. They have been exploring the link between genotype and phenotype evolution through studies of related gene families in *E. coli*. Through these analyses the Riley group has identified a potential role of gene fusion in evolution which may be extremely important in the evolution of genomic complexity. Gene fusions may be responsible for achieving efficiency in localizing related reactions in physical proximity, not in equilibrium with the aqueous phase of the cell. Highlights include:

• The function of 81% of the genes in *E. coli* have been identified (one year ago the functions of 1/3 of the *E. coli* coding regions were unknown)

• Multimodular genes are multifunctional and define physiologically related activities.

Finally, we have provided a report on progress of the Ecogenomics Focus Group (not just the MBL-related activities). Beyond our contributions to the EcoGenomics Focus Group organization activities and meetings, the MBL group has restricted its efforts to constructing libraries for proxy cyanobacterial cultures and making strategic decisions about equipment and methodology that will be employed for analyzing gene expression patterns throughout the hypersaline mat cores. In the very near future, samples returned from the field site will provide a second proxy culture for a sulfate reducing species. Because of the delay in reaching the field site, this project is just getting underway. Highlights for the EcoGenomics Focus Group include:

• The NAI EcoGenomics Focus Group has initiated a comprehensive sampling of biogeochemical processes and microbial diversity in the hypersaline mats of the Guerrero Negro.

• The Keck Foundation awarded funds to the MBL for the construction of an advanced laboratory of ecological and evolutionary genomics.

Roadmap Objectives

#4
Genomic Clues to Evolution

#6
Microbial Ecology

#7
Extremes of Life

Project

Diversity and Physiology of Prokaryotes in Selected Thermophilic and Mesophilic Environments That Might Resemble Early Earth's Biosphere

Senior Project Investigator(s):
A. Teske, V. Edgcomb, H. Jannasch

Accomplishments

Microbial communities in hydrothermal sediments of the Guaymas Basin (Gulf of California, Mexico) were analyzed by 16S rRNA sequencing and by ^{13}C isotopic analysis of archaeal and bacterial lipids. In this way, microbial community composition can be correlated to carbon assimilation pathways that are reflected in the isotopic composition of diagnostic lipids. The Guaymas sediments contained a major lineage of uncultured euryarchaeota (ANME-1 including ANME-1b), and uncultured members (ANME-2) of the Methanosarcinales, the acetoclastic and methyl-disproportionating methanogens. The lipid profiles were congruent with the sequence profiles. Predominant archaeal lipids in the Guaymas Basin sediments included archaeol, diagnostic for non-thermophilic euryarchaota, and sn-2-hydroxyarchaeol, diagnostic for members of the Methanosarcinales. These lipids were extremely ^{13}C depleted ($\partial^{13}C$ = - 80 to - 60 ‰), indicating that they originated from anaerobic methanotrophic archaea that oxidized and assimilated ^{13}C depleted methane that is abundant in Guaymas Basin vent fluids. The archaea occur associated with diverse bacterial populations, predominantly gamma, delta and epsilon-Proteobacteria, green non-sulfur bacteria, and the uncultured candidate subdivision OP11. Bacteria that consume hydrogen and acetate, the reaction products of archaeal methane oxidation, make this process thermodynamically feasible; delta-Proteobacterial sulfate reducers are good candidates.

The discovery of a highly structured, anaerobic methane-oxidizing archaeal ecosystem in the Guaymas sediments was a surprise. Originally, we were searching for novel lineages of deeply branching archaea and bacteria, without expecting detailed results on carbon cycling. We complemented the highly suggestive sequence data with ^{13}C-isotopic signatures from specific archaeal lipids that proved the case for anaerobic methane oxidation. Archaeal and bacterial population structure of hydrothermal sediments at the Guaymas Basin vent sites derived from 16S rRNA sequence and ^{13}C analysis of archaeal and bacterial lipids: indications for anaerobic methanotrophy.

Highlights

We have identified a methanotrophic microbial consortium at a hydrothermal vent site that utilizes geothermal and biogenic methane. In contract to oxygen-respiring, sulfide-oxidizing hydrothermal vent microbes that are commonly the basis of the hydrothermal vent food chain, this archaeal/bacterial consortium thrives without oxygen and requires only sulfate as oxidant.

Field Trips

The samples for this project were collected during a diving expedition to the Guaymas vents (Gulf of California, Mexico) in April and May 1998, with RV ATLANTIS and Research Submersible ALVIN of the Woods Hole Oceanographic Institution.

Cross Team Collaborations

This project has a direct impact on the research strategies of the new NAI Team project "Subsurface Biospheres" at the University of Rhode Island and at the Woods Hole Oceanographic Institute (PI, Steve D'Hondt with A. Teske and K. Hinrichs, Co-PIs).

This microbiology and geochemistry-oriented research team focuses on the composition, environmental constraints, and *in situ* activities of deeply buried microbial communities in the marine subsurface. These are models for extraterrestrial subsurface biospheres on planets with inhospitable surface environments.

The Guaymas studies (particularly, the successful integration of rRNA community surveys and compound-specific ^{13}C isotopic analysis) represent a paradigm for how to approach this deep subsurface project. We expect synergetic effects from comparisons of vent and deep-subsurface datasets and methods.

NASA Mission Involvement

This project informs NASA missions on life forms that are possible on planets with anaerobic subsurface or hydrothermal habitats (e.g., Europa). An anaerobic C1 carbon cycle can be sustained, as long as sulfate is available as oxidant. Such a perspective about the existence and survival of life forms is critical background in considering and planning missions to search for life beyond the Earth.

Future Directions

- Methane oxidation is most likely carried out by a syntrophic consortium of archaea and sulfate-reducing bacteria; we found these consortia in the Guaymas sediments by fluorescence in situ hydridization (FISH). Amplification and sequencing of the phylogenetically informative key genes of sulfate reduction, dissimilatory sulfite reductase, will be used to identify the sulfate-reducing syntrophs in the Guaymas Basin sediments.

- We have large sequence data sets from deeper sediment layers that have not been analyzed so far. We will build a detailed phylogenetic depth profile of bacterial and archaeal populations in the Guaymas sediments, and correlate changing community composition with temperature and chemical gradients.

Roadmap Objectives

#4
Genomic Clues to Evolution

#6
Microbial Ecology

#7
Extremes of Life

Project

Diversity of Eukaryotes in Thermophilic and Mesophilic Environments That Might Resemble Early Earth's Biosphere

Senior Project Investigator(s):
V. Edgcomb, M. Sogin, A. Teske

Accomplishments

Molecular microbial ecology studies have revealed remarkable prokaryotic diversity in extreme hydrothermal marine environments. There are no comparable reports of culture-independent surveys of eukaryotic life in warm, anoxic marine sediments. Using sequence comparisons of polymerase chain reaction (PCR) amplified small subunit ribosomal RNAs, we characterized eukaryotic diversity in hydrothermal vent environments of Guaymas Basin in the Gulf of California. These anoxic sediments and the overlying seawater harbor a mixture of genetically diverse protists. Many sequence isolates represent novel protists, including early branching eukaryotic lineages or extended diversity within described taxa. At least two mechanisms, with overlapping consequences, account for the eukaryotic community structure of this environment. The adaptation to warm anoxic environments is evidenced by specific affinity of environmental sequences to microaerophilic species in molecular trees. This is superimposed on a background of widely distributed aerophilic protists, some of which may migrate into and survive in the sediment while others, e.g. phototrophs, are simply deposited by sedimentary processes. This year we completed the phylogenetic analysis of eukaryotic 18S rRNA sequences from the upper 3 cm layers of two Guaymas Basin cores.

Highlights

- This first molecular survey of eukaryotic microbial diversity in a hydrothermal vent environment shows that these anoxic sediments and the overlying seawater harbor a mixture of genetically diverse protists. Many sequence isolates represent novel protists, including early branching eukaryotic lineages or extended diversity within described taxa.

- At least two mechanisms, with overlapping consequences, account for the eukaryotic community structure of this environment; 1) adaptation to warm anoxic environments, and 2) wide distribution of aerophilic protists in marine environments, some of which may migrate into and survive in the sediment, while others e.g. phototrophs, are simply deposited by sedimentary processes.

Field Trips

The samples for this project were collected during a diving expedition to the Guaymas vents (Gulf of California, Mexico) in April and May 1998, with RV ATLANTIS and Research Submersible ALVIN of the Woods Hole Oceanographic Institution.

NASA Mission Involvement

This project concentrates on surveying hydrothermal vents as part of a comprehensive biodiversity survey of extreme habitats on Earth. Knowing the full range of biodiversity in extreme habitats on Earth allows us to formulate hypotheses on “what to expect” in extraterrestrial biospheres.

Future Directions

• The 18S rRNA survey of the top 3 cm of the Guaymas hydrothermal vent sediments is completed. We will try to amplify 18S rRNA genes from deeper, hotter sediment layers, which could harbor different, more extremophilic or ancestral eukaryotic populations.

• New sequence information can be used to design phylum-specific PCR probes to determine the global distribution of these novel eukaryotic lineages.

Eukaryotic Diversity in the Rio Tinto: Spain’s Acidic/High Metal Extreme Environment

Project

Senior Project Investigator(s):
M. Sogin, A. Zettler, R. Amils

Roadmap Objectives

#3
Models for Life

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

Accomplishments

We have completed a molecular characterization analysis of full-length, eukaryotic, small-subunit ribosomal RNA of isolates obtained from several sampling stations along the Tinto River, including samples taken at the source of the river. Some of our clones showed high similarity to taxa reported in previous studies employing light microscopical techniques. These included the highly conspicuous members of the Euglenozoa (euglenids), Chlorophyta (chlamydomonad and chlorella-like relatives), as well as the Bacillariophyta (diatoms). Members of the Viridiplantae were represented by streptophyte and charophyte relatives.

However, our results also revealed a diversity of other protists not previously identified with conventional techniques. These included members of the stramenopiles (e.g. chrysophytes) and several cercomonad relatives. Among the most interesting clone sequences in the river were those from relatives of amoeboid taxa. In our molecular survey, we found one clone that branched with the Vahlkampfia. Occurrence of vahlkampfids has been noted in other acidic environments. More interestingly, a few of our clones branched with other recently sequenced amoeboid taxa. In initial analyses, these clones branched near the animal/fungal divergence. To obtain a more fully resolved phylogenetic placement for these clones, we added them to a data set with fewer taxa (19) wherein additional sites could be used in the analysis (1550). Our more refined analyses showed these clones branching among the nucleariid amoebae – a group of filose amoebae branching near the animal-fungal divergence and of questionable monophyly. The second group of clones branched with *Filamoeba nolandi*. The current taxonomic placement of *Filamoeba nolandi* is equivocal and reflects much of the state of amoebae systematics.

Our study also revealed a diversity of fungi somewhat different than those previously identified in the river using traditional methods. The majority of our clones belonged to the Ascomycota, but there were a few representatives from the Zygomycota. We have summarized our results in a phylogenetic analysis.

Highlights

In contrast to most extreme environments, eukaryotes rather than prokaryotes dominate parts of the Rio Tinto.

Field Trips

The data summarized from this project were the result of fieldwork carried out in April of 1999 in the Tinto River located in the Huelva district of Southwestern Spain. Linda Amaral Zettler (Marine Biological Laboratory) collected samples with the assistance and guidance of Ricardo Amils (Centro de Astrobiología/ Universidad Autónoma de Madrid), whose lab has been studying the river for 10 years. Although we obtained both sediment and biofilm samples for DNA extraction, we were only successful in amplifying DNA from biofilm samples. We were also successful in establishing cultures of a representative chlorella, chlamydomonad and euglenid species from water samples obtained from this field trip. It is our goal for an upcoming field trip (slated for September, 2001) to isolate cultures of representative phylogenetically diverse protists that we identified from our diversity studies. These cultures will be used in further physiological studies and for studying hydrogen ATPases that may be involved in pH maintenance.

Cross Team Collaborations

We are currently engaged in ongoing collaboration with our colleagues at the CAB (Centro de Astrobiología, Madrid, Spain). As mentioned above, we have already carried out a collection expedition to the Tinto River. Our collaboration represents the first international cooperation at the NAI and serves to further international and interdisciplinary research in extreme environments.

Also, Amils and Gomez from the CAB visited the Marine Biological Laboratory in the fall of 1999 and again in the summer of 2000. Amils gave presentations on the Tinto River (Rio Tinto) to the Woods Hole scientific community on both visits.

Gomez, an astrobiology postdoctoral fellow from Spain, spent a month and a half at the Marine Biological Lab in the fall of 1999. There, he worked on bacterial samples from the Tinto River and learned molecular techniques and methods in phylogenetics. In the summer of 2000, he returned to the MBL to help analyze the eukaryotic sequences obtained from the Rio Tinto and to participate in the three-week intensive Molecular Evolution Workshop held at the MBL in August. Our manuscript summarizing eukaryotic diversity in the RioTinto is in preparation.

NASA Mission Involvement

One of astrobiology's aims is to understand how microbial communities thrive under extreme conditions. Our study site, the Tinto River in Spain, is considered a good model for study about life on Mars because its high iron concentrations are thought to resemble conditions found on Mars.

Our diversity study provides important information about what kinds of phylogenetically diverse microbes can live under acid and high metal extremes. The physiological studies being done as a sister project to our microbial diversity study may provide insights into how organisms are able to adapt to the harsh environments most likely present during the first appearance of life on Earth. This work relates directly to NASA's goal to establish limits for life in environments that provide analogues for conditions on other worlds.

Future Directions

- Publish previous results from our molecular analyses.

- Bring phylogenetically diverse protists into culture to continue physiological studies.

Eukaryotic Origins and the Evolution of Cellular Complexity–Eukaryotic rRNA Evolution: Early Diverging Eukaryotes

Project

Senior Project Investigator(s):
V. Edgcomb and M. Sogin

Roadmap Objectives

#2
Origin of Life's Cellular Components

#4
Genomic Clues to Evolution

#7
Extremes of Life

Accomplishments

Pelobionts are flagellated protists that inhabit micro-oxic and anoxic environments. They represent one group of eukaryotes that has been identified as a potentially deep-branching eukaryotic lineage on the basis of ultrastructure and certain molecular phylogenies. Because they lack stacked dictyosomes, mitochondria, and outer dynein arms in their flagellar apparatus, pelobionts might represent early diverging eukaryotic lineages.

Molecular analyses of DNA-dependent RNA polymerase II suggest the pelobiont *Mastigamoeba invertens* diverged early. Analyses of SSU (small subunit) ribosomal RNAs, in contrast, position pelobionts near the eukaryotic “crown groups”, but not as a monophyletic group. We sequenced SSU rRNA genes from four additional pelobiont taxa (including two new isolates). This work confirmed the sequence for *M. invertens* by *in situ* hybridization, compared the ultrastructural features of these taxa, and subjected these new pelobiont sequences to a rigorous phylogenetic analysis in the context of a larger alignment of eukaryotes, including known sequences from other pelobionts.

Results show that despite the absence of typical eukaryotic organelles, e.g., mitochondria, dictyosomes, chloroplasts, coated vesicles, and peroxisomes, pelobionts are not basal to other eukaryotes in our molecular analyses nor are they specifically related to a previously described pelobiont-like organism, *Mastigamoeba invertens*. This result calls into question the taxonomic assignment of this organism and the phylogenetic methodologies that placed it as a deep-diverging eukaryotic lineage. Independent of the evolutionary models tested, sequence alignments employed, and taxon sampling, the pelobionts (exclusive of *M. invertens*) form a monophyletic group together with *Endolimax nana* in maximum likelihood, distance and parsimony inferences. Many analyses suggest the pelobionts share common ancestry with the Entamoebidae, with *E. nana* sharing an unique common evolutionary history with *Mastigamoeba simplex*. Pelobionts do not represent one of the earliest branching lineages as suggested by RNA polymerase II and ultrastructure.

Highlights

Molecular analyses suggest that the earliest diverging eukaryotes lacked mitochondria and peroxisomes. Analysis of rRNAs show that these characteristics can also be absent in eukaryotic lineages that diverged relatively late. The loss of these organelles has occurred independently several times in the history of eukaryotes.

Field Trips

Field work for this project occurred prior to the Year 3 reporting period. Collection of new pelobiont taxa was performed by Alastair G. B. Simpson, Virginia P. Edgcomb, and David J. Patterson (MBL/University of Sydney) at various locations in Australia between 1996 and 1998.

Cross Team Collaborations

Our project this year involved collaboration with David J. Patterson and Alastair G.B. Simpson of University of Sydney for purposes of sample collection and isolation of organisms.

V. Edgcomb was in Sydney, Australia between June and August in 1998 to collect samples, perform ultrastructural studies, and receive taxonomic training. This has contributed to our current research project work.

NASA Mission Involvement

Any search for life on other planets or small solar system bodies must consider the possible occurrence of both prokaryotic and eukaryotic microbes. This life search definitely relates to our research project.

Through our research project investigations, we have shown that a wide diversity of eukaryotes are just as capable of adapting to anoxic conditions as are prokaryotes. Therefore, extraterrestrial life detection must consider the molecular biology and biochemistry that is characteristic of eukaryotes.

Future Directions

We will complete a detailed analysis of rRNA genes from Carpediomonas and other members of the excavate taxa. These have been proposed to be representative of the earliest diverging lineages. Our initial phylogenetic inferences do not support monophyly for this group although some appear to be very deep branching taxa.

Eukaryotic Origins and the Evolution of Cellular Complexity –Evolution of Tubulins

Project

Senior Project Investigator(s):
V. Edgcomb, M. Sogin, A. Roger

Roadmap Objectives

#2
Origin of Life's Cellular Components

#4
Genomic Clues to Evolution

Accomplishments

Jakobids are free-living, heterotrophic flagellates that might represent early-diverging mitochondriate protists. They share ultrastructural similarities with other eukaryotes that occupy basal positions in molecular phylogenies. Their mitochondria genome architecture is also eubacteria-like, suggesting a close affinity with the ancestral alpha proteobacterial symbiont that gave rise to mitochondria and hydrogenosomes. Tubulin gene phylogenies are in general agreement with mitochondrial gene phylogenies and ultrastructural data, indicating that the "jakobids" may be polyphyletic. Relationships to the putatively deep-branching amitochondriate diplomonads remain uncertain.

To elucidate relationships among jakobids and other early-diverging eukaryotic lineages, we have characterized alpha- and beta-tubulin genes from four jakobids: *Jakoba libera, Jakoba incarcerata, Reclinomonas americana* (the "core jakobids"), and *Malawimonas jakobiformis*. These are the first reports of nuclear genes from these organisms. Phylogenies based on alpha- and beta- and combined alpha- plus beta-tubulin protein data sets do not support the monophyly of the jakobids.

Beta-tubulin and combined alpa- plus beta-tubulin phylogenies show a sister group relationship between *J. libera* and *R. americana*. However, the two other jakobids, *M. jakobiformis* and *J. incarcerata* have unclear affinities. In all three analyses, *J. libera, R. americana* and *M. jakobiformis* emerge from within a well-supported large "plant-protist" clade that includes plants, green algae, cryptophytes, stramenopiles, alveolates, Euglenozoa, Heterolobosea, and several other protist groups. However, animals, fungi, microsporidia, parabasalids, and diplomonads and not included in this affinity relationship. Tubulin gene phylogenies are in general agreement with mitochondrial gene phylogenies and ultrastructural data, indicating that the "jakobids" may be polyphyletic. Relationships to the putatively deep-branching amitochondriate diplomonads remain uncertain. During the Year 3 reporting period we completed phylogenetic analyses of all three data sets and published the results of the work described above.

Since the 1980s, diplomonads and their relatives retortamonads have been proposed as primitive eukaryotes that diverged before the acquisition of mitochondria. On this basis they became key organisms in attempts to understand the evolution of eukaryotic cells. Recent findings of mitochondrial-like genes in *Giardia* suggest that diplomonads may have lost mitochondria. However, the mitochondrial origins of some of these genes are highly questionable, and premitochondrial diplomonad scenarios are still advanced. In this phylogenetic examination we focus on a series of mostly poorly known eukaryotes argued to be related to diplomonads on morphological grounds; the excavate taxa. Phylogenies of SSU rRNA genes, alpha tubulin, beta tubulin and concatenated alpha + beta tubulin all scatter the various excavate taxa across the diversity of eukaryotes. However all phylogenies place the excavate taxon *Carpediemonas* as a close relative of diplomonads (and with SSU rRNA, retortamonads), to the exclusion of all other eukaryotes. This novel relationship is recovered across phylogenetic methods and various taxon-deletion experiments and under different SSU rRNA alignments. Statistical support is strongest under maximum likelihood (when among-site rate variation is modelled) and when the most divergent diplomonad sequences are omitted, suggesting a true relationship rather than a long-branch attraction artifact. As *Carpediemonas* contains double membrane-bound organelles that are reminiscent of mitochondria, we speculate on the possible occurrence of mitochondria in the ancestors of diplomonads. The phylogenetic position of *Carpediemonas* suggests that it will be valuable in interpreting the evolutionary significance of many molecular and cellular biological peculiarities of diplomonads.

Highlights

- Work here convinces us that continued studies of tubulin genes will not be of major importance in identifying early branching patterns in the eukaryotic line of descent.

- These results provide evidence that the excavate hypothesis for the origins of eukaryotes is not likely to be correct.

- Phylogenies based on alpha-, beta-, and combined alpha- plus beta-tubulin genes place the excavate taxon *Carpediemonas* as a close relative of the diplomonads (and with SSU rRNA, retortamonads), to the exclusion of all other eukaryotes. This novel relationship is recovered across phylogenetic methods and various taxon deletion experiments and under different SSU rRNA alignments.

- The phylogenetic position of *Carpediemonas* suggests that it will be valuable in interpreting the evolutionary significance of many molecular and cellular biological peculiarities of diplomonads.

Field Trips

Field work for this project occurred prior to the Year 3 reporting period. Collection of excavate taxa was performed by Alastair G. B. Simpson and David J. Patterson (MBL/University of Sydney) at various locations in Australia between 1996 and 1998.

Cross Team Collaborations

This project involved collaboration with David J. Patterson and Alastair G.B. Simpson of University of Sydney for purposes of sample collection and isolation of organisms.

Alastair Simpson came to MBL during 1997/1998 for training in phylogenetics. During this time, he and Virginia Edgcomb isolated and sequenced the SSU rRNAs for many of the organisms now being used in our project study. In addition, they provided taxonomic training for Virginia P. Edgcomb between June and August in 1998 at various locations in Australia.

NASA Mission Involvement

Any search for life on other planets or small solar system bodies must consider the possible occurrence of both prokaryotic and eukaryotic microbes. This life search definitely relates to our research project.

Through our research project investigations, we have shown that a wide diversity of eukaryotes are just as capable of adapting to anoxic conditions as are prokaryotes. Therefore, extraterrestrial life detection must consider the molecular biology and biochemistry that is characteristic of eukaryotes.

Future Directions

This project has been completed.

Roadmap Objectives

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

Project

Eukaryotic rRNA Evolution: Origins of "Crown Group Taxa"

Senior Project Investigator(s):
M. Sogin

Accomplishments

Previous analyses of small subunit ribosomal RNAs describe the massive evolutionary radiation of eukaryotes that occurred approximately one billion years ago. This event gave rise to plants, animals, fungi, and many other protist groups including stramenopiles, alveolates, and a large number of independent protist lineages. We seek a better understanding of detailed branching pattern for the "crown group" taxa by obtaining additional molecular sequence data and hence greater resolution in our phylogenetic inferences. We are analyzing full length, large subunit rRNA sequences, which have twice as many positions as SSU rRNAs. Our first objective is to study relationships among basal metazoan lineages. After identifying competing hypotheses, we performed maximum likelihood searches for trees that conform to each hypothesis. We employed Kishino-Hasegawa tests to determine if the data (LSU, SSU, and combined) reject any of the competing hypotheses. We also conducted unconstrained tree searches, calculated bootstrap indices, and compared the resulting topologies. We applied Shimodaira-Hasegawa tests to determine if the data reject any of the topologies resulting from the constrained and unconstrained tree searches. LSU, SSU and the combined data strongly contradict two assertions pertaining to sponge phylogeny. Hexactinellid sponges are not likely to be the basal lineage of a monophyletic Porifera or the sister group to all other animals. Instead, Hexactinellida and Demospongia form a well-supported clade of siliceous sponges, Silicea. It remains unclear, based on these data alone, whether the calcarean sponges are more closely related to Silicea or to non-sponge animals. The SSU and combined data reject the hypothesis that Bilateria is more closely related to Ctenophora than it is to Cnidaria. The LSU data alone do not refute either hypothesis, but instead raise the possibility that Bilateria may be sister to Silicea. LSU and SSU data agree in supporting the monophyly of Bilateria, Cnidaria, Ctenophora, and Metazoa. LSU sequence data reveal phylogenetic structure in a data set with limited taxon sampling.

In a related project, we have used similar molecular techniques to explore the phylogeny of the ophistokonths. We have identified the heterotrophic flagellate *Ancyromonas sigmoides* as a potential common ancestor of fungi, choanoflagellates, and metazoan animals. To re-examine and to substantiate these results, we are re-analyzing the original 18S rRNA dataset and have compiled a comprehensive 28S rRNA dataset, including many new sequences that encompass all eukaryotic crown group lineages that are critical for this analysis. We take multicellularity for granted, but almost the entire tree of life, based on rRNA sequences, is microbial and unicellular; multicellular organisms occupy a few recent branch tips. An over-riding question is: What triggered this evolutionary anomaly, multicellularity?

Highlights

- These data show that Bilateria are not more closely related to the Ctenophora than to the Cnidaria. The Bilateria most likely are the sister group to the Silicea.

- The flagellate *Ancyromonas*, isolated from the 9N East Pacific Rise hydrothermal vents, could shed light on the early evolution of eukaryotic multicellularity.

Field Trips

Sponges and ctenophores used in this study were collected locally in Woods Hole. *Ancyromonas sigmoides* was collected in December 1995 from the 9ºNorth East Pacific Rise hydrothermal vents, with RV ATLANTIS II and Research Submersible ALVIN of the Woods Hole Oceanographic Institution. New Sampling trips to Australia are planned for winter 2001/2002, with David Patterson.

Cross Team Collaborations

The metazoan work was done at the MBL when Medina was a post doctoral student in Sogin's laboratory. During that time Medina was supported through joint funding from NAI and the NIH. She has since moved to the San Francisco area and holds a joint appointment at the California Academy of Sciences and the DOE Joint Genome Institute.

The analyses for our project were completed in collaboration with Allen G. Collins, who gathered data for his PhD. thesis in Sogin's laboratory.

Silberman also participated in this work when he as a post doctoral student in Woods Hole, but he has since moved to UCLA where he works with Pat Johnson as part of the UCLA NAI research activities.

Rudi Amann from the Max-Planck-Institute for Microbiology (Bremen, Germany) collaborated with us on 18S rRNA probe development for Ancyromonas.

NASA Mission Involvement

This research provides basic information about the evolutionary history of life on this planet. Our understanding of the evolution of complex metazoans is reasonably well described, but we still have only a poor understanding of basal metazoan lineages. Furthermore information about detailed relationships both between and among animals, fungi, and plants is sketchy.

Although our research has no direct connection with specific missions planned by NASA, it is likely that someday we will visit extraterrestrial sites where multicellular forms are capable of survival. If complex life has arisen on other planets, knowing the evolutionary history of multicellular forms on Earth will be critical in the possible interpretation of multicellular fossils from extraterrestrial sources.

Future Directions

- Continued accumulation of large subunit sequences should increase our understanding of close branching taxa in the eukaryotic crown. During the next months we will focus our attention on several protists with special attention on species related to Nucleria sp.

- The ribosomal RNA phylogeny of Ancyromonas will be complemented by an ultrastructural analysis, in particular with respect to the phylogenetically informative mitochondrial chritae structure of this organism, in collaboration with David Patterson.

- Sequencing of the mitochondrial genome of Ancyromonas will be attempted, for a comprehensive phylogenetic study and placement of Ancyromonas in organelle evolution.

Roadmap Objectives

#2
Origin of Life's Cellular Components

#4
Genomic Clues to Evolution

#6
Microbial Ecology

Project

Genes That Regulate Photosymbiotic Interactions

Senior Project Investigator(s):
R. Gast

Accomplishments

We are interested in understanding how photosymbiotic relationships are controlled through the genetic communication of two cells, and perhaps how this may relate to the evolution of organelles. The presence of an algal symbiont represents the acquisition of novel functions by the host cell. There is an increasing body of evidence for the eukaryotic origin of chloroplasts in cryptophytes, chlorarachniophytes, euglenophytes, heterokonts, haptophytes and dinoflagellates. In these different hosts, the endosymbiotic algae are in various states of genetic and physical reduction. In all cases it is impossible to remove the alga from its host and culture it independently.

Planktonic foraminifera, radiolaria and acantharia are ameboid protists that occur in the pelagic environment of the world's oceans. Many of the species in this group harbor algal symbionts, particularly dinoflagellates. These symbiotic relationships have not evolved to the point where the symbiont is dependent upon the host, so it is possible to separate the two and study the changes in algal gene expression that occur when the free-living organism becomes a symbiont. The sarcodine symbioses will also allow the comparison of symbiotically regulated genes in taxonomically distinct algae, which can lead to the identification of the common genetic elements involved in the associations, as well as what makes each one specific.

In order to reach our overall goal of determining the genetic mechanisms behind algal symbioses and, potentially, organelle evolution, we have begun comparing the mRNA expressed in symbiotic and free-living dinoflagellate symbionts from the radiolarian *Thalassicolla nucleata*. We have cultures of the dinoflagellate (*Scrippsiella velellae*) in our laboratory, which serve as a source of free-living RNA. To obtain RNA from the symbiotic state of *S. velellae*, we collected intact symbioses and dissected the symbionts away from the host. RNA was isolated from cultures and dissected symbionts using Ambion's RNAqueous Kit. The two samples of RNA were processed further by suppression subtractive hybridization. Messenger RNA is converted to cDNA, and hybridization of cDNA tester (symbiotic cDNA) with cDNA driver (free-living cDNA) results in a population of cDNAs enriched for those of the tester (symbiotic). The differentially expressed cDNAs are amplified, and the PCR products are cloned and screened for confirmation of differential expression. This process enriches/selects for genes that are differentially regulated and is available in a kit format from Clontech.

We have completed a subtractive selection for *S. velellae*, and have recovered over 20 differentially expressed clones. Sequence analysis of these clones has indicated that 11 are essentially identical (C4). Although their BLAST searches recover unidentified human chromosomal DNA as the most similar sequences, there is actually not a significant match to anything in the database, potentially indicating a novel gene sequence. Another clone (F7) was analyzed and found to be similar to the gene for Cyplasin S. The matching region is actually a repeat unit, potentially indicating a conserved function or structure.

Highlights

This is one of the first studies to pursue the differential expression of algal genes in photosymbiotic relationships. Although we have only recovered a few genes at this time, we have made significant progress considering that these methods have never been applied to this system or these organisms before. All of the work that we have accomplished is breaking new ground with these organisms.

Field Trips

Cruise on R/V Endeavor , August 17-31, 2000.

The first field collection was an ancillary project on a cruise supported by NSF

Biological Oceanography on a grant to Gast. This cruise took place August 17-31, 2000 onboard the R/V Endeavor. Gast and Beaudoin collected planktonic sarcodines for the isolation of algal symbionts from intact photosymbiotic associations.

Cruise, March 19, 2001.

The second field collection took place on March 19, 2001, again as an ancillary project on a cruise supported by NSF Microbial Observatories as a grant to Caron and Furhman at the University of Southern California. Gast collected planktonic sarcodines on this cruise, again for the isolation of algal symbionts.

Catalina Island, California, June 18-July 3, 2001.

The third field collection took place on June 18 - July 3, 2001, at the University of Southern California field station on Catalina Island. Again, Gast participated in daily cruises to collect planktonic sarcodines.

NASA Mission Involvement

One of the major events in the evolution of eukaryotic cells was the acquisition of endosymbionts and their subsequent transformation to organelles. By studying the regulation of gene expression in symbiotic relationships, we plan to gain insight into how symbionts are acquired and maintained, why certain symbioses form, and what the early steps in organelle formation may be.

This information could potentially help us to understand more about how eukaryotic life on Earth evolved. It would provide insight to help plan missions in the search for life beyond Earth.

Future Directions

- Confirm gene expression in symbiotic state: We will pursue experiments to confirm that the gene sequences we have isolated are only expressed in the symbiotic state of the alga. This will be accomplished by Reverse Transcriptase-PCR of isolated RNA.

- Perform another round of screening for more putative symbiosis specific genes: We will perform one more round of subtractive screening to try and recover more candidates for symbiotically-expressed genes.

- Recover the full length cDNA for symbiotically-expressed genes: We will attempt to recover the full gene sequences for candidates that have been confirmed to be expressed symbiotically. This is necessary for potential identification of gene function, recovery of genomic sequences and for future studies of regulation.

Hyperthermophiles of the Hydrothermal Vent Subsurface and Their Environmental Tolerance

Project

NAI Postdoctoral Fellow: V. Edgcomb
Advisor: A. Teske

Roadmap Objectives

#6
Microbial Ecology

#7
Extremes of Life

#9
Life's Precursors & Habitats in the Outer Solar System

Accomplishments

Hyperthermophilic archaea, isolated from deep sea hydrothermal vents, survive and grow under exposure to extreme heat, pressure, and chemical toxicity. It has been assumed that their environmental tolerance range sets the extreme limits for life, particularly in deep, hot subsurface environments. However, many critical factors and components of this tolerance have not been systematically tested. By defining the critical chemical and physical environmental extremes that limit growth and survival of hydrothermal vent archaea we can determine the likelihood of a deep subsurface biosphere – a possible analog to extraterrestrial habitats. We will also define the conditions under which these organisms can be found, which has applications not only for understanding the ecology and evolution of prokaryotic life on Earth, but for searching for comparable life on other planets. Growth and survival of selected hydrothermal vent archaea are being tested under relevant (simulated *in situ*) environmental conditions - acidic pH, high pressure and temperature, and high sulfide and metal concentrations.

We have completed experiments to determine the limits of tolerance to pressure, acidic pH, and temperature, of two hyperthermophilic, heterotrophic (elemental sulfur reducers) archaea. Both *Pyrococcus* strain GB-D and *Thermococcus fumicolans* are capable of growth at 90°C and pH down to 4.5 at both one atmosphere and 250 atm pressure over a time period of 24 hours. At 100°C and 250 atm, *Pyrococcus* and *Thermococcus* grow at pH down to 5.5 and 4.5, respectively. The deep subsurface is a very heterogeneous environment, and hydrothermal activity can displace organisms temporarily from their niches where conditions are favorable for growth. We tested survival of these two organisms under starvation conditions, and determined that both organisms can survive at 90°C and pH 4.5 at one and 250 atm pressure, however survival decreased with pressures of 250 atm or more. At 100°C, only *Pyrococcus* is able to survive. Interesting synergistic effects of temperature, pH and pressure have been noted for these two organisms. The conditions which define the limits of tolerance of these organisms for growth and survival will be used as a baseline for upcoming experiments incorporating sulfides and metals. Experiments will begin this fall on the remaining two archaea.

Progress to date is in line with goals set in original proposal.

Highlights

• We are testing growth and survival of selected hydrothermal vent archaea under relevant (simulated *in situ*) environmental conditions – high pressure and temperature, acidic pH, high sulfide and metal concentrations; individually and together in ways that approximate natural conditions. These combined tests represent a novel approach to determining physiological tolerance limits of life on Earth. The resulting physiological tolerance profiles of hyperthermophilic archaea will more precisely define their range in the hot, toxic subsurface, and the potential of these organisms as analogs for extraterrestrial microbial life.

• Two cultures of heterotrophic hydrothermal vent archaea, *Pyrococcus* and *Thermococcus*, are capable of growth at 100°C and 250 atm, and acidic pH's of 5.5 and 4.5, respectively.

• The deep subsurface is a very heterogeneous environment, and hydrothermal activity can displace organisms temporarily from their niches where conditions are favorable for growth. Under starvation conditions, both organisms can survive at 90°C and pH 4.5 at one and 250 atm pressure, however survival decreased with pressures of 250 atm or more. At 100°C, only *Pyrococcus* is able to survive. Interesting synergistic effects of temperature, pH and pressure have been noted for these two organisms.

NASA Mission Involvement

Hydrothermal vent environments may serve as a possible analog to extraterrestrial habitats. By defining the critical chemical and physical environmental extremes that limit growth and survival of hydrothermal vent archaea, we can define the conditions under which these organisms can be found. This has applications for directing searches for comparable life on other planets, as a part of future NASA missions.

Future Directions

• Test limits of tolerance to acidic pH, temperature and pressure as above for the chemolithoautotrophic or mixotrophic archaea, *Methanococcus jannaschii* and *Archaeoglobus profundus*.

• Test effects of increasing exposure to sulfide and various metal ion concentrations both individually and synergistic effects, for all four organisms. These experiments will be run at optimal growth conditions, and under conditions of acidic pH, temperature and pressure that have been empirically determined to represent their limit for growth and survival.

micro*scope (http://www.mbl.edu/microscope) New Internet Resources for Microbial Biodiversity

Project

Senior Project Investigator(s):
M. Sogin, D. Patterson, V. Edgcomb

Roadmap Objectives

#2
Origin of Life's Cellular Components

#6
Microbial Ecology

Accomplishments

On May 20th 2001, at the American Society for Microbiology meeting, micro*scope (an innovative web-based bioinformatics resource for microbiology) was launched. **Micro*scope** promotes the assembly and use of information on microbial biodiversity using the internet. This initiative was sponsored by NAI, and has been developed at the MBL NAI (Bay Paul Center, Marine Biological Laboratory, Woods Hole). Micro*scope has educational and biodiversity bioinformatics dimensions. It has been made possible by the use of Biose-IT software developed in-house. Educationally, micro*scope provides resources for educators and students in the form of images, text, talks, LUCID identification guides and a classification structure.

To achieve its bioinformatics objectives, micro*scope does not adopt the more usual strategy of establishing a large centralized database. Rather, it recognizes that names are the universal currency of all taxonomic information. With a database of names, micro*scope can exploit the internet to gather, collate and integrate data from different sources. As entities (species) can have more than one name, the core database has been developed to include all possible names with links to the underlying entities (species). Micro*scope contains generic names of all protists and prokaryotes. Micro*scope currently demonstrates its ability to link data throughout the internet by recovering information from a few selected web sites. Micro*scope includes approximately 3GB of information, inclusive of 1000 images, a classification with 15,000 names; and has approximately 30 contributors.

Highlights

Publication of http://www.mbl.edu/microscope - a visually attractive internet resource for educators.

Cross Team Collaborations

This activity is supported by the Education and Public Outreach (EPO) program within NASA. Thus, we interact with NASA Headquarters and other EPO program developers in our project endeavors.

Future Directions

(1) A suite of 20-24 LUCID identification aids; a system of hierarchical online keys using the model of the three pilot keys currently available through micro*scope (Funded by NAI EPO)

(2) A site management and maintenance structure, for which independent funding is being sought from NSF

(3) To develop micro*scope to integrate traditional ecological data (environmental preferences, biogeography) with environmental genomics for which independent funding is being sought

Roadmap Objectives

#2
Origin of Life's Cellular Components

#4
Genomic Clues to Evolution

Project

Origin of Life: Evolution of Proteins

Senior Project Investigator(s):
M. Riley

Accomplishments

We continue to make steady progress toward enumerating and characterizing the basic protein families that were present in the last common ancestor on Earth. Also our study of the genotype and phenotype relationships for all genes of a single microorganism continues to give information on exactly how a collection of genes in the DNA of an organism translates into a complete, free-living self-reproducing cell. The complete genotype down to DNA sequence is known for *Escherichia coli*. The phenotype conferred by some of these genes is experimentally known, for others, predicted and for others still unknown.

The organism on Earth that we know the most about is *Escherichia coli*. This single-cell bacterium has been minutely studied experimentally for the last 60 years and more. We have collected information on the biological function of each one of the 4408 genes and their gene products (protein or RNA). We have assembled experimentally known information from the literature and we have painstakingly assigned functions to unknown proteins by analogy to similar known proteins. The function of 19% of the genes/gene products is not yet known. However, by careful study of the 81%, we documented over half as experimentally known, the rest predicted. This body of work constitutes an unparalleled opportunity to see in ultimate detail what exactly makes a living cell. A complete knowledge of the relationship of genotype to phenotype for all genes of one organism has implications for knowing the entirety of what is required to fire primitive life.

Also, we have been studying protein families in the context of determining the makeup of the basic protein elements that were adequate to give life to a primitive cell on early Earth. Knowing the basic composition of that early cell should provide clues to the kinds of proteins we might expect to detect on other solar system bodies if life there is based on carbon, oxygen and nitrogen.

To work toward this goal, we have grouped proteins into families both by amino acid sequence similarity and by elements of tertiary structure. The two approaches, sequence and structure, generate some families of proteins in common, and also some families seen only by sequence or only by structure. We have found that some protein families are uniform in biological action, differing only in specificity of molecules acted on. The origins of such families are easy to visualize in terms of molecular evolution as being the consequence of gene duplications followed by change in substrate specificity. Other protein families, however, are more complex. Our analysis has shown that some families, called superfamilies, can be more varied in function. In these cases replicate genes have used the same basic themes of sequence and structure to branch out to a variety of functions of the same general type. We have studied examples in which different enzymes in the same superfamily catalyze only remotely related reactions. Even more extreme are examples in which a single protein uses only one catalytic site but different amino acid residues at that site to catalyze different reactions by the same protein. This model of efficiency and economy could be characteristic of early evolution, giving more catalytic capacity to a single gene product.

In such ways we are reconstructing molecular modes of evolution in order to be prepared to understand comparable events that may have occurred on other bodies in the solar system.

Highlights

- Identification of the functions of 81% of the genes of *E. coli*. We have assembled a nearly complete picture of the essential link between genotype and phenotype for one simple model organism.

- Analysis of the multimodular composition of some genes in *E. coli*. Multimodular genes are multifunctional. These functions are often physiologically related. A given gene's components often have other partners, also functionally related, in other organisms. The role of gene fusion in evolution may be achieving efficiency in localizing related reactions in physical proximity, not in equilibrium with the aqueous phase of the cell.

Cross Team Collaborations

With this project, interchange takes place with Steven Benner at the University of Florida (Gainesville, FL), especially on the modular structure of proteins.

One visit by M. Riley to Gainesville and one visit by Benner to MBL took place this past year. Although Benner's emphasis is on more recent time in evolution than is the case for Riley's work, the important question of modular composition of proteins has been and continues to be addressed by both groups.

NASA Mission Involvement

Ultimately, one hopes to be able to design useful experiments and detection instruments applicable to material from other planets or their moons. With a better picture of the biology and biochemistry of very early life on Earth, we can use this information to help shape the questions we will ask of materials derived from outer space.

Future Directions

- Compare all protein families with 8 or more members that are present in many of the organisms whose genomes have been fully sequenced, in order to identify common families that seem likely each to have descended from a very early ancestor. The goal is to form a picture of the collection of proteins present from early stages of life on Earth.

- With data from above, identify all multimodular proteins that have undergone fusions to different partners in different branches of the tree of life. Some examples are now known anecdotally but systematic analysis should give a coherent picture of the paths taken in recombinational joining of simple unimodular ancestral genes to make different multimodular genes extant in the various branches of phylogeny.

Roadmap Objectives

#7
Extremes of Life

Project

Physiological Regulation of Cytosolic pH in a Eukaryotic Acidophile

NAI Postdoctoral Fellow: M. Messerli
Advisor: M. Sogin

Accomplishments

Survival in acidic, heavy metal rich environments either requires molecular modifications to maintain conserved metabolic processes or the development of new mechanisms to replace these conserved processes. Using a physiological approach we have started screening eukaryotes of the Rio Tinto in Spain (pH 1.7-2.5) to identify the molecular alterations used by these organisms to thrive at low pH. We have measured a cytosolic pH range of 6.2-7.2, with a mean of 6.6 for two autotrophs, a Chlamydomonad and Euglenoid, showing that these organisms protect their cytoplasmic components from the acidic extracellular environment. As lipid bilayer membranes are 6-7 orders of magnitude more permeable to H^+ than metal cations, maintaining this H^+ gradient appears a formidable task. We proposed that these acidophiles employ either a higher activity of energy-driven H^+ transporters at low pH to remove H^+ ions that have leaked into the cell or have developed barriers to greatly reduce the membrane H^+ permeability. We have modified the self-referencing O_2 electrode to function at acidic pH so that we can use O_2 consumption as an indicator of ATP production. We find that loading the cytosol of the Chlamydomonad with H^+ from weak acids, accompanies increased O_2 consumption but that no such increase occurs for the cells at pH 2 compared to pH 7 in the absence of weak acids. This is the first evidence showing that an acidophile does not have a higher energy demand to survive at pH 2 than at pH 7. With this information we suggest that the cells are using a barrier-like mechanism to decrease H^+ diffusion across the plasma membrane. We are now attempting to elucidate the nature of this barrier and determine whether it extends away from the bilayer surface so that it can, in fact, protect membrane surface proteins as well as the cytoplasm.

Highlights

- Acidophiles can maintain a neutral cytosolic pH with an extracellular pH of 2. Previous reports using radiolabelled weak acid partitioning, which is at best a measurement of the average cytoplasmic pH, place the cytosolic pH of acidophiles between 4.25 and 7. We used a fluorescent assay to ensure that we are in fact measuring cytosolic pH and not the pH of organelles.

- Acidophiles do not necessarily have higher oxygen demands at low pH. We interpret these results to mean that the acidophiles do not necessarily have higher energy demands at acidic pH and are therefore using passive mechanisms to slow H^+ diffusion across the plasma membrane.

- We have modified the polarographic probe to function at acidic pH. Self-referencing technology increases the sensitivity of measurements by 10-1000 times. Modifying the polarographic probe has given us heightened sensitivity to measure redox reactions at acidic pH.

Field Trips

Spain, Rio Tinto, September 2001.

Co-Investigator, Linda Amaral Zettler (Marine Biological Lab), will make another trip to the Rio Tinto under the guidance of Ricardo Amils (Centro de Astrobiologĺa) to collect more samples in September, 2001. Of the organisms that we can maintain in culture we will measure the cytosolic pH and sequence the H^+ transporters to determine the sequence diversity to acid tolerance of the protists in the river.

NASA Mission Involvement

At first glance, it appears that acidophiles do not have high energy demands and that they protect the cytosolic environment using passive rather than active mechanisms. This begins to give us an idea of what sorts of energy requirements are needed by organisms that may exist in acidic environments on Mars (Fe^{+3} -produced acid) or on Europa (H_2SO_4). It also gives us ideas regarding general adaptation mechanisms. Passive mechanisms for tolerance are energetically favorable to active mechanisms. This information aids the search for life by giving us ideas of where to look and what to expect to find across different environments.

We also learn technical advances for preparing sensing and collecting equipment for use on NASA missions. We are developing methods for culturing extreme organisms that gives us experience for maintaining cultures of extraterrestrial life. Also, certain measuring equipment such as the self-referencing polarographic probe, that we use to measure O_2 fluxes from acidophiles, did not work at a pH less than 4. Modifying the probe has given us the ability to measure redox reactions with heightened sensitivity at acidic pH.

Another example involves our new understanding of ferric iron as an acid. Depending on the state of the iron on Mars, water and ice there may be very acidic. This will corrode, if not dissolve, metals in search equipment. Preparing acid-tolerant equipment for Mars may not be as important as for Europa. There, sulfuric acid has been detected, and this is a much stronger acid than iron.

Future Directions

- Determine the acidophile membrane permeability to H^+ using the self-referencing H^+ probe under tightly controlled conditions and under 'natural' conditions. Experimentation under tightly controlled conditions will allow comparison of acidophilic bilayer permeability with membranes from nonacidophiles while measurements under natural conditions will show us really what goes on in the native environment. Plasma membranes will be extracted, purified and reconstituted into planar lipid bilayers. H^+/OH^- flux will be directly measured with the self-referencing H^+ probe. This technique has a wide dynamic range of sensitivity, unlike the fluorescent dye technique, and measures H^+ flux as a concentration change (direct) rather than as an electrical conductance (indirect). The bilayer is the first, most prominent source for a passive H^+ barrier.

- Identify the plasma membrane H^+ transporter(s), the inward rectifying K^+ channel and the light activated Ca^{2+} channel and characterize their pH tolerance. Transporter and channel modifications used to function at low pH are not only relevant to acidic adaptation in the Rio Tinto but also to pathological conditions of the stomach and parasitic infection. With inside-out plasma membrane microsomes we will use physiological inhibitors of the known H^+ transporters and ion substitution, in combination with patch clamp recording, to characterize the transporter(s). Using these inhibitors on intact cells has been difficult as most are not soluble at low pH or simply do not pass into the cytosol. Voltage clamp of microsomes resuspended in planar bilayers will enable us to characterize the pH optimum and ion selectivity of the channels under acidic conditions and to understand how selectivity is maintained. We are in the process of cloning these transporters and possibly the ion channels to determine the potential sites of sequence modifications enabling function at low pH.

Protist Diversity in Extreme Environments

Project

Senior Project Investigator(s):
R. Gast and D. Caron

Roadmap Objectives

#6
Microbial Ecology

#7
Extremes of Life

Accomplishments

Natural microbial communities living at the limits of physiological tolerance in extreme environments exist in a variety of marine ecosystems. There has been speculation that some of these environments are similar to physiochemical conditions of primitive Earth or to conditions that are (or were) present on other planets and moons. Thus, studies of microorganisms from extreme environments on Earth may provide insight into early evolutionary events.

Our research on the microbial eukaryotic diversity in extreme environments employs both traditional culture techniques and culture-independent molecular methods. This past year our project has focused on the analysis of deep-sea sediments from the Antarctic. We chose to work with these samples because they can provide a complementary picture to the protistan diversity studies at hydrothermal sites. The non-hydrothermal deep sea tends to be sparsely populated, but is certainly not sterile. The microbial (both prokaryotic and eukaryotic) populations in these areas potentially harbor organisms with different growth strategies. There has been very little work on the microbial populations in this area, an ecosystem that may share characteristics with the deep ocean floor of Europa.

We have been pursuing denaturing gradient gel electrophoresis (DGGE) analysis of the protistan community from the deep sea sediment samples that we collected from a previous Antarctic cruise. It has been very difficult to obtain good banding patterns for these samples, but we have been optimizing our PCR amplification parameters with some success.

In addition to direct genetic analyses, we have recovered protistan cultures from these sediments. Most of our cultures are ameboid protists, but we have also recovered small flagellates and small choanoflagellates. We are planning to compare small subunit ribosomal gene identification of these cultures with ribosomal sequences that we (eventually) recover directly from the sediments to determine whether these cultured protists might be representative of organisms originally in the samples.

Highlights

This work is the first genetic analysis of microbial communities of non-hydrothermal deep sea sediments, and coupled with the analysis of recovered cultures, may potentially yield insights into how this very understudied ecosystem functions.

Field Trips

Samples of ice, water, slush and deep sea sediment for genetic and physiological studies of psychrophillic protists were collected as part of an NSF-sponsored cruise onboard the icebreaker Nathaniel B. Palmer in the Ross Sea, Antarctica. The cruise took place December 1998 through February 1999, and the participants for our group included Rebecca Gast (WHOI), David Caron (USC), and Mark Dennett (WHOI).

NASA Mission Involvement

Phototrophic and heterotrophic protists are essential components of all aquatic ecosystems. Cold water ecosystems are predominant features of the world's oceans because the vast majority of this water is permanently below 2-5°C. Adaptation to extremely low temperature is therefore a fundamental aspect of marine ectothermic species.

Fundamental to understanding the life processes of a major portion of the living biomass on our planet is understanding the structure, diversity, and evolution of cold water protistan communities, along with the adaptations allowing these assemblages to flourish. Our genetic and physiological studies with protists from extreme low temperature environments are also timely with regard to the general consensus that a liquid ocean exists under the icy surface of Europa. Studies of microorganisms living at extremely low temperature potentially have relevance to the evolution and presence of extraterrestrial life on this moon of Jupiter.

Future Directions

- <u>Continue genetic analyses of deep sea sediments</u>: We will continue to try to utilize DGGE (denaturing gradient gel electrophoresis) for the analysis of these samples. We will also target several samples for full length ribosomal clone library analysis.

- <u>Genetically identify cultures</u>: We will sequence the small subunit ribosomal gene from several unique cultured protists. These sequences will be compared to those recovered from the original sample.

Relationship of Genetic Changes to Phenotypic Changes in Organism–Environment Interactions

Project

Senior Project Investigator(s):
M. Cummings

Roadmap Objectives

#4
Genomic Clues to Evolution

Accomplishments

One of the research questions of the NASA Astrobiology Institute is, “How do genetic changes produce phenotypic changes?” This research program seeks to answer this question by using the genetic basis of spectral tuning in animal color vision as a model system. We are sequencing genes for opsins, the protein responsible for color vision, from dragonfly species, which are a facile study system. We collected specimens and isolated poly-A messenger RNA from an additional 72 species bringing the total to 88 species. The new species collected in this reporting period come from two expeditions: Maine, USA, 34 species; and Queensland, Australia, 28 species. The taxa represent a broad sampling of Odonata (Dragonflies): 2 suborders; 11 families; and 52 genera. We obtained complete sequences (for the gene region we are studying) from 26 species, and partial sequences from 9 additional species. These sequences include several distinct opsin types from several species. We are currently collecting sequence data from the new specimens.

Building upon earlier preliminary work, we have completed analyses and model building for the relationship between peptide sequence variation and spectral tuning for 107 vertebrate opsins. This year we will pursue a similar research plan for arthropod opsins and compare the models across these broad taxonomic groups.

The opsin sequence database has been rebuilt to increase flexibility and performance. We used Tangram to include object-oriented design features, Mason to better integrate Perl with the HTML and MySQL as the underlying database system. The database continues to be tested and revised.

Highlights

We completed the largest and most comprehensive study of spectral tuning in vertebrate color vision. Furthermore this work has demonstrated the successful application of novel analytical techniques related to those that have been applied to space shuttle landing to genotype-phenotype relationships. The study has three principal findings: few of the hundreds of variable amino acid positions in vertebrate opsin have an influence on wavelength of light absorbed; several positions appear to be equivalent, or indistinguishable because they co-vary, with regard to their influence on spectral tuning; and most of the variance in spectral tuning can be explained by two or three amino acid changes.

Field Trips

Maine, USA, July 2000.

Michael P. Cummings and Laura A. McInerney; collection of specimens.

Queensland, Australia. May, 2001.

Michael P. Cummings and Laura A. McInerney; collection of specimens and laboratory work.

NASA Mission Involvement

With specific regard to color vision, our studies suggest light wavelength boundaries that might reasonably be applied in assessing the potential of extraterrestrial environments for supporting vision of complex organisms.

More generally, the development and validation of analytical methods for genotype-phenotype relationships establishes a rigorous design and analysis model for mission-based biological experimentation. Our models provide a means for analytically isolating genetic factors from other influences on phenotype such as environment, epigenesis, and experimental error. One consequence is that *phenotypic* effects of space environments can be more properly characterized and distinguished from genetic effects.

Future Directions

The general future directions are to continue laboratory and computer-based aspects of the research. Specific goals include the following:

- Determine opsin sequences for more species, including those collected during recent expeditions

- Construct model for spectral tuning in arthropod opsins using novel analytical techniques related to those that have been applied to space shuttle landing

- Compare vertebrate and arthropod spectral tuning models to determine general taxon-independent genetic influences on color vision

- Apply arthropod model to sequences determined in laboratory, and in doing so, set up specific hypotheses for future laboratory research to validate models

- Continue development work on opsin database for both use in this project and as a generalized system for use in other astrobiology projects involving sequence data

Education & Public Outreach

Career Day Presentation: "My Career - The Classroom Connection"

Linda Amaral Zettler, an NAI Co-I, gave a 20 minute career talk about being a scientist and astrobiologist to five groups of 20 students each at a local middle school.

Lectures/Presentations

Eight lectures and presentations related to the **micro*scope** astrobiology education project developed at the Marine Biological Laboratory, were given by Co-investigator David Patterson. A summary follows: North East Algal Society, Plymouth, Massachusetts; American Society for Microbiology, Orlando, Florida; East Coast Section Society of Protozoologists, Plymouth, Massachusetts; American Society of Phycologists, Estes Park, Colorado; Commonwealth Science and Industry Research Organization (CSIRO), Canberra, Australia; Biosciences in Asia Conference, Sanctuary Cove, Australia; two seminars at James Cook University, Townsville, Australia.

Life on Earth and Elsewhere? Teacher Workshop

This one and a half day hands-on workshop combined lectures on snow algae as a model for life on Mars, extremophiles and the evidence for water on Mars and Europa with hands-on activities drawn from the NASA Astrobiology Institute's Educator Resource Guide (ERG). The **micro*scope** website was also demonstrated.

Living in the Microbial World Teacher Enhancement Workshop

This intensive one week workshop for middle and high school teachers combines hands-on activities in microbial diversity and evolution with lectures and demonstrations from visiting and resident research scientists.

Micro*scope

Micro*scope is an internet resource that promotes microbial diversity with an emphasis on educational materials. It contains an extensive image database of high quality, downloadable digital images of microbes from a variety of environments along with taxonomic, habitat and morphological descriptions of each. Information in the site can be accessed in a variety of ways, including alphabetically, by habitat, by taxonomic affinity, and through the use of interactive LUCID non-dichotomous keys. Also accessible are a complete taxonomic listing of protist families and educational materials including Powerpoint presentations related to protist evolution and microbial diversity. This site will continue to expand to include images of microbes from environments being studied by various astrobiology researchers.

NSTA presentation

An hour long hands-on workshop was given at the National Science Teachers Association annual meeting in St. Louis, MO, entitled "Living in the Microbial World: Hands-on activities in microbial diversity and astrobiology". The presenters, Susan Rutland and Dee Wilkinson provided information on the Woods Hole workshops and the NASA Astrobiology Program and presented concepts related to microbes as the most abundant and little understood inhabitants of our planet as well as the relevance of microbial diversity to astrobiology. Participants were given examples of various methods of capturing microbes, and created low cost plankton nets for their own classrooms.

Presentation to teachers: Origin of Life and Proteins

This was a presentation by Co-I Monica Riley on the origin of life and the origin of proteins to 28 high school teachers from Connecticut.

Presentations to High School Students

Linda Amaral Zettler gave a career talk on her astrobiology research in a presentation entitled "From the Ocean to the Rainforest: A Closer Look at Biodiversity" to high school students at the Massachusetts Marine Educators High School Marine Science Symposium. Carmen Palacios and Jennifer Wernegren presented talks on the use of aphids and ants as models of symbiosis and evolution to students visiting their laboratory at the Marine Biological Laboratory.

Professional Development Workshop: Molecular biological approaches to looking at microbial diversity and evolution

This event offered two one-day professional development workshops for Massachusetts secondary science teachers. The subjects of the workshops were molecular biology tools used to address topics of interest to astrobiologists, microbial diversity of extreme environments, and early evolution of eukaryotes. The workshops consisted of lectures, tours of laboratory and DNA sequencing facilities, and lab bench work.

A similar workshop was given for 11th grade biology students at Falmouth Academy in Falmouth, MA. The workshop consisted of two sessions with 20 students per session.

Script reviewer- DNA Files Radio Program on Astrobiology

Lorraine Olendzenski has been serving as a script reviewer for a one hour segment on astrobiology that is part of "The DNA Files", a radio series for National Public Radio.

Workshop on Molecular Evolution

This workshop, designed for graduate students, faculty, postdocs and researchers, consists of a series of lectures, demonstrations and discussions that cover various aspects of molecular evolution including questions about early genome and microbial evolution on Earth. A distinguishing feature of the workshop is a well-equipped computer laboratory for comparative analysis of molecular data. The laboratory is equipped with Linux and Unix workstations and servers. Authors and experts in the use of computer programs and packages such as Clustal W and Clustal X, COMPARE, FASTA, GAML, GCG, LAMARK, MacClade, Mesquite, PAUP, and PHYLIP provide demonstrations and consultations. Topics include phylogenetic analysis, molecular evolution and genomic comparisons.

Publications

Back to Table of Contents

Diversity and Physiology of Prokaryotes in Selected Thermophilic and Mesophilic Environments That Might Resemble Early Earth's Biosphere

Teske, A., Hinrichs, K-U., Edgcomb, V., de Vera Gomez, A., Kysela, D., Sogin, M. & Jannasch, H. (In Preparation, 2001). Archaeal and bacterial population structure of hydrothermal sediments at the Guaymas Basin vent sites derived from 16S rRNA sequence and C13 analysis of archaeal and bacterial lipids: Indications for anaerobic methanotrophy. *Applied and Environmental Microbiology*.

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Diversity of Eukaryotes in Thermophilic and Mesophilic Environments that might Resemble Early Earth's Biosphere

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Eukaryotic Diversity in the Rio Tinto: Spain's Acidic/High Metal Extreme Environment

Amaral Zettler, L.A., Amils, R., Gomez, F., Keenan, B., Sogin, M.L. (2001). Eukaryotic diversity in an acidic, metal-rich environment: Spain's Tinto River [Abstract]. *General Meeting of the NASA Astrobiology Institute* (p. 147), Carnegie Institution of Washington, Washington, DC.

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Eukaryotic Origins and the Evolution of Cellular Complexity — Eukaryotic rRNA Evolution: Early Diverging Eukaryotes

Amaral Zettler, L.A., Nerad, T.A., O'Kelly, C.J., Peglar, M.T., Gillevet, P.M., Silberman, J.D. & Sogin, M.L. (2000). A molecular reassessment of the *Leptomyxid amoebae*. *Protist*, 151: 275-282.

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Back to Table of Contents

Pennsylvania State University

NASA Astrobiology Institute

Pennsylvania State University Team Members

Ariel D. Anbar
University of Rochester

Michael A. Arthur (PSU)

Mark Barley
University of Western Australia

Michael Bau (PSU)

Ekaterina Bazilevskaya
Pennsylvania State University

Jaime E. Blair (PSU)

Michael J. Borda
SUNY, Stony Brook

Philip Borkow (PSU)

Birthe Borup (PSU)

Susan Brantley (PSU)

Paul Braterman
University of North Texas

Jean Brenchley (PSU)

Jochen Brocks
University of Survey

Lisa L. Brown (PSU)

Tom Bullen (PSU)

Heather Buss (PSU)

Tom Canich (PSU)

Rosemary Capo
University of Pittsburgh

Oliver A. Chadwick
University Of California Santa Barbara

Hsiong Chen (PSU)

Deneyse J. Churchill (PSU)

George D. Cody (CIW)

Linda M. Decker (PSU)

Edward DeLong
Monterey Bay Aquarium

Theresa Diehl (PSU)

Catherine L. Drennan
Massachusetts Institute Of Technology

Amy Eastwood
University of Virginia

Brooke Eidell (PSU)

Jennifer L. Eigenbrode (PSU)

James Ferry (PSU)

Marc Fiddler (PSU)

Sorel T. Fitz-Gibbon
University of California Los Angeles

Katherine Freeman (PSU)

David Geiser (PSU)

Galina Glazko (PSU)

Cathy Gross (PSU)

Robin Guynn (PSU)

Daniel S. Heckman (PSU)

S. Blair Hedges (PSU)

Ellen Herman (PSU)

Achim Hermann (PSU)

Kai-Uwe Hinrichs
Woods Hole Oceanographic Institute

Christopher House (PSU)

Matthew Hurtgen (PSU)

Gary Icopini (PSU)

Takeshi Itoh (PSU)

Daniel Jones (PSU)

Hilary Justh (PSU)

Frank Kachurak (PSU)

Takeshi Kakegawa
Tohoku University

Natalie L. Kardos (PSU)

James F. Kasting (PSU)

Alan Jay Kaufman
University of Maryland

Pushker Kharecha (PSU)

Brian Krapez
University of Western Australia

Andrew Z. Krug (PSU)

Lee R. Kump (PSU)

Andrew C. Kurtz (PSU)

Antonio C. Lasaga
Yale University

Vanessa LeBrun (PSU)

Laura Liermann (PSU)

Graham Logan
Australian Geological Survey Organization

Kristy Longsdorf (PSU)

Gwendolyn Macpherson
University of Kansas

Andres Marin (PSU)

Clifford Matthews
University of Illinois

Kevin McKeegan (UCLA)

Jonathon Millen (PSU)

Jeffrey Miller (UCLA)

Robert D. Minard (PSU)

Joseph Minervini
University of Pittsburgh

Vanya Miteva (PSU)

Jim Moran (PSU)

Kimberly Moran (PSU)

Karl Mueller (PSU)

Hiroshi Naraoka
Tokyo Metropolitan University

Masatoshi Nei (PSU)

Hiroshi Ohmoto (PSU)

Bertil Olsson (PSU)

Shuhei Ono (PSU)

Victoria Orphan
Monterey Bay Aquarium Research Institute

Mark Patzkowsky (PSU)

Alexander Pavlov (PSU)

Robin Penfield
SUNY, Stony Brook

Angela M. Phelps (PSU)

Olena Piontkivska (PSU)

Laura Poling (PSU)

Anthony Prave
St. Andrews University

Janna Rauzin (PSU)

Douglas Rees
California Institute Of Technology

Amanda Reynolds
University Of Pittsburgh

Jessica Schendel
University of Puget Sound

Martin A. Schoonen
SUNY, Stony Brook

J. William Schopf (UCLA)

Mark Sephton
Planetary Sciences Research Institute

Peter P. Sheridan (PSU)

Erin Shields (PSU)

Kerry S. Smith (PSU)

Colin Snape
University of Nottingham

Phil Sorber (PSU)

Sherry Stafford
University of Pittsburgh

Rebecca L. Stauffer (PSU)

Brian W. Stewart
University of Pittsburgh

Neil S. Suits
Colorado State University

Roger E. Summons
Massachusetts Institute Of Technology

Amanda Thompson (PSU)

Dennis Walizer
University of Pennsylvania

Yumiko Watanabe (PSU)

Richard Wilkin (PSU)

Ping Xu (PSU)

Kosei E. Yamaguchi (PSU)

Faith Yarberry
University of North Texas

Aubrey Zerkle (PSU)

Executive Summary

Accomplishments

The Penn State Astrobiology Research Center (PSARC) is composed of 16 Full Members (PI and Co-I's) who are full-time faculty members of The Pennsylvania State University (Arthur, Brantley, Brenchley, Ferry, Freeman, Hedges, House, Kasting, Kump, Minard, Nei, Ohmoto, Patzkowsky), the University of Pittsburgh (Capo, Stewart), or the State University of New York at Stony Brook (Schoonen), plus their research staff (post-doctoral fellows, graduate and undergraduate students, technicians) and administrative staff. During the third year with NAI, a large number of people have been involved in the research activities of the PSARC, including 8 Research Associates, 27 graduate students, 12 undergraduate students, 2 technicians, 1 Administrative Assistant, and 2 IT assistants. We also have 26 active Associate Members who are collaborating with the Full Members, as well as many other researchers with our Co-Investigators. Currently, we also have 4 Associate Members who are in Education and Public Outreach, supporting PSARC activities.

The main goal of research at PSARC has been to increase our understanding of the connection (interplay) between the environment and the biota on Earth, especially during the early stage of evolution. Attainment of this goal will greatly enhance our ability to predict and identify life elsewhere in the Solar System. The above goal has been pursued primarily from multidisciplinary and multidimensional research focused on the following topics:

1. Environment of prebiotic Earth and the origin of life
    a. Experimental Approach (Schoonen, Kasting, Minard)
    b. Prebiotic chemistry of hydrogen cyanide (Minard)

2. Biochemistry of archaea and bacteria
    a. Enzymes of ancient metabolic pathways (Ferry)
    b. Biochemistry of psychrophilic organisms (Brenchley)

3. Microbe-mineral interactions (Brantley)

4. GEOPULSE: Gene Expression of Observations for Planetary Life Study (House, Ferry, Freeman, Brantley)

5. Timescale for the evolution of life on Earth: Molecular evolution approach (Hedges and Nei)

6. Evolution of atmospheric O_2, climate, and the terrestrial biosphere (Kump, Kasting, Ohmoto, Capo, and Stewart)

7. Causes and consequences of the diversification and extinction of metazoans
    a. Neoproterozoic variations (Arthur)
    b. Paleontological approach (Patzkowsky)

Excellent progress has been made in all phases of our research projects. A total of 48 papers related to NAI, excluding those in press, were published by the PSARC members during this year. The PSARC members also gave more than 82 presentations at various international/national scientific meetings.

Highlights

Martin Schoonen

- Presence of iron in the Hadean oceans may have been important in shielding organic molecules from UV irradiation.
  - Pyrite-water interaction may have led to the formation of hydrogen peroxide on the early Earth. The presence of hydrogen peroxide would have forced organisms to develop protection mechanisms early on. Eventually this may have led to the evolution of a reaction center capable of using hydrogen peroxide as an electron donor. This reaction center may have evolved into a water-splitting reaction center.

James G. Ferry

- The characterization of carbonic anhydrases from the methanoarchaea suggest these enzymes were essential in ancient autotrophic pathways for cell carbon biosynthesis.
  - The crystal structure of the first beta-class carbonic anhydrase from a procaryote lays a foundation for investigations into the evolution of carbonic anhydrases and the determination of their physiological roles in ancient microbes.

Susan L. Brantley

- We have shown that Fe dissolved from a soil mineral by siderophore-producing bacteria is shifted up to 0.8 ‰ lighter than Fe in the silicate. A smaller such isotopic shift is observed for Fe released abiotically by two chelates, and the magnitude of the shift increases with affinity of the ligand for Fe, consistent with a kinetic isotope effect during hydrolysis of Fe at the mineral surface. Fe released abiotically without chelates shows no isotopic shift. When recorded in the rock record, Fe isotopes could document Fe transport by organic molecules where such molecules were present in the geologic past.

- We have shown that microbes preferentially mobilize Mo, Ni, Cu, and other metals from silicates.

- We have shown that microbes can be removed from mineral surfaces using SDS, and this treatment does not alter the surface chemically or physically. After treatment with SDS, etching underneath microbes can be imaged.

Christopher House

- Methanogens can more significantly fractionate carbon isotopes than many other prokaryotes.

• We have directly identified an archaeal group of microorganisms that consumes methane. The technique developed, FISH-SIMS, will be an important technique for studying microbes that are currently unculturable.

• We also published the first paper demonstrating carbon isotopic analysis on Precambrian microfossils.

• Whole-genomic method of phylogenetic analysis of free-living microorganisms seems to be robust as more genomes are added.

S. Blair Hedges

Several discoveries were made with genomic analyses:

• evidence for a late origin of cyanobacteria, approximately 3 Ga

• evidence for multiple symbiotic events in the origin of eukaryotes, in contrast to the current single-symbiosis model

• evidence for a basal position of nematodes with respect to arthropods and vertebrates, in contrast to the widely accepted Ecdysozoa hypothesis joining arthropods with nematodes

• evidence for an early (Neoproterozoic) colonization of land by fungi and plants, possibly affecting global climate and the evolution of animals

Lee Kump

A link between mantle plumes, iron formation, glaciation, and the establishment of an oxygen-rich atmosphere was established. In a paper by Kump, Kasting and Barley (2001), which appeared in the new electronic journal of the American Geophysical Union, Geochemistry Geophysics Geosystems, they propose that plumes of the latest Archean brought oxidized mantle to the surface from near the core-mantle boundary, reducing the oxygen sink associated with the oxidation of reduced volcanic gases, and thus allowing for the buildup of oxygen released by cyanobacteria during photosynthesis. This oxidation event may also have triggered the first global Snowball Earth glaciation of 2.3 Ga ago. The article was highlighted in Nature with an article by Norman H. Sleep (Nature v. 410, pp. 317-319, 2001).

Hiroshi Ohmoto

• Discovery of the evidence for oldest life on land. We have demonstrated that the kerogen preserved in a paleosol in the East Transvaal district is a remnant of cyanobacterial mats that developed on soil surfaces about 2.6 Ga ago. This discovery, reported in Nature (Nov., 2000), places the development of terrestrial biomass more than 1.4 Ga earlier than previously reported.

• Discovery of 2.3 Ga laterites in Finland. This discovery also suggests that the development of terrestrial biomass and of an oxygenated atmosphere occurred prior to 2.3 Ga ago.

• Discoveries in Archean marine sediments and banded iron formations of geochemical signatures suggesting rapid evolution of the atmosphere and biosphere. The development of an oxygenated atmosphere and oceans, and of the diverse ecosystems, probably occurred more than 3.5 Ga ago.

• Survival of uraninite and siderite under an oxic atmosphere. Our laboratory studies suggest that these minerals, previously considered as stable under an anoxic atmosphere, in fact, dissolve faster under an O_2-poor and CO_2-rich atmosphere compared to under the present atmosphere. Therefore, the presence of detrital grains of siderite and uraninite in some Archean sediments may be supporting, rather than contradicting, evidence for Ohmoto's model postulating an oxic Archean atmosphere.

• Linking the evolution of atmosphere, biosphere, and lithosphere. For the first time in geosciences, we have developed a kinetic approach to quantitatively model the evolution of atmospheric O_2 and CO_2, the redox structure of oceans, the biological activity in the oceans and on land, the mantle degassing, and the volume/area of the continental crust.

Rosemary Capo/Brian Stewart

• Neodymium isotopes provide possible means of dating ancient soils. Application of the Sm-Nd geochronology systems to paleosols from several locations has yielded dates consistent with likely soil formation ages. This is a possible new method for paleosol geochronology, and demonstrates the relative immobility of this isotope system to post-depositional disturbances.

• Neodymium isotope analysis of >2.4 Ga marine carbonate suggests a mantle (hydrothermal) source for rare earth elements (and possibly iron) in the Paleoproterozoic oceans. The flux of these elements from the continents appears to have been minimal during this period.

Michael Arthur

• Variations in sulfur isotopic values of seawater sulfate and sulfate concentrations in carbonate rocks of Neoproterozoic age suggest that the oceanic sulfate concentration was low and that isotopic changes suggest susceptibility to rapid changes in sulfur cycling.

• Numerical modeling supports limited data from the rock record that indicates that the oxygen isotopic composition of seawater was relatively constant and similar to that at present throughout most of the Precambrian.

Mark Patzkowsky

Both paleographic changes (movement of Gondwana toward the South Pole) and eustatic sea level prior to the onset of glaciation may have been important factors determining the timing and severity of the Late Ordovician glaciation. These preliminary results also suggest that, contrary to previous studies, a drop in pCO_2 levels may not be a necessary condition to start the Late Ordovician glaciation.

Roadmap Objectives

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#12
Effects of Climate & Geology on Habitability

#16
Bringing Life with Us Beyond Earth

Project

Biochemistry of Archaea and Bacteria: Biochemistry of Psychrophilic Organisms

Senior Project Investigator(s):
J. Brenchley

Accomplishments

Our objective is to study the diversity and properties of *psychrophilic* (cold-loving) microorganisms that can serve as models for the possible origin, evolution, and survival of psychrophiles from other cold, non-terrestrial sources. Our isolation and examination of bacteria belonging to new species and genera will shed light on the potential diversity of life elsewhere and provide clues to avenues for their cultivation.

Our work includes the isolation of numerous *psychrophiles* and the cloning of genes encoding *glycosyl hydrolase*. One isolate of particular interest was obtained from a hypersaline pond in Antarctica. We have shown that the isolate is a member of the Planococcus genus and that it is both psychrophilic and halotolerant (grows at low temperatures and high salt). Additional isolates from Antarctic samples have physiological traits and 16S rRNA gene sequences that differ from those found for other bacteria and may represent organisms belonging to a new genus.

In addition, we have developed procedures for obtaining and examining possible chemoautotrophic psychrophilic anaerobes from a Greenland ice core. A first step was the development of aseptic techniques for obtaining portions of the ice core as inocula without contaminating the sample. Samples from the center of the ice core were inoculated into a basal autotrophic medium and incubated at temperatures from 0°C to 15°C. We have demonstrated reproducible turbidity of cultures following repeated dilutions into new anaerobic media and incubation at 0°C over the course of several months. Microscopic examination demonstrates that there are several cell shapes and sizes in the mixed population. We are in the process of developing procedures for isolating the specific organisms in these cultures. In addition, we have attempted numerous cell lysis procedures in order to obtain sufficient DNA to allow amplification and cloning of the 16S rRNA genes.

NASA Mission Involvement

This research project is relevant to NASA's missions that search for extraterrestrial life and work with the detection of life in samples returned to Earth.

Future Directions

- We will continue the attempts to grow, isolate and characterize the anaerobes from the ice cores.
- We will continue our characterization of new psychrophilic organisms and their cold-active enzymes as models for life in cold environments elsewhere.
- We will amplify and sequence the 16S rRNA genes and use the data to prepare phylogenetic trees.

These results will determine the evolutionary relationships of the ice-core organisms with the previously isolated organisms, especially those identified as deep branching organisms. The characterization of isolates will provide important information needed in the interpretation data from the geochemical analyses of the ice cores and for examining other environments for psychrophilic organisms.

Biochemistry of Archaea and Bacteria: Enzymes of Ancient Metabolic Pathways

Project

Senior Project Investigator(s):
J. Ferry

Roadmap Objectives

#2
Origin of Life's Cellular Components

Accomplishments

We previously reported that carbonic anhydrases of the independently-evolved beta- and gamma-classes are widespread in procaryotes. We also reported that these enzymes have ancient origins, with the gamma-class dating to the origin of life. The novel nature of these discoveries prompted us to write the first review article on procaryotic carbonic anhydrases with the view to stimulate further research into these enzymes, which are likely to have played a role in the acquisition of carbon dioxide during the evolution of autotrophic pathways in early life forms. We also previously reported on the purification of a beta-class carbonic anhydrase from *Methanobacterium thermoautotrophicum*, which is a thermophilic methane-producing archaeon. This current reporting period we continued our characterization of this enzyme, the first beta-class carbonic anhydrase from any procaryote. Kinetic analyses showed that the enzyme has a zinc-hydroxide mechanism common to the other two independently-evolved classes of carbonic anhydrases. The crystal structure was determined which revealed the overall fold and identified the active site. The structural results establish that the *M. thermoautotrophicum* enzyme represents a sub-class (termed the “cab-type”) of the beta-class, and that the beta-class is more diverse than either the alpha-class (predominantly found in mammals) or the gamma-class. The major difference between ‘plant’ and ‘cab’ type beta-class carbonic anhydrases is in the organization of the hydrophobic pocket thought to bind carbon dioxide. The active site architecture also identifies a route for transfer of a proton from the zinc-bound water molecule required for the zinc-hydroxide mechanism of catalysis. The overall results have provided further insight into the evolution of carbonic anhydrases and provided a foundation for continued studies to understand the catalytic mechanism and the roles they play in the autotrophic metabolism of deeply branching microbes in the phylogenetic tree of life.

Highlights

- The characterization of carbonic anhydrases from the methanoarchaea suggest these enzymes were essential in ancient autotrophic pathways for cell carbon biosynthesis.

- The crystal structure of the first beta-class carbonic anhydrase from a procaryote lays a foundation for investigations into the evolution of carbonic anhydrases and the determination of their physiological roles in ancient microbes.

Future Directions

- Further characterize examples of carbonic anhydrases from the beta- and gamma-class with the view to identify properties that provide an understanding of the evolution of the catalytic mechanism

- Conduct a genetic analysis of *Methanosarcina thermophila* to determine the physiological role of the gamma-class carbonic anhydrase from this methanoarchaeon

- Continue to characterize the carbon monoxide dehydrogenase/acetyl-CoA synthase from *M. thermophila*. Also continue efforts to obtain a crystal structure of the enzyme in collaboration with Cathy Drennan

Biogeochemical and Environmental Influences on the Composition of Late Archean Kerogens and Extracts

Project

Senior Project Investigator(s):
K. Freeman

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

Accomplishments

Our research focuses on unraveling the biogeochemical record of carbon and oxygen cycling in the late Archean by determining relationships between physical environments and biological sources of preserved organic matter. We are testing the hypothesis that depositional environments are linked to microbial ecosystems through the creation and stabilization of oxygen microenvironments or stratified water columns by integrating stratigraphic and organic geochemical studies.

Sample collection of 2.8-2.5 Ga rocks from the Hamersley Basin, Western Australia, is complete. Sample preparation for kerogen carbon isotope analysis and elemental (CHNS) abundance analyses is in progress.

Molecular biomarker analyses and a test for contamination are integral parts of our study and involve detailed investigation of both extractable organic matter and non-extractable kerogen. Molecular analysis of trace components in the extracts of core samples was completed in October & November. Kerogen hydropyrolysis and analysis of products are also in progress.

Field Trips

Western Australia, Hamersley Basin, August 1999.

Samples were collected from the core and a single outcrop for organic geochemical and sedimentary studies. This trip was lead by Hiroshi Ohmoto, Penn State University, and guided by Mark Barley and Brian Krapez of the University of Western Australia.

Western Australia, Hamersley Basin, August 2000.

Samples were collected across the basin in order to establish organic geochemical relationships to depositional environments and/or temporal and geographic variability. More than 200 core samples and 200 outcrop samples were collected. Mark Barley and Brian Krapez assisted with logistics and provided guidance in determining the best field sites to sample and log. Matt Hurtgen, an astrobiology doctoral student at Penn State, assisted in fieldwork.

NASA Mission Involvement

In our research investigation of an uncertain ancient Earth biogeochemistry, we are in a position analogous to what we might face when trying to unravel the biogeochemistries of other planets. Therefore, our research helps NASA missions by setting the foundation for extraterrestrial geochemical studies in the search for life elsewhere in the Solar System. It also will help establish tests for the indigeneity of biomarkers in ancient rocks from Earth and elsewhere in sample return missions.

Future Directions

- Molecular analysis of hydropyrolysis products
- Molecular analysis of specific biomarkers (i.e., branched alkanes)
- Compound specific isotope analysis of biomarkers
- Carbon isotope and elemental analyses of >200 bulk and kerogen samples

Roadmap Objectives

Project

Causes and Consequences of the Diversification and Extinction of Metazoans: Paleontological Approach

Senior Project Investigator(s):
M. Patzkowsky

#5
Linking Planetary & Biological Evolution

#14
Ecosystem Response to Rapid Environmental Change

#15
Earth's Future Habitability

Accomplishments

We have built an extensive database (nearly 1000 fossil collections) of Neoproterozoic and Cambrian macrofossil occurrences in order to evaluate the timing of the Cambrian explosion as well as its environmental and geographic context. The data were entered into the Paleobiology Database housed at the National Center for Ecological Analysis and Synthesis (NCEAS) in Santa Barbara, California (http://flatpebble.nceas.ucsb.edu/public/). A preliminary report on the Paleobiology Database was recently published in the Proceedings of the National Academy of Sciences. We have met most of our main objectives, which were to (1) redesign the database and implement a standardized structure and (2) to begin building the Neoproterozoic and Cambrian data.

We also made progress on investigating the Late Ordovician mass extinction. We are working on an analysis of taxonomic and environmental patterns of extinction and recovery on Laurentia. We have collected an extensive database over the last year. We have also performed simulations of Late Ordovician climate using the GENESIS 2.0 global climate model in order to investigate the role of paleogeography, atmospheric pCO_2 levels, and sea level in causing the Late Ordovician glaciation. We have met all of our main objectives, which were to make preliminary studies into the physical factors underlying the Late Ordovician glaciation and the paleontological patterns of Late Ordovician mass extinction and recovery.

Highlights

Both paleographic changes (movement of Gondwana toward the South Pole) and eustatic sea level prior to the onset of glaciation may have been important factors determining the timing and severity of the Late Ordovician glaciation. These preliminary results also suggest that, contrary to previous studies, a drop in pCO_2 levels may not be a necessary condition to start the Late Ordovician glaciation.

Cross Team Collaborations

The work on the Paleobiology Database has involved collaboration with Charles Marshall (NAI-Harvard Team). This has resulted in one abstract and one publication. Effort on the Cambrian explosion during the next year will come during a sabbatical leave at Harvard (Spring 2002) and work with Charles Marshall.

Future Directions

- Make preliminary analyses of the Paleobiology Database to constrain better the timing of the Cambrian explosion and to determine its geographic and environmental context.

- Complete analyses of Late Ordovician mass extinction in Laurentia.

- Complete manuscript on the Late Ordovician climate simulations. Begin simulations with an ocean circulation model (MOM 2.0; Modular Ocean Model) that uses GENESIS (Global ENvironmental and Ecological Simulation of Interactive Systems) output to investigate the role of ocean heat transport and ocean circulation in the Late Ordovician global environmental changes.

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#12
Effects of Climate & Geology on Habitability

#14
Ecosystem Response to Rapid Environmental Change

Project

Causes and Consequences of the Diversification and Extinction of Metazoans: Neoproterozoic Variations in Carbon and Sulfur Cycling

Senior Project Investigator(s):
M. Arthur

Accomplishments

Topic 1: Understanding geochemical signals of aerobic/anaerobic interfaces: Stable isotope (C, S), orgC-S-Fe, pyrite framboid size, and trace metal interrelationships in sediments. Modern baseline studies for application to Archean and Proterozoic records. We have largely completed our studies of signals in modern environments (Peru margin, Black Sea; see publications) and are beginning to apply the techniques to understanding the ancient record of anoxia.

Topic 2: Secular variations in sulfate sulfur isotopes for the Neoproterozoic. Objectives to detect hypothesized changes in the sulfide oxidizing biota that may have led to a large increase in the sulfide-sulfate sulfur isotope difference and to examine possible changes in oceanic sulfate concentration and isotopic composition that occurred in association with hypothesized "Snowball Earth" episodes. We have made great progress in this project having now produced sulfur isotope records for the Neoproterozoic of Australia, Namibia and the western U.S. We have found at least four major positive excursions in seawater sulfur isotopic composition, two of which are closely associated with "cap carbonates" following glacial epochs.

Topic 3: Phosphogenesis and the Cambrian explosion. We have not produced any publications as yet in this area. One manuscript is in preparation by Suits and Arthur which explores the role of sulfide oxidizing bacterial mats as the nucleation sites for massive deposits of carbonate-fluorapatite, which then lowered the phosphate concentration in seawater and allowed biogenic calcification to ensue. This is a focus for the next 2 years

Highlights

- Variations in sulfur isotopic values of seawater sulfate and sulfate concentrations in carbonate rocks of Neoproterozoic age suggest that the oceanic sulfate concentration was low and that isotopic changes suggest susceptibility to rapid changes in sulfur cycling.

- Numerical modeling supports limited data from the rock record that indicates that the oxygen isotopic composition of seawater was relatively constant and similar to that at present throughout most of the Precambrian.

Field Trips

NW Australia, June/July, 2000.

M. Hurtgen and J. Eigenbrode (both of PSARC) sampled Archean and Early Proterozoic carbonates for stable isotope analysis to examine evidence for early sulfur cycling and the origin of the pronounced negative carbon isotope excursion at about 2.7 Ga as they might bear on the "rise of oxygen."

Death Valley, November, 2000.

M. Arthur and M. Hurtgen (PSARC) sampled Neoproterozoic rocks in the Death Valley, CA region as a supplement to collections provided by A. Prave (St. Andrews Univ., Scotland) for geochemical and stable isotope studies of so-called "Snowball Earth" associated events.

Cross Team Collaborations

During our Death Valley field excursion, we interacted with Anthony Prave (St. Andrews Univ., PSARC Affiliate) Bruce Runnegar (UCLA, NAI), Frank Corsetti (Univ. Southern California) and Martin Kennedy (UC Riverside) in discussions of field relations of Neoproterozoic strata and their bearing on the Snowball Earth Hypothesis.

Future Directions

- complete sulfur isotope studies of secular variation in the Neoproterozoic in order to document global patterns and validate the Namibian record already completed, S. Australia and Great Basin samples

- complete preliminary study of Archean sulfate sulfur isotopes for NW Australian samples (around 2.7 Ga to elucidate possible ranges of oceanic sulfate concentration and implications for pO_2)

- complete work on Precambrian/Cambrian boundary global phosphogenic episode and the possible role of sulfide-oxidizing bacteria at the sed/water interface: implications for "explosion" of fauna with mineralized shells

Roadmap Objectives

#1
Sources of Organics on Earth

Project

Environment of Prebiotic Earth and the Origin of Life: Experimental Approach

Senior Project Investigator(s):
M. Schoonen, J. Kasting, R. Minard

Accomplishments

This year we essentially finished our planned experimental work on the fate of CO on an early Earth. We studied the kinetics of the hydration of CO and the fate of the hydration products. Formate decomposition is first order overall under anoxic, ambient pressure conditions. In neutral to alkaline solutions, pH is not a significant factor in the overall rate of formate decomposition, but as the pH approaches the pKa of formic acid, it becomes apparent that formic acid decays much more rapidly than formate. The decomposition products include formaldehyde. The rate of the decomposition reaction is unaffected by the presence of minerals or dissolved metals. UV-C radiation accelerates the formate decomposition rate ~100-fold and promotes the formation of acetate, particularly in alkaline solution. Perhaps one of the more interesting results of this study in terms of prebiotic chemistry is the preservation of acetate in irradiated formate/$FeCl_2$ solutions. The $Fe(OH)_3$ precipitate is such an effective shield for acetate that the compound remained stable in UV-C, even after the formate concentration had been photolyzed. The 100mM FeII experimental concentrations simulate the levels in the Archean ocean at the time of banded iron formations.

We have also followed up on our discovery that hydrogen peroxide is produced in anoxic solutions through a reaction between water and pyrite. Subsequent experiments with a pyrite analog, NiS_2, show the same effect. A mechanism involving defect sites on the surface of metal disulfides has been formulated. This finding may be of significance to local environments on the early Earth. Its presence may have been a driving force in the evolution to the oxygenic photosynthetic system.

Highlights

- Presence of iron in the Hadean oceans may have been important in shielding organic molecules from UV irradiation.

- Pyrite-water interaction may have led to the formation of hydrogen peroxide on the early Earth. The presence of hydrogen peroxide would have forced organisms to develop protection mechanisms early on. Eventually this may have led to the evolution of a reaction center capable of using hydrogen peroxide as an electron donor. This reaction center may have evolved into a water-splitting reaction center.

Future Directions

- Conduct hydrothermal experiments to determine the fate of CO hydration products at temperatures in excess of 100ºC, simulating the mixing of vent solution with Hadean seawater.

- Conduct an experimental study to determine the rate of ammonia formation in seawater-basalt interaction as well as seawater – iron/iron wustite interaction. Complement these experiments with theoretical thermodynamic equilibrium calculations.

- Determine the stability of amino acids and other simple organics in the presence of pyrite. Evaluate if the hydrogen peroxide formed by pyrite-water interaction could have been detrimental on an early Earth.

Environment of Prebiotic Earth and the Origin of Life: Prebiotic Chemistry of Hydrogen Cyanide

Project

Senior Project Investigator(s):
R. Minard

Roadmap Objectives

#1
Sources of Organics on Earth

#2
Origin of Life's Cellular Components

#9
Life's Precursors & Habitats in the Outer Solar System

Accomplishments

The role of hydrogen cyanide polymer chemistry in the origin of life has provoked much speculation. In spite of extensive efforts by many groups, a satisfactory understanding of this polymer's structure and mechanism of formation still eludes us. Our studies involve the synthesis of HCN polymers under a variety of conditions and the application of modern powerful analytical methods for structure elucidation.

We have shown that analysis of HCN polymer by TMAH thermochemolysis/GC-MS has yielded considerable new structural insight. When this method is applied to the analysis of solids produced in experiments simulating Titan's atmospheric chemistry, a large number of degradation products matched those derived from analysis of HCN polymer by this same method. These results provide evidence that N_2/CH_4 tholins contain HCN polymers, which could therefore be major components of Titan's atmosphere. Along these lines, we observed the ready absorption and polymerization of HCN into nonpolar materials. When HCN gas is sealed with liquid dodecylbenzene, HCN polymer separated from this hydrocarbon liquid within a week. Also, we are examining cyano-self-addition directed by layered double hydroxide mineral surfaces. Initially pink products were formed, which become darker purple with time. Recently, TMAH thermochemolysis GC-MS analysis has shown the presence of glycine, triazine, adenine and xanthine.

We have examined the polymerization reactions of HCN, aminomalononitrile (AMN, HCN trimer) and diaminomaleonitrile (DAMN, HCN tetramer) and glyconitrile under a variety of conditions. AMN and DAMN appear to be more reactive than HCN itself at equal HCN "equivalent" concentration.

Using Laser Desorption-Ionization-Mass-Spectroscopy (LDI-MS) we have found that hydrogen cyanide polymers, (HCN)n, formed from HCN monomer, showed MH+ peaks for n = 8 to 25 with the maximum occurring at n = 15 to 17. HCN polymer formed from diaminomaleonitrile (DAMN, HCN tetramer) showed a similar LDI spectra with no enhancement for peaks at n = 4, 8, 12, 16, etc., implying the breakdown of the tetramer to monomer followed by monomer repolymerization. The LDI-MS spectra of polymer formed from aminomalononitrile (HCN trimer) showed oligomers from m/z 200 to 800, but these did not match an (HCN)n series. In other studies, the water soluble products derived from stirring HCN polymer with water at room temperature for 1 to 3 days consisted primarily of two compounds, urea and a compound with formula $C_6H_8N_6$ as determined by APCI-exact mass MS whose structure we are determining using Nuclear Magnetic Resonance (NMR) techniques.

Natural abundance ^{13}C solids NMR analysis confirms previous observations of a major peak at 166 ppm. We find this same strong absorption in Titan tholin. We have now prepared (HCN)n highly enriched in ^{13}C and ^{15}N. Solids $^{13}C/^{15}N$ NMR analysis of this labeled polymer should provide considerable structural detail of HCN polymer.

Cross Team Collaborations

We have been collaborating with George Cody of the Carnegie Institution Team. We are comparing NMR data on HCN polymer from our lab and Titan aerosol polymer (tholin) and Murchison meteorite NMR data from Cody's lab.

NASA Mission Involvement

HCN is ubiquitous in the Universe, and its chemistry leads to both amino acid and purine/pyrimidine production. One of the most facile reactions of HCN is its self-polymerization to yield a heterogeneous polymer that may be present on moons, in comets, or in interplanetary dust particles that NASA and ESA are planning to probe.

In fact, prior missions (Giotto, 1985-1992) and now Stardust (1999-2006) have given evidence of macromolecular nitrogen-containing solids consistent with HCN polymers or related aqueous altered products. It is essential that we know how to characterize macromolecular solids that NASA and other missions (Cassini-Huygens, 1997-2008) will likely encounter as we explore potential biotic or prebiotic chemistry in the solar system.

Efforts to characterize macromolecular materials fully and effectively are key to adequate preparation for probe or sample return missions. Our research work on the HCN polymer system and its aqueous alteration products contributes to NASA endeavors in the probe or sample return missions.

Future Directions

- In collaboration with Karl Mueller, we have started new solids NMR studies of HCN polymer which show tremendous improvement in spectral quality over previously reported studies. We are optimistic that this will finally reveal HCN polymer structure in detail.

- We plan to more securely characterize the molecular weight distributions of HCN polymers using LDI-MS (laser desorption ionization mass spectrometry) and laser-TOF-MS (time-of-flight mass spectrometry).

- We expect to continue to fully characterize the water soluble products from aqueous and non-aqueous HCN polymerizations. This will include a mass spectral study of the products formed from HCN polymer formed under anhydrous conditions before and after interaction with $H_2{}^{18}O$.

- We wish to examine the report by Eastman that HCN polymers form dendritic structures using optical, atomic force and electron microscopy.

- We plan to examine the interaction of HCN polymer/hydrocarbon composites (mimics of Titan aerosols) with water to see if vesicle formation can be observed. If so, we will investigate the affinity of these materials for sugars and phosphate.

- We plan to test the hypothesis of Matthews that HCN polymers formed under anhydrous conditions could act as condensing agents to form peptide or sugar-phosphate bonds.

- Ultimately, we would like to design experiments involving a variety of polymerization conditions (gas phase, mineral surface, etc.) and post-polymerization modifications that will test the hypothesis that HCN polymers could demonstrate catalytic and self-alignment templating properties consistent with an emergent self-replicating biopolymer.

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#7
Extremes of Life

#11
Origin of Habitable Planets

#12
Effects of Climate & Geology on Habitability

#14
Ecosystem Response to Rapid Environmental Change

Project

Evolution of Atmospheric O_2, Climate, and the Terrestrial Biosphere

Senior Project Investigator(s):
B. Stewart and R. Capo

Accomplishments

We are continuing work on paleosols (ancient soil horizons) in order to determine atmospheric conditions and evidence for possible terrestrial biota at the time of soil formation. We are focusing on the major and trace element characteristics of the ~3 Ga Steep Rock paleosol, Ontario, Canada. We have found striking trace element and Nd isotope patterns through a combination of whole rock and laser ablation inductively coupled plasma mass spectrometry (ICP-MS) analysis, including a possible preserved Sm-Nd isochron recording soil formation age. We have also initiated work on the ~2.4 Ga Hokkalampi paleosol, Finland. Results show possible preservation of Sm-Nd soil formation ages from at least two profiles in the Hokkalampi sequence, with a range of redox conditions from oxidized, Fe-rich areas to reduced, Fe-depleted areas. We are also comparing oxidized and reduced paleosols from the Permo-Pennsylvanian Monongahela Group. This work also shows rare earth element (REE) fractionation during pedogenesis, independent of redox conditions. The paleosol isotopic results provide the groundwork for development of an *in situ* geochronology instrument for planetary surfaces. To understand the similarities and differences between modern and ancient soils, we are working to study the isotopic and geochemical evolution of Hawaiian basalts.

Additional work centers on ancient and modern carbonate as recorders of secular variations in marine chemistry, depositional environment, and paleoclimate indicators. We continue to apply Nd isotopes to Paleoproterozoic marine carbonate sequences from Australia and South Africa. Our work to date suggests that most REE in the carbonate is derived from hydrothermal rather than continental sources, consistent with banded iron formation studies. Our studies of Pleistocene carbonate from Owens Lake, California are designed to understand the water-carbonate speciation of REE, and to relate chemical variations to climate shifts. This provides the baseline for studies of ancient carbonates.

Highlights

• Neodymium isotopes provide possible means of “dating” ancient soils. Application of the Sm-Nd geochronology systems to paleosols from several locations has yielded dates consistent with likely soil formation ages. This is a possible new method for paleosol geochronology, and demonstrates the relative immobility of this isotope system to post-depositional disturbances.

• Neodymium isotope analysis of >2.4 Ga marine carbonate suggests a mantle (hydrothermal) source for rare earth elements (and possibly iron) in the Paleoproterozoic oceans. The flux of these elements from the continents appears to have been minimal during this period.

NASA Mission Involvement

Our work on isotope systematics of the ancient Earth relates to ongoing development of an *in situ* geochronology instrument for martian surface analysis. This instrument is currently being developed by the Principal Investigators in collaboration with JPL personnel. The geochronology instrument development project is funded by NASA's Planetary Instrument Definition and Development Program (PIDDP).

Future Directions

• Continue work on paleosols. This will include (1) determining major controls on redox conditions during soil development, (2) developing and publishing a model for Nd isotope systematics of paleosols, (3) relating isotope systematics to post-depositional disturbances, and (4) comparing ancient soils to modern analogs formed under varying conditions.

• Complete work on Nd isotope composition of Precambrian carbonate with M. Bau. Develop and publish model for REE transport and deposition in carbonate under different depositional conditions

• Initiate work on possible >2.4 Ga red beds from Canada (with H. Ohmoto). This will include *in situ* trace element analyses by laser ablation ICP-MS (with G. L. Macpherson) and careful isotopic analysis of selected phases for geochronological constraints.

• Begin work related to Mission to Early Earth Focus Group. Graduate student S. Stafford will be going on the July, 2001 field trip to Australia, and will initiate isotopic and geochemical studies of outcrop and, ultimately, core samples from the selected localities.

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

Project

Evolution of Atmospheric O_2, Climate, and the Terrestrial Biosphere

Senior Project Investigator(s):
L. Kump

Accomplishments

The Rise of Oxygen: A model for the rise of atmospheric oxygen approximately 2.3 Ga indicates that the interval of intense mantle plume activity in the latest Archean not only stimulated the widespread deposition of banded iron formation, but also created more oxidized source regions for volcanic emissions in the upper mantle, reducing an important Archean oxygen sink, and thus allowing for the establishment of an oxygen-rich atmosphere. A consequence of this change in atmospheric composition was the first Snowball Earth event of the earliest Paleoproterozoic.

Ge/Si Ratio in Precambrian Cherts: Germanium behaves in a geochemically similar pattern to silicon, but has a more important seafloor hydrothermal source. Thus, Ge/Si ratios in the ocean represent a balance between riverine and hydrothermal inputs. In addition, the Ge/Si ratio of the riverine source depends on the intensity of chemical weathering on land. Cherts precipitated from the ocean potentially record the Ge/Si ratio of ancient oceans, thus providing insights into the nature of elemental cycling of Precambrian oceans. We have analyzed a number of Precambrian cherts from the Gunflint (2.0 Ga) and Overnwacht (3.2 Ga) formations. We find that the Ge/Si ratio in these rocks is 2-9x higher than modern cherts, suggesting an overall increased influence of hydrothermal inputs and/or intensity of chemical weathering, both of which can be supported theoretically. Interestingly, iron-formation-associated cherts have the highest ratios, suggesting a hydrothermal source for the iron in banded iron formations (BIFs).

Oceanic Modeling: We have developed a biogeochemical module for the GFDL Modular Ocean Model that is unique in its ability to simulate biogeochemical cycles in anoxic world oceans. We have applied the model to the problem of Late Permian anoxia and mass extinction to test the idea that hypercapnia and H_2S toxicity contributed to the extinction.

Models of Microbial Mat Biogeochemistry: We have generated a 1-dimensional numerical model of the biogeochemistry of microbial mats that considers interactions among cyanobacteria, purple, and colorless sulfur bacteria. Our ultimate goal is to explore the sensitivity of mat structure and function under a variety of boundary conditions appropriate for modern and ancient environments, looking for characteristics that might leave a geological record and indicate whether, for example, the overlying waters were oxygen or hydrogen-sulfide rich.

Highlights

A link between mantle plumes, iron formation, glaciation, and the establishment of an oxygen-rich atmosphere was established. In a paper by Kump, Kasting and Barley (2001), which appeared in the new electronic journal of the American Geophysical Union, Geochemistry Geophysics Geosystems, they propose that plumes of the latest Archean brought oxidized mantle to the surface from near the core-mantle boundary, reducing the oxygen sink associated with the oxidation of reduced volcanic gases, and thus allowing for the buildup of oxygen released by cyanobacteria during photosynthesis. This oxidation event may also have triggered the first global Snowball Earth glaciation of 2.3 Ga. The article was highlighted in Nature with an article by Norman H. Sleep (Nature v. 410, pp. 317-319, 2001).

Future Directions

- Develop a functioning model of microbial mat biogeochemistry, including explicit treatment of aerobic and sulfate-reducing heterotrophic bacteria and isotopic balances for carbon and sulfur

- Extend the microbial model to include aquatic microbial ecosystems. Evaluate the model by collecting field data from a modern, permanently stratified lake that supports an archean consortium of bacteria very similar to those found in microbial mats (Green Lakes, NY State)

- Begin the development of a soil-genesis model. This model will ultimately be used to explore the sensitivity of mineralogy and chemical composition of soils to variations in boundary conditions, particularly to the O_2 and CO_2 content of the atmosphere.

- Continue the collaboration with Jim Kasting, exploring mechanisms for the purported Paleoproterozoic rise in atmospheric O_2. Focus fieldwork and sample acquisition on this time period, perhaps in conjunction with the Mission to Early Earth Focus Group (core acquisition in Western Australia)

Roadmap Objectives

#1
Sources of Organics on Earth

#5
Linking Planetary & Biological Evolution

#11
Origin of Habitable Planets

Project

Evolution of Atmospheric O_2, Climate, and the Terrestrial Biosphere

Senior Project Investigator(s):
J. Kasting

Accomplishments

This report covers work done in association with the rise of atmospheric O_2. We have studied the amount of greenhouse warming expected from CH_4 during the Archean Era. The thesis is that atmospheric CH_4 levels should have been high in a low-O_2 environment. This idea is supported by thermodynamic calculations that predict that methanogenic bacteria should have converted most of the available atmospheric H_2 into CH_4. We also predict that the transition from an O_2-poor to an O_2-rich atmosphere around 2.3 Ga could have triggered the Huronian glaciation by destroying the methane component of the atmospheric greenhouse.

We have described a possible mechanism for delaying the rise of atmospheric O_2. The problem is that O_2 concentrations did not increase substantially until ~2.3 Ga, whereas cyanobacteria were evidently producing O_2 at least 400 million years earlier, and possibly well before that. We suggest that oxidized lithospheric slabs subducted deep into the mantle were brought back to the upper mantle by convection at 2.3 Ga, and that this caused a change in upper mantle redox state that, in turn, caused volcanic gases to become more oxidized.

We also have described quantitative modeling of an organic haze layer produced by photochemical reactions in a CH_4-rich Archean atmosphere. We suggest that the isotopically light (i.e., low $\delta^{13}C$) kerogens seen in Late Archean sediments are, in fact, the remnants of such an organic haze layer. This work is an outgrowth of work that we have been performing under separate funding from NASA's Exobiology program.

Future Directions

We are currently working on an atmospheric model of sulfur species in the Archean atmosphere. The goal is to try to explain mass-independent S isotope fractionation in Archean sediments.

Evolution of Atmospheric O_2, Climate, and the Terrestrial Biosphere

Project

Senior Project Investigator(s):
H. Ohmoto

Accomplishments

The major goal of the research carried out by Ohmoto's group is to understand the connection between the chemical evolution of the atmosphere and oceans and the biological evolution in the oceans and land. Specific questions include: (1) the pO_2 history of the atmosphere and the causes for its evolution; (2) the oxidation history of oceans; (3) the controlling mechanisms for atmospheric pO_2 and pCO_2; (4) the timings of emergence of important organisms in the oceans and on land; and (5) the influence of atmospheric and oceanic chemistry, and of biological activity, on the geochemical cycles of important elements. These questions have been approached from field-related geochemical investigations of a variety of rocks (e.g., paleosols, shales, carbonates, banded iron formations, uraniferous conglomerates) of 3.5 – 1.8 Ga in age, laboratory experiments on redox-sensitive minerals; and computer simulations. Satisfactory progress has been made in all fronts; new and important discoveries have been made to strengthen the theory that postulates the very early (~4.0 Ga) development of an oxygenated atmosphere and a complex biosphere.

Paleosol Project: Geochemical investigation on 2.6 Ga paleosols in the Eastern Transvaal district of South Africa resulted in a discovery of remnants of indigenous cyanobacterial mats that developed on mineral surfaces during soil formation. This discovery has been widely publicized in news media as evidence for the oldest life on land, and as evidence for the early development of an ozone shield and an oxygenated atmosphere. During fieldwork in Finland during the Summer of 2000, we discovered several outcrops of paleolaterites of ~2.3 Ga in age. Formation of laterites, which are soils highly enriched in ferric iron, requires an oxygen-rich atmosphere and organic acids that were generated by soil organisms. Therefore, this discovery also supports the theory that an oxygen-rich atmosphere and the terrestrial biomass developed before 2.3 Ga.

Roadmap Objectives

#1
Sources of Organics on Earth

#2
Origin of Life's Cellular Components

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#10
Natural Migration of Life

#12
Effects of Climate & Geology on Habitability

#14
Ecosystem Response to Rapid Environmental Change

#15
Earth's Future Habitability

Shales and Carbonates Project: A very large number of shale and carbonate samples were collected by us during the past 10 years from the Kaapvaal Craton in South Africa (3.5 – 2.0 Ga), the Pilbara – Hamersley district in Australia (3.5 – 2.4 Ga), southern Ontario district, Canada (2.75 – 1.9 Ga), and from other areas in the world. Detailed investigations on the mineralogical and geochemical characteristics (both inorganic and organic) of these samples have been carried. An important finding from these investigations is that the geochemical cycles of redox-sensitive elements (e.g., C, S, N, Fe, Mn, Mo, U, V, Ce) through the atmosphere – hydrosphere – lithosphere have been basically the same since at least ~3.5 Ga. This finding also supports the model of an early rise of atmospheric oxygen. The results of carbon and sulfur isotope analyses of the above samples suggest the divergence of organisms to produce aerobic organisms (e.g., cyanobacteria, methanotrophs, and eukaryotes) and anaerobic organisms (sulfate-reducing bacteria, methanogenic bacteria) occurred before 3.5 Ga.

Banded Iron Formations (BIFs) Project: Banded iron formations, especially of the Lake Superior-type BIFs, have been considered by many previous researchers as the best evidence for an anoxic atmosphere prior to ~2.0 Ga. Our preliminary investigations on the geochemical characteristics of the Algoma-type BIFs, which are more common than the Superior-type BIFs, suggest that they formed in deep oceans by mixing of Fe^{2+}-bearing hydrothermal solutions and O_2-bearing, deep ocean water. "The oxygenated deep oceans" requires the atmospheric pO_2 to be greater than 20-50 % of the present level. Because the Algoma-type BIFs formed through geologic history, our finding also suggests that the atmospheric pO_2 level has been essentially the same since ~4.0 Ga.

Uraninite Project: The presence of detrital grains of uraninite, pyrite and siderite in some quartz-pebble conglomerates of pre-2.4 Ga age have been used by many researchers as strong evidence for an anoxic atmosphere, because these minerals have been considered to be unstable under an O_2-rich atmosphere but stable under an O_2-poor atmosphere. However, the results of laboratory experiments and theory suggest that the dissolution rates of uraninite and siderite are in fact faster in an O_2-poor and CO_2-rich environment compared to an O_2-rich and CO_2-poor environment; these minerals eventually dissolve out after a long exposure to rainwater under all pO_2 conditions. Therefore, the presence of these minerals in some unusual sedimentary rocks (quartz pebble conglomerates) cannot be used as a measure of atmospheric oxygen level.

Modeling of the Atmospheric Evolution: We have carried out a quantitative evaluation of the various geochemical parameters that influence the production and consumption fluxes of O_2 and CO_2 through the atmosphere, ocean, sediment, crust, and soil reservoirs. Based on this, we have demonstrated that the atmospheric pO_2 level is likely to have remained (and will remain) within ±50 % of the present level as long as the oxygenic photosynthetic organisms are active. The atmospheric pO_2 level has been regulated primarily by the coupling of two negative feedback mechanisms. One is an increasing (or decreasing) flux of organic carbon burial in marine sediments, responding to a decreasing (or increasing) pO_2

in the atmosphere. The other is an increasing (or decreasing) flux of O_2 consumption by soils, responding to an increasing (or decreasing) pO_2. Using the kinetic equations developed in the above study, we have also been carrying out simulation of the long-term (>1 billion years) evolution of atmospheric pO_2 and pCO_2 and of sedimentary chemistry, especially of the contents and isotopic compositions of carbon in marine sediments (carbonates and organic C), under a variety of scenarios for the evolution of the continental crust and the mantle degassing. This is the first attempt by any researcher to quantitatively link the evolution of the atmosphere, oceans, continents and mantle. Our preliminary computations suggest that the pO_2 was likely to have risen essentially to the present level within 50 million years of the first appearance of cyanobacteria about 4 Ga ago and remained about this level while the pCO_2 gradually decreased from ~1000 times the present level. This assumes that the degassing of H_2 and other reducing gases decreased from about 8 times the present flux and the continental volume increased from ~10 % of the present value over a 4 billion year period.

Highlights

- Discovery of evidence for oldest life on land. We have demonstrated that the kerogen preserved in a paleosol in the East Transvaal district is a remnant of cyanobacterial mats that developed on soil surface about 2.6 Ga ago. This discovery, reported in Nature (Nov., 2000), places the development of terrestrial biomass more than 1.4 Ga earlier than previously reported.

- Discovery of 2.3 Ga laterites in Finland. This discovery also suggests that the development of terrestrial biomass and of an oxygenated atmosphere occurred prior to 2.3 Ga.

- Discoveries in Archean marine sediments and banded iron formations of geochemical signatures suggesting rapid evolution of the atmosphere and biosphere. The development of an oxygenated atmosphere and oceans, and of the diverse ecosystems, probably occurred more than 3.5 Ga.

- Survival of uraninite and siderite under an oxic atmosphere. Our laboratory studies suggest that these minerals, previously considered as stable under an anoxic atmosphere, in fact, dissolve faster under an O_2-poor and CO_2-rich atmosphere compared to under the present atmosphere. Therefore, the presence of detrital grains of siderite and uraninite in some Archean sediments may be supporting, rather than contradicting, evidence for Ohmoto's model postulating an oxic Archean atmosphere.

- Linking the evolution of atmosphere, biosphere, and lithosphere. For the first time in geosciences, we have developed a kinetic approach to quantitatively model the evolution of atmospheric O_2 and CO_2, the redox structure of oceans, the biological activity in the oceans and on land, the mantle degassing, and the volume/area of the continental crust.

Field Trips

Northeastern Finland, June 5-23, 2000.

Objective: To investigate ~2.3 Ga paleosols in the Hokkalampi & Hallavaara areas, 2.1 Ga banded iron formations in the Tenetti area, ~1.9 Ga black shales & massive sulfide ores in the Outokumpu area, and 2.2 – 2.0 Ga carbonate sequence in the Jatulian Formation. Participants: Hiroshi Ohmoto and Yumiko Watanabe (PSU), Sherry Stafford (U. of Pittsburgh), Naoki Watanabe (Niigata Univ., Japan), Jukka Marmo and Juha Karhu (Geological Survey of Finland).

Cross Team Collaborations

- Active participation in the Mission to Early Earth Focus Group

- Collaboration with Clark Johnson of JPL/Wisconsin Team on Fe isotope geochemistry of Precambrian paleosols

- Exchange of information and discussions with Bruce Runnegar and James Farquhar of the UCLA Team on sulfur isotope geochemistry of Precambrian rocks

- Exchange of information and discussions with Dick Holland of the Harvard Team on atmospheric evolution

- The Associate Members of PSARC who are not listed in the above summary of research activity, but who have been collaborating with the Ohmoto Research Group, include: Nick Beukes (Rand Afrikaans University), Greg Retallack (University of Oregon), Ken-ichiro Hayashi (Tohoku University), Hu Barnes (PSU), Mark Barley (University of Western Australia), and Anthony Prave (University of St. Andrews).

NASA Mission Involvement

A popular geological theory postulates that the oxygen content of Earth's atmosphere did not reach its present level until about 2 billion or 600 million years ago. However, results of a variety of our investigations suggest that Earth's atmospheric oxygen content rose extremely rapidly, reaching its present level in less than 30 million years and maintaining about this level since then. Emergence of cyanobacteria (first oxygen-producing organism) occurred about four billion years ago.

Results of our study strongly support the theory that a major divergence of organisms, producing a variety of aerobic and anaerobic organisms, occurred about 4 Ga ago. It is probable that Earth's early atmosphere already contained appreciable amounts of ozone (produced from oxygen) and methane (produced by methanogenic bacteria). This research provides a theoretical justification for NASA's program to detect life on other planets by searching for ozone and methane in their atmosphere.

Our research also studies a variety of rock geochemical characteristics, e.g., Ce anomaly, Fe^{3+}/Fe^{2+}, content of redox sensitive elements, plus sulfur and carbon isotopic compositions. From these geochemical qualities, we have identified rocks that formed under different environments, e.g., oxic vs. anoxic or in different ecosystems. These various characteristics can be used to identify the atmospheric conditions and the potential of life (past and present) of other planets, thus relating to NASA missions.

Future Directions

Paleosol Project

• Characterization of organic matter and its host rocks in the 2.4 Ga paleosol at Kalkloof area, S. Africa to constrain the type of organisms that lived on land 2.4 Ga ago

• Characterization of mineralogy and geochemistry of the 2.3 Ga paleolaterite sections in Finland to understand the behaviors of various elements during soil formation 2.3 Ga ago

• Continuation of geochemical investigations on 2.7 Ga paleosol at Mt. Roe, Pilbara, Australia to constrain the timing and mechanisms of Fe loss from this paleosol

Shales and Carbonates Project

• Geochemical investigations of 2.7 Ga black shales in the Abitibi and Wawa districts, Ontario, Canada to understand the relationships between the changes in the redox structure of seawater and those in microbial ecology

• Continuation of the geochemical investigations on shales and carbonates from South Africa (3.5 – 1.8 Ga) and those from the Pilbara – Hamersley district (3.5 – 2.4 Ga)

• Preliminary investigation on the stromatolite and organic matter in the sedimentary sequence at Steep Rock, Ontario, Canada in order to understand the ecological systems ~3.0 Ga ago

Banded Iron Formations (BIFs) Project

• Geochemical investigations of the 2.7 Ga Algoma-type BIFs (Banded Iron Formations) in Ontario, Canada, will be carried out to test our new theory that this type of BIFs formed in deep, oxygenated oceans. The focus will be placed on the rare earth element geochemistry.

Uraninite Project

• Continuation of laboratory experiments on the dissolution kinetics of uraninite in order to establish the rate law as a function of pO_2, pCO_2 and pH

• Preliminary experiments on the dissolution rates of siderite as a function of pO_2

Modeling of the Atmospheric Evolution

• Simulations of the atmospheric evolution, incorporating the sulfur and iron geochemical cycles

- Simulations of the diagenetic processes of marine sediments in order to understand the relationships among the chemical and isotopic compositions of sediments (S and C contents, $\delta^{34}S$ of pyrite and sulfates) and those of seawater

Roadmap Objectives

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#8
Past and Present Life on Mars

Project

GEOPULSE (Gene Expression Observations for Planetary Life Study)

Senior Project Investigator(s):
C. House

Accomplishments

Carbon Isotopic Fractionation in Diverse Microorganisms: We are studying the carbon isotopic fractionation in diverse prokaryotes.

Pyrobaculum as a model organism for the study of geomicrobiology in a hydrothermal vent setting: Progress on this project has been slower than expected. We have run one preliminary metal extraction experiment looking at *Pyrobaculum islandicum* in a low iron medium in the presence of basaltic glass. We have also been developing a PCR-based method of monitoring changes in gene expression with *Pyrobaculum aerophilum*.

Carbon isotopic analysis of individual microfossils and microorganisms: Great progress on the application of secondary mass spectrometry to microbial samples has been made over the last year. In collaboration with the Monterey Bay Aquarium Research Institute, we have used the ion microprobe to identify the archaeal microorganisms responsible for anaerobic methane oxidation. Work on the analysis of microfossils has not proceeded as quickly as planned pending the development of new standards.

Whole genome-based phylogenetic trees of life: We have updated our whole genome-based tree of life to include 27 free and non–free living genomes. We have compared the results from the free-living and the non-free-living genomes. We have also compared our results with other published methods of whole genomic phylogenetic analysis, as well as investigated the usefulness of partial genome sequences.

Highlights

• Methanogens can more significantly fractionate carbon isotopes than many other prokaryotes.

• We have directly identified an archaeal group of microorganisms that consumes methane. The technique developed, Fluorescent *In-Situ* Hybridization-Secondary Ion Mass Spectrometry (FISH-SIMS), will be an important technique for studying microbes that are currently unculturable.

• We also published the first paper demonstrating carbon isotopic analysis on Precambrian microfossils.

• We have found that the whole-genomic method of phylogenetic analysis of free-living microorganisms seems to be robust as more genomes are added.

Cross Team Collaborations

• The paper submitted to Organic Geochemistry represents the results of a collaboration between the Penn State and UCLA Teams.

• Future work on the carbon isotopic analysis of RNA will require collaboration with Woods Hole (MBL Team), as they are developing an alternative method for this important type of analysis.

• The work on Pyrobaculum as a model organism for the study of geomicrobiology in a hydrothermal vent setting requires collaboration between the Penn State Team (microbial geobiology) and the UCLA Team (Pyrobaculum genetics).

• Carbon isotopic analysis of individual microfossils and microorganisms, specifically on anaerobic methane oxidation, required the collaboration of NAI team members at Penn State University, UCLA, and Woods Hole (MBL), as well as collaboration with MBARI (Monterey Bay Aquarium Research Institute). The microfossil work requires collaboration of the Penn State and UCLA Teams.

• The work on whole genome-based phylogenetic trees of life represents collaboration between the Penn State and UCLA Teams. House at Penn State works closely with Sorel Fitz-Gibbon at UCLA to develop our new methods of genomic analysis. Sorel is actively working on the genomic server built at Penn State and managed by Phil Sorber.

NASA Mission Involvement

Focus on the carbon isotopic composition of total biomass, lipids, or RNA is an important way to study fossil or modern microorganisms from the environment without growing them in the laboratory. These developing techniques will be important tools in the search for past or present microbial life on the Earth or Mars.

Our research project may reveal genetic responses of hyperthermophiles to geochemical changes. The evolutionary history of the genes identified can then be investigated using phylogenetics. The project may also help identify biosignatures of microbial life.

In order to investigate microbial biomass (fossil or modern) on other planets, we will need methods that do not depend on our ability to grow the organisms. The analysis of organisms capable of anaerobic methane oxidation (AMO) currently also requires development of such techniques. Therefore, the development of our new technique (FISH—SIMS: fluorescence in situ hybridization—secondary ion mass spectrometry) is fundamental to the goals of astrobiology.

This project will help us understand early events of microbial evolution on Earth. Such information may be very important for understanding what one can expect to have been possible on early Mars.

Future Directions

Carbon Isotopic Fractionation in Diverse Microorganisms

- Calibrate the fractionation between total cell biomass and lipids in methanogens

- Continue to compare carbon isotopic fractionation between methanogens and the archaeoglobales

- Establish a GCMS (Gas Chromatography/Mass Spectrometry) method for the analysis of target RNA

- Compare our GCMS method with LCMS (Liquid Chromatography/Mass Spectrometry) method under development at Woods Hole

Pyrobaculum as a model organism for the study of geomicrobiology in a hydrothermal vent setting

- Test the PCR-based gene expression method on *Pyrobaculum aerophilum* and *E. coli* grown under aerobic and anaerobic conditions at 95°C

- Analyze iron leaching capability of *Pyrobaculum islandicum*, then the tungstun leaching capability of *Pyrobaculum aerophilum*

- Initiate new geomicrobiology experiments with *Pyrobaculum islandicum*, *Pyrobaculum aerophilum*, and *Methanopyrus kandleri*

Carbon isotopic analysis of individual microfossils and microorganisms

- We plan to continue to use the ion microprobe to investigate the isotopic composition of microbial cells. This work may include carbon, nitrogen, and hydrogen isotopic studies.

- We also plan to work on standards that will help the analysis of microfossil material.

Whole genome-based phylogenetic trees of life

- Establish a MySQL database of genes from all published microbial genomes and the results of pair-wise sequence comparisons

- Update whole genomic analysis to include the 26 free-living genomes now available

- Investigate eukaryotic evolution using the whole genome genomic analysis approach

- Compare our protein groups with COGS and TIGRfams

- Develop a method for detection of derived genomes

Microbe-Mineral Interactions

Project

Senior Project Investigator(s):
S. Brantley

Roadmap Objectives

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

Accomplishments

Although the terrestrial average crustal abundances and concentrations in surface waters of Fe, Mn, Zn, Ni, Cu, Co, and Mo are extremely low, each of these metals is used in bacterial enzymes, coenzymes, and cofactors. While it is well known that microbes excrete siderophores to extract Fe from their environment, it is not understood how these siderophores attack minerals to provide the FeIII, nor is it understood how bacteria extract other micronutrients. We are investigating how soil microbes extract these metals from common minerals. In previously reported work, we showed that microbes mobilize Fe from silicate minerals and that this Fe is isotopically light compared to the host mineral. In this past year, we have shown that microbes can mobilize Mo (*Azotobacter vinelandii*), Ni (*Methanobacterium thermoautotrophicum*), and Cu (*Bacillus mycoides*) from silicates. We are attempting to measure the isotopic signatures of these mobilized metals, using multi-collector inductively coupled plasma-mass spectrometry. We have also investigated mobilization of P from the common mineral apatite, and we have documented etch pit types formed as a consequence of biotic and abiotic etching. In this last project, we have also identified a technique to remove microbes from silicate surfaces without chemically or topographically disturbing the surface. Our progress has been very accelerated: we have found that every microbe tested has the ability to mobilize metals, when those metals are needed as micronutrients. Future work with isotopic measurements could identify a suite of biosignatures.

Highlights

- We have shown that Fe dissolved from a soil mineral by siderophore-producing bacteria is shifted up to 0.8 ‰ lighter than Fe in the silicate. A smaller such isotopic shift is observed for Fe released abiotically by two chelates, and the magnitude of the shift increases with affinity of the ligand for Fe, consistent with a kinetic isotope effect during hydrolysis of Fe at the mineral surface. Fe released abiotically without chelates shows no isotopic shift. When recorded in the rock record, Fe isotopes could document Fe transport by organic molecules where such molecules were present in the geologic past.

- We have shown that microbes preferentially mobilize Mo, Ni, Cu, and other metals from silicates.

- We have shown that microbes can be removed from mineral surfaces using SDS, and this treatment does not alter the surface chemically or physically. After treatment with SDS, etching underneath microbes can be imaged.

Cross Team Collaborations

We are collaborating actively with Ariel Anbar of the University of Rochester, who is also interactive with additional NAI teams, namely Harvard University, Jet Propulsion Laboratory, and University of Colorado, Boulder.

Graduate student Robin Guynn in our group received a Director's Travel Award to work with Anbar. Anbar is working with us to measure Fe and Mo isotopes.

Future Directions

- Identify the mechanism of fractionation of Fe isotopes from silicates during microbial mobilization

- Measure the extent of isotopic fractionation of Mo, Ni, and Cu from silicates during microbial mobilization

- Identify new mechanisms of mobilization of P from apatite during microbial growth in P-limited media

- Identify patterns of abiotic and biotic etching on silicate minerals

Timescale for the Evolution of Life on Earth: Molecular Evolution Approach

Project

Senior Project Investigator(s):
B. Hedges

Roadmap Objectives

#2
Origin of Life's Cellular Components

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#7
Extremes of Life

#8
Past and Present Life on Mars

#11
Origin of Habitable Planets

#12
Effects of Climate & Geology on Habitability

#14
Ecosystem Response to Rapid Environmental Change

Accomplishments

Studies continue on the evolution of prokaryotes and eukaryotes, with emphasis on clarifying relationships and divergence times using genomic data, and relating this to Earth history. We completed an initial study of prokaryote genomes where we discovered that many eukaryotic genes of eubacterial origin arise basally among eubacteria rather than being close to alpha-proteobacteria. The simplest explanation is that they arose through an earlier, pre-mitochondrial, symbiotic event. Our time for the origin of cyanobacteria is later than anticipated yet precedes the time period (2.4-2.0 Ga) that some believe represents the major rise in oxygen. We completed a study of fungi and land plants with a large number of nuclear protein-coding genes indicating that land was colonized much earlier (>1 Ga) than the fossil record indicates (~480 Myr ago). We proposed that this early colonization of land by eukaryotes affected the climate of the Proterozoic, possibly leading to the Neoproterozoic snowball Earth events and Cambrian explosion of animals. When compared with previous goals set, we found that productivity (discoveries) has been higher than expected, but that attempting to predict what specific topics and areas of study will yield breakthroughs is difficult. The approach we have taken is to follow the general goals of the project but to be flexible enough to direct energy towards areas showing greatest rewards at any one time.

Highlights

Several discoveries were made with genomic analyses:

- Evidence for a late origin of cyanobacteria, approximately 3 Ga

- Evidence for multiple symbiotic events in the origin of eukaryotes, in contrast to the current single-symbiosis model

- Evidence for a basal position of nematodes with respect to arthropods and vertebrates, in contrast to the widely accepted Ecdysozoa hypothesis joining arthropods with nematodes

- Evidence for an early (Neoproterozoic) colonization of land by fungi and plants, possibly affecting global climate and the evolution of animals

Cross Team Collaborations

Hedges co-chairs the EvoGenomics Focus Group, which held several inter-team meetings during Year 3. These included video conferences, a breakout session during the NAI meeting in April 2000, and a workshop at UCLA (9-11 March 2000). These inter-team meetings concentrated on making plans for collaborative genomic studies of metazoan evolution in the Neoproterozoic.

NASA Mission Involvement

Resolving the timing of different stages in the evolution of life on Earth will directly or indirectly affect most NASA missions related to astrobiology. Detection of life elsewhere relies strongly on biosignatures, with their interpretation based on the evolution of life on Earth.

For NASA missions in our solar system (e.g., Mars, Europa), biosignatures in the substrate (e.g., sediments) will be important. Extrasolar NASA missions (e.g., Terrestrial Planet Finder) will emphasize biosignatures in the atmospheres of planets.

As we more clearly resolve the relationship between the evolution of oxygen-producing microbes (e.g., cyanobacteria) and the development of Earth's atmosphere, biosignatures will have a greater intrinsic value. Also, knowing when (from molecular clock evidence) different groups of prokaryotes arose on Earth and their relationship with the terrestrial environment may help focus searches for evidence of past life on other planets in our solar system.

Future Directions

- Origin of cyanobacteria: complete genomes of several additional species of cyanobacteria have become available, and these will be analyzed to refine the time of origin of this important group of prokaryotes.

- Eukaryotes: additional analyses will be conducted to clarify the number and timing of symbiotic events in the origin of eukaryotes, and diversification of major groups.

Timescale for the Evolution of Life on Earth: Molecular Evolution Approach

Project ▶ Roadmap Objectives

Senior Project Investigator(s):
M. Nei

#4
Genomic Clues to Evolution

Accomplishments

The major goals of our research are to develop reliable statistical methods for constructing phylogenetic trees and estimating divergence times for distantly related organisms, and to analyze protein or DNA sequence data to obtain some idea about the early stage of evolution of life. This year we investigated the efficiencies of various methods of phylogenetic reconstruction from molecular data and showed that the three most widely used methods, i.e., neighbor joining, maximum parsimony, and maximum likelihood methods. All give essentially the same results as long as the reliability of branching patterns is tested by the bootstrap method. There is no need to find the most parsimonious tree or the highest likelihood tree using extensive computer time. We also investigated the accuracies of various methods of estimating divergence times and showed that the most commonly used individual protein (IP) method generally gives overestimates. We then developed two concatenate distance (CD) methods to estimate divergence times. One was to compute average gamma distances for all proteins weighted with sequence length (dAG), and the other was to concatenate all protein sequences and then compute a single gamma distance (dMG). Theoretical and empirical studies have shown that the time estimates obtained by these methods are generally less biased than those obtained by the IP method. Using the new CD methods, we estimated that the times of divergence between eubacteria and eukaryotes, between protists (mostly Plasmodium genes) and other eukaryotes, and between plants, fungi, and animals were 3, 1.7, and 1.3 Ga ago, respectively.

Future Directions

We plan to develop a new method of time estimation for the case where the evolutionary rate varies among different groups of organisms. We also plan to analyze the genomic sequences for some endocellular parasitic genomes (e.g., mitochondria and Buchnera) and their putative ancestral bacteria to understand the mechanism of evolution of eukaryotic organelles.

Education & Public Outreach

Distribution of NAI and PSARC materials at PSTA

The annual Pennsylvania Science Teachers Association Conference was held in Champion, Pennsylvania. NAI and PSARC brochures were distributed to interested educators who visited a booth sponsored by the Pennsylvania Space Grant Consortium and the PA-NASA Education Collaborative.

Penn State's Astrobiology Minor program

Undergraduate students at Penn State can now earn a Minor in Astrobiology. The program includes a total of 18 credits of coursework from the departments of Geosciences, Astronomy & Astrophysics, Biology, Microbiology, and Biochemistry & Molecular Biology. The Astrobiology Minor is designed to educate students in this interdisciplinary field by covering the varied scientific disciplines that contribute to our general understanding of life, the origin of life, the past history of life on Earth, possible futures for life on Earth, and the possible existence of life on other planetary environments.

Presentation at the NJSTA

Penn State graduate student Erin McMullin gave a presentation as part of a one day Astrobiology Symposium at the New Jersey State Teachers Association Conference. The New Jersey Space Grant Consortium and The NJ Technology Council Educational Foundation cosponsored the symposium. The theme for the symposium was "Life in Extreme Environments." The late Gerry Soffen gave an overview of astrobiology and where NASA is heading. John Charles, NASA, gave a presentation on Mars exploration.

Production and distribution of an Astrobiology magazine supplement

“Astrobiology: The Search for Life in the Universe” is a hardcopy, color magazine publication that is written at the high school level. The piece was originally published by Research Penn State magazine in their January 2001 issue and is based on a lecture series that was held at Penn State in January 2000. The lecture series and the magazine supplement feature NAI scientists from Penn State University, The University of Colorado, and Harvard University as well as researchers from the SETI Institute and Rice University. The Pennsylvania Space Grant Consortium is distributing the publication to interested educators, students, researchers and others across the country and internationally.

Space Day 2001 at Penn State

This annual one-day event, sponsored by the Pennsylvania Space Grant Consortium, is designed to showcase the exciting space-related research being carried out at Penn State. We displayed an overview of the research goals of the

Penn State Astrobiology Research Center and distributed informational literature. Several faculty, Postdocs, and graduate students exhibited posters describing their astrobiology research. Two teachers from the 2000 Teacher Workshop demonstrated astrobiology related hands-on activities. We conducted drawings to distribute educator kits.

Teacher Workshop - "Astrobiology: The Origins and Early Evolution of Life"

This five-day residential workshop for in-service teachers is designed to expose middle and high school science teachers to current thinking about the origin and early evolution of life in a way that they can pass on to their students. The course combines lectures by several Penn State faculty, hands-on classroom activities, lab tours, and a field trip to a local bog. Some of the classroom activities were taken from TERC's draft High School curriculum. Evaluations were conducted using daily participant surveys. Following the workshop, this year's teachers were added to our existing listserv discussion group to facilitate and track the application of knowledge from the workshop back in the classrooms. Funding for the workshop is provided by the Penn State Astrobiology Research Center and the Pennsylvania Space Grant Consortium. Housing, meals and some travel support were provided. Teachers are charged tuition for two graduate credits from Penn State. The workshop is approved by the Pennsylvania Department of Education for Act 48 Professional Development credit.

WISE WEEK - Science Projects in Astrobiology

Bob Minard and Chemistry undergraduate student, Nick Hartman, participated in WISE Week 2001, a one-week residential program for young women in the 11th grade. WISE Week is a program of Penn State's WISE Institute (Women In Science and Engineering). The high school students were engaged in a weeklong project designed to expose them to some of the important questions in astrobiology and to some of the approaches used to answer those questions. Students chose to participate in one of two projects: "Chemical Analysis for the Fingerprint of Life - Analysis of samples for biomolecules using Gas Chromatography/Mass Spectrometry". Or "Spectral Analysis for the Fingerprint of Life - Signals from across the cosmos".

Women In Science and Engineering Research (WISER)

Faculty from the Penn State Astrobiology Research Center participate in this two semester research internship program, which is sponsored by the Pennsylvania Space Grant Consortium. The program is designed to retain women students in the science and engineering fields by providing first-year undergraduate students invaluable experience and mentoring at the critical early stages of their undergraduate career. Students begin their research in the Spring semester and continue during either the subsequent Summer or Fall semester. This past year, Sue Brantley, Blair Hedges, and Chuck Fisher involved WISER students in their astrobiology related research.

Publications

Back to Table of Contents

Biochemistry of Archaea and Bacteria: Biochemistry of Psychrophilic Organisms

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Back to Table of Contents

Scripps Research Institute

NASA Astrobiology Institute

La Jolla, California

Principal Investigator

M. Reza Ghadiri

Scripps Research Institute Team Members

Ignacio Alfonso -Rodriguez
Scripps Research Institute

Jamie Bacher
University of Texas, Austin

Steven A. Benner
University of Florida

Ronald K. Castellano
Scripps Research Institute

John Chaput
University of California Riverside

Chong W. Choi
Scripps Research Institute

Stephen L. Craig
Scripps Research Institute

Milan Crnogorac
Scripps Research Institute

Ujjwal Das
University of Florida

Robert Devitt
University of Texas, Austin

Andrew D. Ellington
University of Texas, Austin

Eric Alexander Gaucher
University of Florida

Ronald Geyer
University of Florida

M. Reza Ghadiri
Scripps Research Institute

Kevin Guckian
Scripps Research Institute

Azizul Haque
University of California Riverside

Paul H. Kim
University of California Riverside

Dong-Kye Lee
University of California Riverside

Matthew Levy
University of Texas, Austin

Pillar Lopez-Deber
Scripps Research Institute

John S. McCaskill
German National Research Center

Subbarao Nallagatal
University of California Riverside

Csaba Nemes
Scripps Research Institute

Colin Nuckolls
Scripps Research Institute

Liam Palmer
Scripps Research Institute

Photon Rao
University of Florida

Julius Rebek
Scripps Research Institute

Alonso Ricardo
University of Florida

Timothy Riedel
University of Texas, Austin

Michael Robertson
University of Texas, Austin

Dmitry Rudkevich
Scripps Research Institute

Alan Saghatelian
Scripps Research Institute

Mary Schmitz-Brown
University of Texas, Austin

Surajit Sinha
University of California Riverside

Kathy Soltani
Scripps Research Institute

Letha Sooter
University of Texas, Austin

Christopher Switzer
University of California Riverside

J. Michael Thomson
University of Florida

Paul Wash
Scripps Research Institute

Yohei Yokobayashi
Scripps Research Institute

Executive Summary

Accomplishments

What constitutes life, requirements for the origins of life and evolution, and how living systems may be identified elsewhere in the universe are some of the most fundamental questions in astrobiology. Under the auspices of the Scripps Research Institute, a "virtual institute" has been assembled to explore multidisciplinary experimental approaches to self-replicating molecular species and Darwinian chemistry, the most important hallmarks of life. By comparing and contrasting the results of diverse but complementary sets of experiments, we seek to garner a better understanding of life and its origins. In the past year our team has made significant progress in several areas of research.

Benner's group at the University of Florida has confirmed that hydrogen bonding is central to base pairing in DNA, laying to rest a five-year discussion suggesting that specific base pairing might be had between two DNA strands without the involvement of hydrogen bonds. This result provides an important piece of a model to help us guess how non-terran genetic systems might be structured, and how they might be detected on Mars, Europa, and elsewhere. They have also made significant progress in developing polymerases to support *in vitro* selection with functionalized nucleic acids on an expanded genetic alphabet, directed towards generating, in the laboratory, artificial experimental models for life. In an effort to couple geology to genomics, Benner's group has organized a database of protein sequences containing a complete history of macromolecular life on Earth in order to extract genomic clues that couple events in the molecular record with mass extinctions, paleoecology biosphere transitions, and planetary events. We have also developed simple assays to detect organic chemicals on the surface of Mars.

Ghadiri's group has continued their efforts in the design and characterization of novel self-organized molecular systems that display emergent properties such as replication and parasitism. Furthermore, they provided important experimental evidence for the origin of homochirality in living systems. The origin of homochirality in living systems is often attributed to the generation of enantiomeric differences in a pool of chiral prebiotic molecules, but none of the possible physiochemical processes considered can produce the significant imbalance required if homochiral biopolymers are to result from simple coupling of suitable precursor molecules. This implies a central role either for additional processes that can selectively amplify an initially minute enantiomeric difference in the starting material, or for a nonenzymatic process by which biopolymers undergo chiroselective molecular replication. Given that molecular self-replication and the capacity for selection are necessary conditions for the emergence of life, chiroselective replication of biopolymers seems a particularly attractive process for explaining homochirality in nature. Ghadiri's group reported recently that a 32-residue peptide replicator is capable of efficiently amplifying homochiral products from a racemic mixture of peptide fragments through a chiroselective autocatalytic cycle. The chiroselective amplification process discriminates between structures possessing even single stereochemical mutations within otherwise homochiral sequences. Moreover, the system exhibits a dynamic stereochemical "editing" function; it makes use of heterochiral sequences that arise through uncatalyzed background reactions to catalyze the production of the homochiral product. These results support the idea that self-replicating polypeptides could have played a key role in the origin of homochirality on Earth.

Ellington's group has made significant strides toward filling in several key intermediates in a hypothetical ascent of molecules from simplistic origins to complex catalysts presumed present in the RNA world. They have described *in vitro* selection of a simple deoxyribozyme that may have resembled some of the earliest self-replicators. In addition, Ellington's group evolved a ribonucleoprotein enzyme that may have resembled catalysts found at the boundary between the ancient RNA and modern protein worlds. Finally, their studies, directed at generating an unnatural organism, have gone far in testing the bounds of terrestrial chemistry.

Rebek's group continues to evaluate molecular requirements for the process of self-replication in abiotic systems. They have devised a system that shows a new form of autocatalysis. It involves encapsulated reagents that work on the principles of molecular recognition but without any direct contact between reagents and products. The emergent properties of this system are related to those of self-replicating molecules.

Switzer's group has expanded efforts directed at design and discovery of alternative nucleic acid structures. They demonstrated that the iso-C-iso-G base-pair exhibits intrinsic fidelity during non-enzymatic transcription. This result is in complete contrast with the limited fidelity seen during past experiments involving replication, transcription and translation using natural enzymes and the unnatural iso-C-iso-G base-pair, and implies iso-C-iso-G could have contributed to early biopolymer replication on Earth or elsewhere. In addition they established that the non-standard iso-C-iso-G nucleotide base-pair can support recombination. This is the first time the question of recombination ability has been assessed for an unnatural base-pair, and is an important component of base-pair fitness. Switzer's group has also shown that an acyclic, phosphodiester linked, glycerol nucleotide template (an attractive pre-RNA candidate) can participate in non-enzymatic template-directed copying reactions. Prior to this study, such a result was considered unlikely, and implies this primitive nucleotide candidate could have contributed to early biopolymer replication.

Roadmap Objectives

#2
Origin of Life's Cellular Components

#3
Models for Life

Project

Self-Reproducing Molecular Systems and Darwinian Chemistry -- Rebek's Laboratory

Senior Project Investigator(s):
J. Rebek

Accomplishments

Autocatalysis based on molecular recognition is a phenomenon generally associated with molecular replication. In templated autocatalysis, the product selects specific reactants and acts as a template that makes copies of itself. Because the product is self-complementary, product inhibition is an inevitable consequence of template-mediated replication. Here, we introduce an autocatalytic system in which no direct contact exists between reagents and products. Autocatalysis is observed, but the products do not compete for reaction sites; the two species simply exchange residences. The autocatalytic behavior, therefore, is viewed more correctly as an emergent property of the system as a whole, rather than a property of specific molecules within the system

Highlights

During the current year, we have devised a system that shows a new form of autocatalysis. It involves encapsulated reagents that work on the principles of molecular recognition. The emergent properties of this system are related to those of self-replicating molecules.

Future Directions

For the coming year, we intend to explore how capsules could be used to create metastable systems. We will place incompatible reagents in separate capsules and trigger the reaction between them by adding small molecules that release the reagents from the capsules.

Self-Replicating Molecular Systems and Darwinian Chemistry -- Benner's Laboratory

Project ▶ Roadmap Objectives

Senior Project Investigator(s):
S. Benner

Accomplishments

Benner's group has made major contributions towards the objectives set forward by the NASA Astrobiology Roadmap. They: (a) established the importance of hydrogen bonding in nucleobase pairing, a critical step towards guessing how non-terran genetic systems might be detected, for example, on Mars and Europa, (b) continued the development of polymerases to support *in vitro* selection with functionalized nucleic acids on an expanded genetic alphabet, directed towards generating in the laboratory artificial experimental models for life, (c) developed an assay to detected at femtomole levels the organic molecules that are most likely to be near the surface of Mars and that are likely to be similar to a class of organic molecules on early Earth, (d) worked with Mars mission planners Mars Exploration Program Advisory Group (MEPAG) to design their search for the signatures of life on Mars, (e) served on a panel to review planetary protection protocols, (f) joined with the Ecogenomics Focus Group, extracting genomic clues that couple events in the molecular record with mass extinctions, paleoecology biosphere transitions, and planetary events that couple geology to genomics, (g) organized a database of protein sequences containing a complete history of macromolecular life on Earth (as it can be presently inferred from genomic sequence data, (h) developed sophisticated evolutionary models (non-stationary forms of gamma distributions, for example) that analyze change in function in microorganisms, in particular, in response to changes in microbial ecology and movement in and out of extreme environments.

Highlights

- Confirmed that hydrogen bonding is central to base pairing in DNA, laying to rest a five year discussion proposing that specific base pairing might occur between two DNA strands without the involvement of hydrogen bonds. This provides an important piece of a model to help us predict how non-terran genetic systems might be structured, and how they might be detected on Mars, Europa, and elsewhere.

- Developed simple assays to detect organic chemicals on the surface of Mars.

Cross Team Collaborations

Benner's Florida group has exchanged visitors with the EcoGenomics Focus Group.

#1
Sources of Organics on Earth

#2
Origin of Life's Cellular Components

#3
Models for Life

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#9
Life's Precursors & Habitats in the Outer Solar System

#12
Effects of Climate & Geology on Habitability

#13
Extrasolar Biomarkers

#15
Earth's Future Habitability

#16
Bringing Life with Us Beyond Earth

#17
Planetary Protection

Monica Riley from Woods Hole came to Gainesville to use the “Master Catalog.” This is an evolutionarily organized database of protein sequences containing a complete history of macromolecular life on Earth (as it can be presently inferred from genomic sequence data). Steven Benner went to Woods Hole from Gainesville to research specific issues associated with the Phanaerozoic Project.

Visits from Florida to Pasadena with the JPL Team contributed chemistry to the design of instrumentation suitable for the *in situ* detection of organic molecules on Mars. The Florida group sent samples to the JPL group, and received samples from the JPL group, to facilitate work in each laboratory.

With the Johnson Space Center group, the Florida group has now received samples of SNC meteorites to search for organic compounds within them.

Discussions within our Team between the Ellington and Benner groups may result in collaborations on optimal *in vitro* evolution experiments.

NASA Mission Involvement

Benner (Principal Investigator for this research project) has been involved with NASA missions in the activities listed below.

- Served on a MEPAG (Mars Exploration Program / Payload Analysis Group) panel (Mars Peer Review Group) working to design the search for signatures of life on Mars

- Worked with the Grand Challenge project of the JPL team headed by Frank Grunthaner to design instrumentation for *in situ* analysis of martian organics. The JPL Grand Challenge project focus is identifying the chemical signatures of life on other planets.

- Served on a panel to review planetary protection protocols

Future Directions

- Complete the collaboration with Frank Grunthaner, at JPL, to generate instrumentation to implement assays that we developed to detect the most likely organic molecules on the near-accessible surface of Mars. We hope to include these assays on a future mission.

- Complete a study on the possibility of alternative backbones in alternative genetic systems from non-terran sources

- With the EcoGenomic Focus Group, make major progress on the “Phanerozoic Project”, an ambitious effort to use purely computational methods to couple the molecular history of life on Earth with the history as told by geology, planetary physics, paleontology, paleoclimatology, and paleo-ecology

Self-Reproducing Molecular Systems and Darwinian Chemistry -- Ellington's Laboratory

Project

Senior Project Investigator(s):
A. Ellington

Accomplishments

During the past year Ellington's laboratory has focused on filling in several key intermediates in a hypothetical ascent of molecules from simplistic origins to complex catalysts that may have been present in a putative RNA world. Short oligonucleotides may have been generated by prebiotic mechanisms. Some of these oligonucleotides would have been self-replicating. The first step beyond simple, templated self-replication would likely have been the acquisition of catalysis. We have now selected doppelgangers of these early catalysts, simple deoxyribozyme ligases. To enhance our ability to explore sequence space, we have also automated the process for catalyst selection. Catalytic activity need not have been inherent in nucleic acids alone, but would have been likely augmented by cofactors or effectors. Interestingly, it now appears increasingly unlikely that the opposite strategy would have been pursued, that mere binding of a ligand would not have brought about the acquisition of catalytic activity. This finding has additional, important implications for theories of the origin of the genetic code. To demonstrate the ability of nucleic acid catalysts to synergize with molecules in their environment, we have adapted the selected deoxyribozyme ligase to function as an allosteric enzyme, dependent upon ATP for its activity. We have similarly adapted a selected ribozyme ligase to be dependent upon protein and peptide cofactors. Such nucleoprotein enzymes may have stood at the transition between the ancient RNA and modern protein worlds.

In a separate project, we have selected an *E. coli* strain that completely incorporates an unnatural amino acid throughout its proteome. This 'unColi' demonstrates that canonical, terrestrial chemistries are adaptable, and serves as an avatar for organisms that may be found on other planets.

Highlights

- *In vitro* selection of a simple deoxyribozyme that may have resembled some of the earliest self-replicators.

- Evolved a ribonucleoprotein enzyme that may have resembled catalysts found at the boundary between the ancient RNA and modern protein worlds.

- Generated an unnatural organism that pushes the bounds of terrestrial chemistry.

Roadmap Objectives

#2 Origin of Life's Cellular Components

#3 Models for Life

#4 Genomic Clues to Evolution

#8 Past and Present Life on Mars

#9 Life's Precursors & Habitats in the Outer Solar System

#16 Bringing Life with Us Beyond Earth

Cross Team Collaborations

We have worked closely with Steven Benner at the University of Florida at Gainesville on the incorporation of unnatural nucleotides into nucleic acid enzymes. This work is in progress, but we expect results within the coming year. We have also begun to collaborate with Reza Ghadiri at Scripps Research Institute on simple, ‘chimeric’ self-replicators. Reza has given us self-replicating peptides for which we are attempting to develop nucleic acid receptors (aptamers). The receptors may be better able to catalyze peptide replication, while the peptides may in turn may be able to assist in the assembly of the aptamers from simple oligonucleotides.

Future Directions

In the coming year we will (a) attempt to select self-replicating oligonucleotides from a random sequence pool, (b) discern whether cross-catalysis could have fostered transitions between different chemical species by using both peptide nucleic acids (PNAs) and peptides to act as templates for and/or substrates with short, self-replicating oligonucleotides, (c) split the allosteric deoxyribozyme ligase described above into two or more pieces, and determine if the ligase can self-assemble and self-replicate, (d) probe the interdependence of RNA and protein chemistries in selected nucleoprotein enzymes, and (e) continue to move the ‘unColi’ farther away from terrestrial life forms by the incorporation of additional unnatural amino acids and vitamins into its metabolism. Finally, we are interested in simulating the martian environment and determining whether organisms such as the ‘unColi’ (which has a greatly reduced UV cross section) might have a significant survival advantage in such environments.

Roadmap Objectives

#2
Origin of Life’s Cellular Components

#3
Models for Life

#9
Life’s Precursors & Habitats in the Outer Solar System

Project

Self-Reproducing Molecular Systems and Darwinian Chemistry -- Ghadiri’s Laboratory

Senior Project Investigator(s):
M. Ghadiri

Accomplishments

Ghadiri’s laboratory investigates the importance of molecular Darwinistic processes in the origins of life by exploiting catalytic and self-reproducing (autocatalytic) polypeptide constructs. The goal of our research program is to design, discover, and understand the primary factors responsible for directing self-organization of inanimate molecules into the animate chemistry of living systems. Our approach has been to rationally design and recreate various forms of autocatalytic peptide networks in the laboratory and study how the interplay of molecular information and nonlinear catalysis can lead to self-organization and expression of emergent properties. In the past year we have completed studies on: (1) the design of An Exponential Replicator, fulfilling the structural and kinetic requirements for the onset of Darwinian evolution, (2) established the emergence of a peptide replicator in high salt, highlighting the effects of environmental factors on template-directed catalysis, (3) disclosed the first example of a chiroselective peptide self-replication, addressing the important issue of origin of homochirality in terrestrial proteins, (4) characterized a reciprocal autocatalytic peptide network, illustrating how self-reproduction can emerge out of mutually autocatalytic set of chemical reactions, and (5) designed and characterized a parasitic peptide network, demonstrating the emergence of host-parasite relationship among similar molecular species.

Highlights

The origin of homochirality in living systems is often attributed to the generation of enantiomeric differences in a pool of chiral prebiotic molecules, but none of the possible physiochemical processes considered can produce the significant imbalance required if homochiral biopolymers are to result from simple coupling of suitable precursor molecules. This implies a central role either for additional processes that can selectively amplify an initially minute enantiomeric difference in the starting material, or for a nonenzymatic process by which biopolymers undergo chiroselective molecular replication. Given that molecular self-replication and the capacity for selection are necessary conditions for the emergence of life, chiroselective replication of biopolymers seems a particularly attractive process for explaining homochirality in nature. We reported recently that a 32-residue peptide replicator is capable of efficiently amplifying homochiral products from a racemic mixture of peptide fragments through a chiroselective autocatalytic cycle. The chiroselective amplification process discriminates between structures possessing even single stereochemical mutations within otherwise homochiral sequences. Moreover, the system exhibits a dynamic stereochemical “editing” function; it makes use of heterochiral sequences that arise through uncatalyzed background reactions to catalyze the production of the homochiral product. These results support the idea that self-replicating polypeptides could have played a key role in the origin of homochirality on Earth.

Future Directions

One of the major goals of our research program has been to construct molecular ecosystems and study the behavior of large collections of catalytic and autocatalytic peptides, the process of self-organization, and network formation under variety of reaction conditions and environmental stimuli. We plan to continue our ongoing efforts in this area.

Roadmap Objectives

#1
Sources of Organics on Earth

#2
Origin of Life's Cellular Components

#3
Models for Life

#9
Life's Precursors & Habitats in the Outer Solar System

Project

Self-Reproducing Molecular Systems and Darwinian Chemistry -- Switzer's Laboratory

Senior Project Investigator(s):
C. Switzer

Accomplishments

The primary goal of the Switzer laboratory is to synthesize Alternative Nucleic Acids (ANAs) to attempt the optimization of polymer structure subject to the constraints of prebiotic availability, template-directed reproduction, replication conservative mutation, and fitness. We have identified ANAs by taking small steps in "structure-space" away from RNA (the best model for a molecule bearing features both universal and unique to life) that may avoid some of the problems inherent in fulfillment of the aforementioned constraints. Specifically, the Switzer laboratory is examining ANAs with novel changes to (i) base-pairing domains, (ii) backbone charges and (iii) the sugar. These studies will help to define chemical parameters for molecular evolution. Moreover, our work addresses whether nucleic acid-like molecules are sufficient to enable the origin of life and what limitations exist for life elsewhere in the universe based on a single biopolymer (eg. RNA) rather than multiple biopolymers (DNA, RNA, proteins, carbohydrates). Accomplishments by our group for the past year include: (i) the discovery of surprising fidelity during non-enzymatic transcription of an unnatural base-pair, (ii) the discovery that DNA recombination is possible with artificial Watson-Crick base-pairs, (iii) an assessment of how tethered cations affect DNA topology, and (iv) the discovery that a DNA analogue bearing a simple acyclic sugar replacement of glycerol in place of ribose can support template-directed synthetic reactions.

Highlights

- Demonstrated that the iso-C-iso-G base-pair exhibits intrinsic fidelity during non-enzymatic transcription. This result is in complete contrast with the limited fidelity seen during past experiments involving replication, transcription and translation using natural enzymes and the unnatural iso-C-iso-G base-pair, and implies iso-C-iso-G could have contributed to early biopolymer replication on Earth or elsewhere.
- Established that the non-standard iso-C-iso-G nucleotide base-pair can support recombination. This is the first time the question of recombination ability has been assessed for an unnatural base-pair, and is an important component of base-pair fitness.
- Established that an acyclic, phosphodiester linked, glycerol nucleotide template (an attractive pre-RNA candidate) can participate in non-enzymatic template-directed copying reactions. Prior to this study, such a result was considered unlikely, and implies this primitive nucleotide candidate could have contributed to early biopolymer replication.

• Showed that electrostatic perturbation of the DNA helix by asymmetrically tethered ammonium ions is strongly influenced by tether flexibility. This result contributes to knowledge of the electrostatic environment about DNA in general and how proteins bend DNA in particular.

Future Directions

• Expand the number of monomers and sequences capable of efficient non-enzymatic, template-directed replication by using an informational polymer related to nucleic acids incorporating new, but prebiotically plausible, features. This will be done using the general approaches (ie., altered base domains, altered formal charges) that were proposed originally to arrive at alternative nucleic acids with improved properties.

• Examine the ability of prebiotically plausible, and previously unknown, alternative nucleic acid derivatives to satisfy the constraints of replication conservative mutation. This will be done using the general approaches (ie., altered base domains, altered formal charges) that were proposed originally to arrive at alternative nucleic acids with improved properties.

• Develop assays to explore the fitness of the above and previously reported alternative nucleic acids from our laboratory.

Using Functional Genomics to Infer the Biology and Chemistry of the Last Common Ancestor

Project

NAI Postdoctoral Fellow: E. Gaucher
Advisor: S. Benner

Roadmap Objectives

#3
Models for Life

#4
Genomic Clues to Evolution

Accomplishments

Our goal is to test the thermostability of an ancestral reconstructed protein from bacteria using a combination of laboratory and computational approaches. To this end, phylogenetic analyses have been performed in order to determine extant bacterial relationships (phylogenetic tree) using sophisticated maximum-likelihood models. The ancestral sequence at the base of the bacterial lineage was subsequently inferred. The next step of the project will entail the reconstruction of the ancestral sequences in the laboratory using standard molecular biology techniques. This, in turn, will allow us to test the thermostability of the ancestral sequences. We are making appropriate progress towards the previously set goals as established in my initial application.

Highlights

- <u>Concepts of Functional Divergence</u>

Our ability to detect functional divergence among genomic sequences can be greatly influenced by our perception of function/behavior. The goal is to inform the genomics community of possible biases that limit the exploration of genomic sequences as they relate to biological systems in general.

- <u>Approaches for Detecting Functional Divergence</u>

We are interested in elucidating the various patterns of protein divergence. Specifically, we are analyzing shifts in evolutionary rates as an approach to detect altered biological function.

- <u>Validation of Functional Divergence Predictions</u>

Predictions based on our previous analyses have recently been confirmed, highlighting the power of combining computational and experimental biology.

Future Directions

- Test elongation factor protein thermostability from the last common ancestor of the bacterial lineage. Reconstruct ancestral sequences in the laboratory, test thermostability, and draw conclusions regarding the environment in which bacteria lived before their adaptive radiation.

- Use NED (Neutral Evolutionary Distances) tools to date evolutionary events. Attempt to correlate genomic duplication events to paleontological and climatological records.

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Back to Table of Contents

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Back to Table of Contents

University of California, Los Angeles

NASA Astrobiology Institute

University of California, Los Angeles Team Members

Eric E. Becklin
University of California Los Angeles

Andrew D. Czaja
University of California Los Angeles

James Farquhar
University of Maryland

James G. Ferry
Pennsylvania State University

Jason Finley
University of California Los Angeles

Sorel T. Fitz-Gibbon
University of California Los Angeles

James R. Garey
University of South Florida

Ruth D. Gates
University of California Los Angeles

James G. Gehling
University of South Australia

Andrea M. Ghez
University of California Los Angeles

Michael Ghil
University of California Los Angeles

Kathleen Grey
Geological Survey of Western Australia

T. Mark Harrison
University of California Los Angeles

S. Blair Hedges
Pennsylvania State University

Tracey Herrera
University of California Los Angeles

Andrew N. Hock
University of California Los Angeles

Seth D. Hornstein
University of California Los Angeles

Christopher H. House
Pennsylvania State University

David K. Jacobs
University of California Los Angeles

Ravi Jain
University of California Los Angeles

Patricia J. Johnson
University of California Los Angeles

Per M. Jogi
University of California Los Angeles

Michael A. Jura
University of California Los Angeles

D. Keith Kirts
University of California Los Angeles

Dana N. Kovaric
University of California Los Angeles

Frank T. Kyte
University of California Los Angeles

David LaFreniere
University of California Los Angeles

James A. Lake
University of California Los Angeles

Barbara A. Laval
University of California Los Angeles

Crispin T.S. Little
University of California Los Angeles

Donald R. Lowe
Stanford University

James R. Lyons
University of California Los Angeles

Craig E. Manning
University of California Los Angeles

Caer-Eve McCabe
University of California Los Angeles

Christopher McCarthy
Carnegie Institution of Washington

Kevin D. McKeegan
University of California Los Angeles

Ian S. McLean
University of California Los Angeles

Laurel Methot
University of California Los Angeles

Jeffrey H. Miller
University of California Los Angeles

Michael A. Mischna
University of California Los Angeles

Stephen J. Mojzsis
University of Colorado Boulder

Jonathan E. Moore
University of California Los Angeles

William B. Moore
University of California Los Angeles

James M. Nakatsuka
University of California Los Angeles

William I. Newman
University of California Los Angeles

Keith R. Olwin
University of California Los Angeles

David A. Paige
University of California Los Angeles

Maria C. Rivera
University of California Los Angeles

Andrew J. Roger
Dalhousie University

Bruce N. Runnegar
University of California Los Angeles

Matthew R. Saltzman
Ohio State University

Gerald Schubert
University of California Los Angeles

Jeffrey D. Silberman
University of California Los Angeles

Leigh Ann Smith
University of California Los Angeles

Inseok Song
University of California Los Angeles

Karl O. Stetter
University of California Los Angeles

Ferenc Varadi
University of California Los Angeles

Michael J. Vendrasco
University of California Los Angeles

M. Indira Venkatesan
University of California Los Angeles

Malcolm R. Walter
Macquarie University, Australia

John T. Wasson
University of California Los Angeles

Richard A. Webb
NASA Ames Research Center

Alycia J. Weinberger
Carnegie Institution of Washington

Benjamin Zuckerman
University of California Los Angeles

Executive Summary

Accomplishments

Research at UCLA continues to be focused on six main themes: (1) extrasolar planetary systems; (2) geobiology and geochemistry of early Earth and Mars; (3) evolution of Earth's early life; (4) genomic evolution and the tree of life; (5) celestial influences on planetary environments; and (6) exploration for life in the solar system. Over the past year, activities of NAI Focus Groups have allowed us to explore several of these themes in conjunction with partners across the astrobiology community. Mission-oriented achievements include the selection of the Artemis project (David Paige, PI) as one of ten Scout mission concepts. These accomplishments are developed more fully below.

Astronomy

Song, Zuckerman and their colleagues are targeting the youngest and nearest stars in their search for dusty disks and directly observable planets. Telescopic observations of about 300 candidate objects, mostly at Siding Spring Observatory, Australia, have revealed a sizeable group of 10 million year-old southern stars (the ß Pictoris moving group) that are near enough to enable advanced coronographic and adaptive optic methods to detect and image cooling giant planets when comparable facilities become available in the southern hemisphere. This result dramatically exceeds expectations of only a year ago. In a parallel study, Jura and Chen recently reported the probable detection of a massive population of asteroids around a nearby star. Looking forward to the first science flights of the SOFIA airborne infrared telescope in 2004, astronomers are beginning to explore astrobiological applications such as the detection and characterization of organics in the interstellar medium and nearby accretion disks.

Geobiology and Geochemistry

Excitement in geochemistry comes from discoveries, made by Farquhar and his collaborators at UC San Diego, of a record of atmospheric mass-independent isotope effects recorded in modern and ancient sedimentary rocks. Lyons has developed a photochemical model for oxygen isotopes that implicates ozone, rather than peroxide, as the influential molecule and also predicts large $\delta^{17}O$ anomalies in upper atmosphere nitrates. The recent discovery of nitrate particles in the Earth's polar stratosphere suggests that this study will have unexpected and important spinoff applications for present-day atmospheric chemistry. In the more complex and less well understood sulfur system, important progress has been made in developing new techniques and standards for multi-collector ion microprobe analysis of ^{32}S, ^{33}S, ^{34}S, and tentatively ^{36}S, in both sulfides and sulfates (Mojzsis, McKeegan, Lyons, Runnegar, and collaborators). Work on 3.5 Ga old samples from Western Australia provides strong support for the Farquhar hypothesis that sulfur cycling during the Archean was greatly influenced by shortwave ultraviolet radiation in an anoxic atmosphere. This research imposes severe constraints on ideas about bacterial sulfur metabolism during early Earth history. Similarly, the discovery of 4.3-4.4 Ga-old detrital zircons from Western Australia have already led to the inference of an ocean-scale hydrosphere interacting with the crust by that time (Mojzsis, Harrison). The ambitious plan now is to obtain sufficient quantities of these Hadean zircons (10,000?) to investigate significant environmental questions such as the age of the atmosphere (using ^{136}Xe derived from extinct ^{244}Pu) and rates of crustal recycling on the early Earth (Harrison).

Following the pioneering study of ion microprobe measurements of the carbon isotope composition of individual Precambrian microfossils reported last year (House and colleagues), this technique has been used to measure the carbon isotopic compositions of cells targeted by fluorescent in-situ hybridization (FISH) within novel microbial consortia (House, McKeegan, and collaborators). This work led to unexpected collaborations among scientists of the Monterey Bay Aquarium Research Institute, the Woods Hole Oceanographic Institution, and the Penn State and UCLA teams. Importantly, it also led to the identification of archeal microorganisms responsible for anaerobic methane oxidation.

Metamorphic terranes on Akilia Island, Greenland, are thought to contain both the oldest sediments and the oldest evidence for biological activity (Mojzsis, Harrison, McKeegan, and collaborators). Since these interpretations depend on strongly contested geologic observations, Manning, Mojszis, Harrison, and colleagues mapped the Akilia outcrop at a scale of 1:250 and confirmed the great antiquity of crucial sample sites. Ion probe depth-profiling of a selected zircon crystal then provided unequivocal evidence for a >3.83 Ga age for both the sedimentary rocks and the organic matter they contain.

Paleobiology

Field work in the Bangemall Basin, Western Australia, allowed Gehling, Runnegar, and collaborators to observe and sample numerous horizons of Mesoproterozoic megafossils that are preserved in exactly the same way as the soft-bodied Ediacarans of the terminal Proterozoic. Preliminary work suggests the possibility that these megafossils, previously thought to be eukaryotic algae, may be novel microbial consortia, but the large collection will not be studied in detail until later in 2001. Research continues on understanding early Archean conical stromatolites from Western Australia using the differential calculus of interface physics (Jögi, Runnegar). As realistic simulations in 2+1 dimensions require sizeable grids ($> 2^{10}$ nodes), much time has been devoted to developing codes for massively parallel supercomputers. Some of this work will be done in collaboration with the Spanish Centro de Astrobiología (Perez-Mercader).

Molecular techniques are increasingly being used to understand early animal evolution. In addition to the massive metazoan genome sequencing effort being proposed by the Evolutionary Genomics Focus Group, Jacobs, Gates, and colleagues used engrailed gene expression in mollusks to suggest that the skeletons of all bilateral animals have a common origin, an idea that bears on the nature of the Cambrian Explosion. Additional work on genes involved in intercellular signaling, as well as neural and sensory development in cnidarian flatworms, is throwing light on the origins of sense organs and, hence, on nervous systems.

Genomics and Evolution

Fitz-Gibbon and House are extending their whole-genome analyses based on the presence or absence of protein-coding genes as phylogenetic markers. This computationally formidable task has now been extended to include 26 published genomes. They are investigating the effects of lateral gene transfer and the statistical effects of adding partial gene sequences. The latter work has an important bearing on the utility of expressed sequence tags (ESTs) for phylogenetic analysis and it complements collaborative work within the Evolutionary Genomics Focus Group on horizontal gene transfer (Lake, Rivera, Riley). An intermediate target is understanding, at the genomic level, the evolution of the eukaryotic cell (Lake). Complementary research on deeply-branching, free-living, and pathogenic protists is aimed at understanding the evolution of the eukarotic cell in the context of anaerobic and aerobic energy production by means of organelles (Johnson, Roger, Silberman). A large number of new rRNA gene sequences has been obtained and the organisms are now being searched for genes targeted to organelles, including those organelles that may have been lost.

Planetary Science

Kyte recently participated in an Antarctic expedition aboard the Polarstern, a research ship operated by the German Alfred Wegener Institute for Polar and Marine Research. Two of the seven weeks of the cruise were devoted to geophysical and geological exploration of an area in the Bellingshausen Sea which is the only known site of an asteroid impact into a deep-ocean basin. At least 15 of the cores recovered contain ejecta from this Tertiary-age impact, and they will be used to understand the processes and environmental effects of an oceanic event of this scale.

Additional insight into large impacts within the habitable zone may come from long numerical integrations of solar system dynamics (Varadi, Ghil, Runnegar). Integrations involving different conditions point to a chaotic perturbation of the inner solar system at or near the Cretaceous-Tertiary (K-T) boundary, 65 million years ago. Varadi and colleagues are exploring the possibility, using asteroids as test particles, that this chaotic transition may have nudged the K-T impactor into an Earth-crossing orbit. In a different study, W. Moore has developed a mission-testable, end-member model for heat production within Europa that has important implications for the possibility of maintaining life in Europa's subsurface ocean.

Solar System Exploration

NASA has selected the Artemis multi-Scout mission as one of ten potential payloads for launch in 2007. The study team is led by UCLA (Paige, PI), and includes as partners TRW and JPL. The plan is to send an orbiter carrying three or more small landers which will be targeted to a wide range of latitudes, including the polar regions. The landers will incorporate miniaturized versions of Mars Polar Lander instruments, with the additional capability of detecting organic compounds, plus a nanorover.

NAI Focus Groups

UCLA team members are active participants in the majority of NAI Focus Groups (FG). Paige is an active member of the Mars FG; Jacobs, Lake (co-Chair), and Runnegar are deeply involved in the EvoGenomics FG; W. Moore is working closely with the Europa FG; Kyte and Ward are candidate co-Chairs for the proposed Impacts FG; and team members have substantial interests in the Astromaterials, Mission to Early Earth, and EcoGenomics Focus Groups.

Astrobiology at UCLA: An Integrated Multidisciplinary Approach to Research and Education

Project

Senior Project Investigator(s):
B. Runnegar, M. Harrison, J. Lake,
B. Zuckerman

Accomplishments

Research at UCLA continues to be focused on six main themes: (1) extrasolar planetary systems; (2) geobiology and geochemistry of early Earth and Mars; (3) evolution of Earth's early life; (4) genomic evolution and the tree of life; (5) celestial influences on planetary environments; and (6) exploration for life in the solar system. Over the past year, activities of NAI Focus Groups have allowed us to explore many of these themes in conjunction with partners across the astrobiology community. Progress in several of these areas has exceeded expectations because the problems were well chosen (Hadean environment, Archean sulfur cycles, geomicrobiology, etc.) and they were attacked with state-of-the-art instrumentation (ion microprobes, adaptive optics, fast computers, etc.). In other cases, longer-term goals are being achieved progressively. This is particularly true for genomic investigations and those that are aimed at solar system exploration. Mission-oriented achievements include the selection of the Artemis project (David Paige, PI) as one of ten Scout mission concepts for possible launch to Mars in 2007.

Roadmap Objectives

#1
Sources of Organics on Earth

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#11
Origin of Habitable Planets

#12
Effects of Climate & Geology on Habitability

#13
Extrasolar Biomarkers

Highlights

- Discovery of the ß Pictoris moving group, a new association of the youngest and nearest star systems to Earth and likely to house imageable planets (Song and Zuckerman, submitted)

- Probable detection of massive population of asteroids around a nearby star (Jura and Chen, in preparation)

- Prediction that atmospheric nitrates will exhibit large mass-independent fractionations of oxygen isotopes (recently confirmed by observation of Death Valley nitrates) (Lyons, accepted)

- Confirmation of effects of atmospheric photochemistry on sulfur cycling during the Archean (Runnegar et al., in preparation)

- Detection of ocean-continental crust interaction in 4.3 billion year-old zircon crystals from Western Australia (Mojzsis et al., 2001)

- Carbon isotopic compositions of bacterial consortia members measured by a collaboration between microbiologists and geochemists (House et al., accepted)

- Discovery that animal (metazoan) skeletons may be underwritten by a common molecular mechanism (Jacobs et al., 2001)

- Geological mapping of Akilia Island, Greenland, confirms >3.83 Ga age for oldest sedimentary rocks and their contained organic matter (Manning et al., in preparation)

- A zoo of novel anaerobic protists grafted onto the tree of life (Silberman et al., submitted)

- Chaos in the inner solar system may have been the ultimate cause of the Cretaceous-Tertiary impact (Varadi et al., in preparation)

- Artemis multi-Scout mission proposal selected as one of ten potential payloads for launch to Mars in 2007 (Paige, PI)

Field Trips

Western Greenland, Akilia Island, July 2000.

Objective: to map and sample Archean metasedimentary rocks that contain isotopic evidence for an early origin for life on Earth. Participants from University of California, Los Angeles: Craig Manning, Steven Mojzsis, Natalie Caciagli, Mark Harrison

Western Australia, Bangemall Basin and Pilbara region, September 2001.
Objective: to explore and collect Mesoproterozoic megafossils from the Bangemall Basin and sulfides and sulfates from the early Archean Warrawoona Group (explained more fully in Executive Summary). Participants: Mikhail A. Fedonkin, Russian Academy of Science, Moscow; James G. Gehling, South Australian Museum; David McB. Martin, Geological Survey of Western Australia; Martin Van Kranendonk, Geological Survey of Western Australia; Ian R. Williams, Geological Survey of Western Australia; Ellis L. Yochelson, U.S. Geological Survey, Washington, D.C.; Four field assistants.

Australia - New South Wales, Australian National University Siding Spring Observatory, December 2000 and January 2001.
Used the 2.3 meter telescope to survey stars selected on the basis of criteria for extreme youth from existing star catalogs. Participants: Benjamin Zuckerman and Inseok Song, University of California, Los Angeles.

Cross Team Collaborations

UCLA team members are active participants in the majority of NAI Focus Groups (FG). Paige is an active member of the Mars FG; Jacobs, Lake (co-Chair), and Runnegar are deeply involved in the Evolutionary Genomics FG; W. Moore is working closely with the Europa FG; Kyte and Ward are candidate co-Chairs for the proposed Impacts FG; and team members have substantial interests in the Astromaterials, Mission to Early Earth, and EcoGenomics Focus Groups.

Other cross team collaborations are mentioned elsewhere in this report.

NASA Mission Involvement

Among the various astrobiology projects at UCLA, there are three research activities with direct contributions to NASA missions.

- E. Becklin is Director of the SOFIA airborne infrared observatory, due to begin science flights in 2004.

- W. Moore has relied heavily on the magnetometer results from the spacecraft Galileo to interpret and model the lithospheric structures of the four Galilean moons of Jupiter. (Margaret Kivelson, Principal Investigator of the Galileo magnetometer, is a colleague at UCLA). Moore is now developing concepts for instruments that can be flown to Europa to test his end-member models for Europa’s internal heat balance.

- D. Paige is Principal Investigator of the Artemis multi-Scout mission proposal, which has been selected as one of ten potential payloads for launch to Mars in 2007.

Future Directions

- Continue to expand the catalog of very young (<30 Ma), nearby, T Tauri stars such as the TW Hydrae, Tucana, and Horologium A associations and the new and exciting ß Pictoris moving group. Most are best seen from the southern hemisphere so continuing collaboration with astronomers at ANU's Siding Spring Observatory, NSW, Australia is planned for 2001-2002 (Song, Zuckerman).

- Participate in the development of adaptive optics, coronographic, and near infrared capabilities at the Keck Observatory, Hawaii, and with the airborne SOFIA infrared telescope. The goals here are the imaging of young stars and dusty accretion disks as well as the detection and characterization of organic molecules (Becklin, Ghez, Jura, McLean, Song, Zuckerman).

- Test the hypothesis that atmospheric nitrates have large $\delta^{17}O$ anomalies that can be preserved in geological materials. Particulates collected at the Santa Monica, California pier and late Cenozoic nitrates from the Atacama desert, Peru will be analyzed for ^{16}O, ^{17}O, and ^{18}O using multi-collector ion microprobe methods (Lyons, McKeegan, Runnegar). This work represents an unexpected applied spinoff that requires further evaluation.

- Extend the already successful study of the sulfur cycles in early Archean environments by sampling (July 2001) sulfides and sulfates from exploration coreholes drilled by Sipa Resources that intersected pristine volcanic massive sulfide (VMS) deposits formed deep within the Archean ocean. These hot vent deposits will complement the shallow marine environments already sampled within this time window (Farquhar, Harrison, Lyons, McKeegan, Runnegar).

- Explore the interaction of the methane-producing thermophile Methanopyrus with oxidative (magnetite) and reductive (olivine) mineral substrates under gases of different compositions. Here the objectives are to develop new techniques for examining cooperativity among inorganic and biological redox reactions (House, Manning).

- Work towards industrial-scale sampling of the Jack Hills and Narryer conglomerates, Western Australia, in order to obtain sufficient (~10,000) of the fraction of detrital zircons (~4%) that will yield U-Pb ages > 4.2 Ga. Products of early short-lived isotopes (e.g., ^{136}Xe derived from ^{244}Pu) that provide information about the environment on the Hadean Earth should then be measurable (Harrison, McKeegan, Mojzsis).

- Cast, measure, photograph, and describe the ton or so of samples containing Mesoproterozoic megafossils collected from the Bangemall Basin, Western Australia in 2000. This work will explore the role of microbial binding of sediment surfaces in the preservation of fossils here and on Mars (Gehling, Grey, Runnegar).

- Continue attempts to simulate the topographic features of conical stromatolites from the early Archean of Western Australia using the differential calculus of interface physics on massively parallel machines. These Archean structures are features that would be readily visible, in the right geological seeting, from a rover on Mars. It is important that we understand their origin here on Earth (Jögi, Perez-Mercader, Runnegar).

- Find and sequence genes involved in the production of gravity receptors and eyes in lower metazoans (sponges, cnidarians, and flatworms) in order to explore the origin of sensation and innervation within animals (Gates, Jacobs).

- Continue to develop and refine whole-genome comparisons of protein-coding genes in order to test the small subunit ribosomal RNA tree of life and to evaluate historical effects and noise levels resulting from horizontal gene transfers (Fitz-Gibbon, House).

- Test models for modularity in gene assembly and the role of horizontal gene transfer in molding the history of life by computing comparisons within and between microbial genomes (Benner, Riley, Rivera, Lake).

- If funds become available, obtain several thousand partial gene sequences (ESTs) from 30-40 kinds of animals representing all of the phyla for which no genome sequence is available and many of the phyla (arthropods, brachiopods, echinoderms, molluscs) that have excellent fossil records. The goals are to understand metazoan relationships and to explore the timing of the metazoan radiation in the context of snowball Earth and the Cambrian explosion (an EvoGenomics FG proposal; Benner, Boore, Garey, Hedges, Jacobs, Lake, Marshall, Rivera, Runnegar).

- Use the offshore sinkhole Jewfish Spring, Florida, as an accessible sulfide-rich habitat for exploring bacterial and metazoan symbioses/adaptations as an analog for deep sea vent communities (pilot project funded by NAI Director's Discretionary Fund; Garey).

- In conjunction with other scientists, work up cores containing ejecta from the Eltanin oceanic asteroid impact obtained during the Austral summer of 2001 (Kyte).

- Continue running year-long numerical integrations of solar system dynamics to explore chaotic transitions such as the one that coincided, approximately, with the K-T boundary and may have been responsible for placing the end-Cretaceous impactor into an Earth-crossing orbit. This work will focus on improving the accuracy of the simulations, testing them via the stratigraphic record, and using asteroids as test particles to study transitions in the dynamics.

- Field work in the early Archean Western Australia aimed at re-examining evidence for the antiquity of life on Earth given new interpretations of long-supported criteria including carbon, oxygen, and sulfur isotope ratios, microfossils, and stromatolites (Knauth, Lowe, Lyons, Runnegar).

- Development of the Artemis multi-Scout mission concept in conjunction with partners at TRW and JPL (Paige).

Education & Public Outreach

Astrobiology at UCLA: An Integrated Multidisciplinary Approach to Research and Education

Astrobiology Superstars is a bi-monthly free public lecture series at the Midnight Special Bookstore on the Santa Monica Promenade, three blocks from the Pacific ocean. The series features researchers and professors from the UCLA Center for Astrobiology speaking on their area of expertise.

Six presentations and lectures were given at various science conferences and universities nationally and internationally. Presenters discussed astrobiology; what it is and where it is going and covered the many aspects of astrobiology.

The presentations on fossils and fossilization are aimed at elementary and middle school audiences. Presentations consisted of a brief lecture (some with slides), discussion and hands-on examination of fossils. A field trip to the tidepools was led for a 3rd grade class.

Publications

Back to Table of Contents

Astrobiology at UCLA: An Integrated Multidisciplinary Approach to Research and Education

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Back to Table of Contents

University of Colorado, Boulder

NASA Astrobiology Institute

University of Colorado, Boulder Team Members

Ariel D. Anbar
University of Rochester

Kat Anderson
University of Colorado Boulder

John Bally
University of Colorado Boulder

Erica Barth
University of Colorado Boulder

Carole E. Cleland
University of Colorado Boulder

Shelley Copley
University of Colorado Boulder

John Daspit
University of Colorado Boulder

Ico de Zwart
University of Colorado Boulder

Joyleen Desai
University of Colorado Boulder

Jasvinder H. Dhillon
University of Colorado Boulder

Gamelyn G. Dykstra
University of Colorado Boulder

Tian Feng
University of Colorado Boulder

William E. Friedman
University of Colorado Boulder

Kirk Harris
University of Colorado Boulder

T. Mark M. Harrison
University of California Los Angeles

Christine Hathaway
University of Colorado Boulder

Jennifer Heldmann
University of Colorado Boulder

Bruce M. Jakosky
University of Colorado Boulder

Bernard Marty
CRPG-CNRS

Kevin McGouldrick
University of Colorado Boulder

Kevin McKeegan
University of California Los Angeles

Steve Mojzsis
University of Colorado Boulder

Elinor Newman
University of Colorado Boulder

Glen Niccore
University of Colorado Boulder

Norman R. Pace
University of Colorado Boulder

Bo Reipurth
University of Colorado Boulder

Teresa L. Segura
University of Colorado Boulder

John Spear
University of Colorado Boulder

Nicholas Strohecker
University of Colorado Boulder

Henry B. Throop
University of Colorado Boulder

Owen B. Toon
University of Colorado Boulder

Stacy Varnes
University of Colorado Boulder

Jeffrey J. Walker
University of Colorado Boulder

Jennifer Winther
University of Colorado Boulder

Michael Yarus
University of Colorado Boulder

Executive Summary

Accomplishments

University of Colorado Center for Astrobiology has supported the following general activities:

- Sponsored public symposium on “Is there intelligent life elsewhere?”
- Sponsored graduate and undergraduate courses in astrobiology
- Prepared proposal to the CU Graduate School for a Graduate Certificate in Astrobiology
- Filled a faculty position in astrobiology with Prof. Stephen Mojzsis in Dept. of Geological Sciences
- Sponsored creation of an undergraduate “Astrobiology Society”
- Sponsored monthly astrobiology colloquium and informal Co-Investigator research forum

Research activities sponsored by the Center for Astrobiology using support from the NASA Astrobiology Institute included:

(i) Constraints on planet formation (John Bally, lead). We found compelling evidence for growth of dust grains in the outer portions of the largest protoplanetary disks in the Orion Nebula, providing insight into the very first stages of planet formation. Assessment of hazards of radiation and dynamical interactions for planet formation indicate that only 3-10 % of young stars will possess planetary systems.

(ii) Planetary climates (Brian Toon, lead). The efforts center on understanding processes that affect early climate on terrestrial planets. Results to date for Mars indicate (a) that impact release of crustal water may have played a substantial role in maintaining an early climate conducive to life and (b) that carbon dioxide clouds on Mars have radiative effects that make it difficult for them to contribute to the putative early greenhouse warming of Mars.

(iii) Setting the stage for the origin of life on Earth (Steve Mojzsis, lead). This is a new investigation this year exploring the geological environment on early Earth and the relevance for the origin and early history of life on Earth. However, efforts within the last twelve months by this group included using 4.3 Ga old zircons to obtain evidence for extensive liquid water and weathering at or near the Earth’s surface at that time.

(iv) RNA world and origin of life (Mike Yarus, lead). We have made progress in developing techniques for Direct Isolation of Catalysts or Enzymes (DICE) and anticipate carrying out the first isolations this coming year. We have synthesized new Transition State Analogues (TSAs) for the ribosomal peptidyl transferase ribozyme, and begun a set of selections for RNAs that bind these compounds; we hope that these new selections will show that the peptidyl transferase itself (or some close relative) can emerge directly from randomized RNA sequences, which would support the RNA World hypothesis.

(v) Toward a molecular phylogeny of a metabolic enzyme, maleylacetoacetate isomerase (Shelley Copley, lead). This is a new task added this year. The goal is to use the enzyme maleylacetoacetate (MAA) isomerase to study the evolution of metabolic pathways and the spread of metabolic genes. MAA has a puzzling phylogenetic distribution, in which the distribution is patchy, as might be expected if the gene has been distributed by infrequent lateral transfer events or lost selectively in certain lineages.

(vi) Molecular analysis of microbial ecosystems in extreme environments (Norman Pace, lead). Projects center around development and use of rRNA-based molecular methods to survey and study the microbial constituents of ecosystems in extreme environments. Recent results include the discovery of seven new kingdom-level phylogenetic groups of eucaryotes in anaerobic environments and the identification of hydrogen (as opposed to sulfur) as the fundamental energy source for thermophilic communities in Yellowstone hot springs.

(vii) Symbiosis and the origin of multicellularity in photosynthetic organisms (William Friedman, lead). We are studying symbiosis in order to understand major events in the evolution of life on Earth, including, for example, the origin of multicellularity. Results include beginning to understand one of the most important symbioses in evolutionary history, the mycorrhizal (plant-fungus) association in early land plant lineages; and analyzing the role of these symbioses in the colonization of land by photosynthetic organisms.

(viii) Energetics of life on other planets (Bruce Jakosky, lead). We are analyzing the geochemical environment of the surface and subsurface of Mars in order to determine the energy available from water-rock chemical reactions to support possible metabolism. Preliminary results have been obtained from models using the mineralogy and petrology of martian meteorites as constraints, and suggest that chemical energy is relatively limited due to the comparatively small contrast, by terrestrial standards, in oxidation state between groundwater and hydrothermal fluid.

(ix) Philosophical issues in astrobiology (Carol Cleland, lead). We are exploring the nature of scientific investigations as applied to astrobiology, in order to better understand historical vs. experimental sciences and the implications for, for example, the search for life on Mars. Results are used to point out the problems inherent in the Viking biology experiments and the difficulty of reaching concrete conclusions from them.

(x) Societal issues in astrobiology (Bruce Jakosky, lead). We are using astrobiology as a way to understand the relationship between science and society, the importance of educating the public as to the nature of science (and, equally importantly, of non-science), and the role of science in society today.

(xi) Commitment to hire new faculty. We met the commitment in our initial proposal to hire a new faculty member in astrobiology, by hiring Dr. Stephen Mojzsis as Assistant Professor in the Dept. of Geological Sciences. His research specialty is in the environment on the early Earth and the implications for the origin and early evolution of life on Earth and for the potential for life elsewhere. He arrived on campus in January 2001.

(xii) Undergraduate Astrobiology Society. We created an undergraduate “Astrobiology Society” to engage undergraduates in the excitement and breadth of the field. We just created it in spring 2001, and have met to have telescope viewing of the planets and an informal talk on astrobiology by one of the CU Co-Is. Future activities will include talks by visitors, a field trip to a local outcrop of the K-T boundary layer, and a visit to Lockheed Martin to see spacecraft assembly and operations.

(xiii) Proposal to the CU Graduate School to create a Graduate Certificate in Astrobiology. A graduate certificate is, in essence, the graduate equivalent of an undergraduate minor. We are going this way in lieu of creating a separate graduate degree in astrobiology, as the latter would serve to split students from their most natural intellectual home. Rather, a certificate provides added value to their degree with training in the broader aspects of astrobiology.

(xiv) Undergraduate and graduate classes in astrobiology. Sponsored teaching a graduate class in astrobiology (spring 2001), drawing some 16 students from both the physical and biological sciences. Also, sponsored teaching an undergraduate non-majors class in “Extraterrestrial Life” (fall 2000 and spring 2001), each time drawing about 70 students from all majors around campus. CU provided teaching assistant support as part of its commitment to the astrobiology program.

Astrophysical Constraints on Planet Formation

Project

Roadmap Objectives

#11
Origin of Habitable Planets

Senior Project Investigator(s):
J. Bally

Accomplishments

We have continued our exploration of planet formation in OB association star clusters (hot, young, massive stars). We have analyzed the colors of several proto-planetary disks in the Orion Nebula from 500 nm to 1870 nm. The translucent outer edge of the largest disk is achromatic, implying grains larger than 5 mm. We have also developed a model of grain growth in the presence of UV radiation induced photo-ablation.

We have shown that most young stars are born in either very over-dense but transient star clusters or in multiple star systems in which circumstellar disks may be truncated or destroyed by the close passage of nearest neighbor stars. Our best estimate is that only 3 to 10% of young stars will possess planetary systems. The rest of the stars lost their disks prematurely due to photo-ablation, dynamical interactions, or are in multiple star systems or bound clusters where planetary orbits may suffer large perturbations.

Highlights

We have found compelling evidence for grain growth in the outer portions of the largest proto-planetary disks in the Orion Nebula. Thus, we have seen the very first stages of planet formation.

NASA Mission Involvement

The current Hubble Space Telescope (HST) is of fundamental importance to our work, as it is the only telescope that can deliver the angular resolution needed to study circumstellar disks at the distance of Orion (1500 light years).

Our continuing survey of nearby star forming regions, (NOAO) will provide fundamental constraints on the likely abundance of planets around normal stars. Such information has provided a constraint on the design of the Terrestrial Planet Finder mission.

Future Directions

- Extend the search for large grains to longer wavelengths. We have been granted time on the Keck 10 meter telescope to use the adaptive optics system for near infrared imaging and spectroscopy of the largest disks in Orion. The Keck adaptive optics system is expected to deliver better images than the Hubble Space

Telescope. We will extend this study to a wavelength of 5 µm, which will constrain the population of grains to a size of about 15µm

• Propose to use the Very Large Array (VLA) radio telescope to search for evidence of cm to mm size particles and giant planets formed by gravitational instability. The VLA could in principle detect large grains by the thermal emission that such grains might produce. Indirect evidence for giant planets might be found by searching for non-thermal radio emission from the extended magnetosphere or thermal emission from a photo-ionized gas accreting onto a giant planet that is more than 30 AU from the central star.

• Continue our survey of the Perseus and Orion clouds to determine the locations of their young stars with respect to sources of UV radiation. We have obtained 14 nights of observing time at the National Optical Astronomy Observatories during fall 2001 for this purpose.

• Mine the 2MASS near infrared sky survey to constrain the formation frequency of stars in the solar vicinity.

• I will be taking on a new graduate student, and mentoring an undergraduate student. Both students will be involved with the analysis of new HST data and our ground-based observing campaigns.

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

#9
Life's Precursors & Habitats in the Outer Solar System

#12
Effects of Climate & Geology on Habitability

Project

Energetics of Life on Other Planets

Senior Project Investigator(s):
B. Jakosky

Accomplishments

The goal of this project is to understand the geochemical environment of other planets, Mars in particular, and the implications for geochemical resources to support potential life. We are exploring the chemical reactions available from interactions between water and rock on the Mars surface and in the subsurface and the amounts of energy released that could be utilized by putative martian organisms to support metabolism. Preliminary analysis using global estimates of the mineralogy and geochemistry indicate that there is a small but plausibly adequate supply of energy to support life. We are doing more detailed analysis of specific locations in order to better understand the constraints.

Progress so far has involved getting the EQ3/6 and SUPCRT92 software packages up and running at Colorado and understanding the subtle issues in how they work and what the calculations really mean. We have validated the models against previous calculations. We are applying the models to determine the energy available from reactions in the martian meteorites (starting with LEW 88516 as representative of the martian crust). Preliminary results indicate that chemical energy is relatively limited due to the comparatively small contrast, by terrestrial standards, in oxidation state between groundwater and hydrothermal fluid.

Future Directions

During the coming year, we will (i) expand upon the simplified hydrothermal model and from it produce a series of models including different rock and groundwater compositions to reflect different local geological environments (as derived using Mars meteorite, *in situ* spacecraft analysis, and orbital remote sensing analysis of surface composition and mineralogy) as well as different atmospheric compositions reflecting different climatic regimes, and (ii) apply the results to specific sites using the determination of surface mineralogy at high spatial resolution using results from the Mars Odyssey THEMIS experiment.

Molecular Analysis of Microbial Ecosystems in Extreme Environments

Project

Senior Project Investigator(s):
N. Pace

Accomplishments

Projects revolve around the development and use of ribosomal RNA (rRNA) based molecular methods to survey and study the microbial constituents of ecosystems in extreme environments without the requirement for cultivation of the organisms. This cultivation-independent approach to ecosystems analysis is essential rather than classic methods because most microbes, >99%, can not be cultured using standard techniques. With the molecular methods, rRNA genes are cloned directly from environmental DNA, and then sequenced to gain a phylogenetic snapshot of the organisms represented by the cloned genes. Some properties of organisms can be inferred from the phylogenetic results, and the sequences can be used to design hybridization probes to visualize organisms and their interactions in the environment. Studies relevant to NAI include:

Roadmap Objectives

#2
Origin of Life's Cellular Components

#3
Models for Life

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#10
Natural Migration of Life

#14
Ecosystem Response to Rapid Environmental Change

#16
Bringing Life with Us Beyond Earth

#17
Planetary Protection

• Antarctic and Colorado endolithic communities: Primary productivity in rocks occurs through the action of endolithic microbial communities, photosynthesis-driven communities in the outer few cm of any rock surface exposed to light. These communities so far have received only limited study with classic microscopy and culture techniques. Ongoing rRNA gene analyses of two selected communities from Antarctica and two in Colorado have revealed many novel kinds of organisms, some closely related to described organisms, but others very different. Although previously considered dominated by cyanobacteria, an abundance of chloroplast sequences has been detected. The nature of the eucaryal component expected is not yet known. Remarkably, an abundance of representatives of the Thermus/Deinococcus division of bacteria has been detected that were previously unknown, but are related to the radiation resistant deinococci. These new organisms also may predictably be similarly robust; the radiation resistant property of the deinococci is now thought to be toward dessication/oxidative damage.

• Yellowstone high-temperature settings: The laboratory has for many years studied thermophilic ecosystems at Yellowstone and elsewhere. Current activities continue to explore the makeup of properties of communities driven by hydrogen-metabolism, probably the dominant form of primary productivity in high-temperature settings anywhere.

• Anaerobic environmental eucaryotes: We have been conducting an rRNA-based survey of eucaryal phylotypes in anaerobic settings, for instance anaerobic marine and freshwater sediments. Recent results have identified a wealth of novel eucaryotic microbial diversity, including seven (!) novel kingdom-level clades, some among the most deeply divergent of eucaryal rRNA sequences. Attempts to learn more about those organisms represented by the sequences are underway.

• Studies of hypersaline microbial mats in concert with other NAI representatives began with an expedition to Guerrero Negro.

Highlights

• Discovery of seven new kingdom-level phylogenetic groups of eucaryotes in anaerobic environments, including forms that branch deeply in the eucaryal phylogenetic tree, and are highly divergent from known organisms

• Identification of hydrogen, not sulfur, as the fundamental energy source for thermophilic communities in Yellowstone hot springs

Field Trips

Yellowstone National Park, September, 2000.
Sampled hot springs for hydrogen. Participants: J. Spear, J. Walker.

Yellowstone National Park, October, 2000.
Sampled hot springs for hydrogen. Participants: J. Spear, J. Walker.

Guerrero Negro BCS, May, 2001.
Studied and sampled hypersaline microbial mats. Participants: N. Pace, J. Spear, R. Ley.

Philosophical and Societal Issues in Astrobiology

Project

Senior Project Investigator(s):
B. Jakosky

Roadmap Objectives

#18
Currently this project does not fit in a current category

Accomplishments

Our goal is to explore the connections between astrobiology and society and, more broadly, between the sciences and the science community and the broader society. The tremendous public interest in astrobiology is at a level far beyond the practical relevance of the topic, and suggests a public interest in the broader issues of exploration. In addition, we can use this interest as a way to get the public interested in science and science education, and to inform the public as to what science is (and, of equal importance, what science is not). As a way of furthering the exploration of these issues, We have (i) prepared and delivered many times a seminar on “What is astrobiology and why do we care?”, (ii) organized and participated in a special session panel discussion on “Societal connections of planetary exploration and the search for life elsewhere” at the 2001 Lunar and Planetary Science Conference as a way of raising these issues to the science community (about 250 scientists attended the session).

Highlights

Panel discussion at the 2001 Lunar and Planetary Science Conference on “Societal connections of planetary exploration and the search for life elsewhere”

NASA Mission Involvement

Considerations and activities in this project place NASA missions into the context of their ultimate goals with respect to the interests of the public. It also supports the NASA Administrator’s views on why we are exploring the planets and universe.

Future Directions

Complete book on “What is astrobiology and why do we care?”

Roadmap Objectives

#18
Currently this project does not fit in a current category

Project

Philosophical Issues in Astrobiology

Senior Project Investigator(s):
C. Cleland

Accomplishments

Our project deals with analysis of the differences in methodology between historical science and experimental science vis-à-vis the testing of hypotheses, as applied to astrobiology. We completed a more accurate account of the research practices of experimental scientists and historical research the previous year. We discovered that experiment plays a large number of different roles in science besides the classical role of testing hypotheses. We also determined that classical experimental research is best thought of in terms of an experimental program rather than (as traditional) isolated experiments. An experimental program involves a series of experiments each one designed in light of the results of previous experiments. Further investigation revealed that this is the central problem with robotic experiments like the Viking Lander metabolic experiments. One cannot determine in advance of actual experimental data what controls are adequate to rule out false positives and false negatives. These conclusions have implications for the development of instrument packages for detecting extraterrestrial life, namely that the probability of success is higher if one takes a historical approach (which involves emphasizing diversity of evidence collected and minimizing the use of antecedently designed controlled experiments).

We also began working on the question of whether "life" can be defined. Preliminary investigations suggest that trying to define "life" is a mistake. The purpose of definition is to analyze the meanings of terms. Supplying the current meaning of the term "life" is no more likely to provide us with an understanding of the inherent nature of life than supplying the sixteenth century meaning of "water" is likely to provide us with an understanding of the inherent nature of water. What is required is a theoretical framework for biology that would support identity statements analogous to "water is H_20."

Highlights

I discovered that classical experimental research is best thought of in terms of an experimental program rather than (as traditional) isolated experiments; an experimental program involves a series of interdependent experiments, each one designed in light of the results of previous experiments.

I discovered that robotic experiments (like the Viking Lander metabolic experiments) do not qualify as good experimental research, and that the emphasis in the design of future instrument packages for detecting extraterrestrial life ought to be on a historical approach, which involves doing a broad sweep (with little attention to controlled experiments) for very different sorts of evidence.

I discovered that attempts to answer the question "What is life?" by defining "life" are mistaken; what is required is a theoretical framework for biology that would support identity statements analogous to "water is H_20."

NASA Mission Involvement

In this project, we have no direct involvement in any NASA missions.

Nevertheless, my work on the nature of historical science and its differences from classical experimental science plus my current work on the nature/problem of "defining" life have significant implications for missions.

Perspective from my research to characterize historical science and classical experimental science suggests a particular preferred emphasis in design of future instrument packages for life detection. This activity might best be done using a broad sweep (historical science), rather than a series of controlled experiments (classical experimental science).

Future Directions

My goal for next year is to continue to work on applications of my analysis of historical and classical experimental research for astrobiological research, with particular emphasis on the design of robotic instrument packages for detecting extraterrestrial life. My other (related) goal is to work on the question "What is life?" Preliminary investigations indicate that trying to define "life" is a mistake. Next year I plan to look at current work in biology, particularly in the origins of life, for some hints about what an adequate theoretical framework would be like. This is a particularly tough project, so I do not anticipate finishing it by this time next year, although I hope to have it well underway.

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

#11
Origin of Habitable Planets

#12
Effects of Climate & Geology on Habitability

#15
Earth's Future Habitability

Project

Planetary Climates

Senior Project Investigator(s):
O. Toon

Accomplishments

We have made considerable progress understanding the role of large impacts in freeing water on Mars. We have cataloged the larger craters on the planet, and have performed simulations showing that considerable heating of subsurface reservoirs can occur. We have also completed a study showing that carbon dioxide clouds were not important for warming early Mars. New work is being conducted on the small stream beds recently observed on Mars.

Highlights

- Numerous craters on Mars show that very large impacts have occurred. These can place thick sheets of hot rock on the surface and lead to the release of great volumes of water, possibly explaining the river valleys on Mars.

- While carbon dioxide clouds may have occurred on Mars in the past they are unlikely to have been optically thick enough to create a significant greenhouse effect. Essentially the clouds warm the air where they occur which tends to dissipate the clouds.

Cross Team Collaborations

We have collaborated with the NASA Ames team through work with Margaret Tolbert, and Jim Kasting. In particular we have initiated studies with Kasting (Ames team and Penn State) on the escape of hydrogen from early Earth. We are working with Chris Mckay and Margaret Tolbert (Ames) on organic aerosols and on stream beds on Mars.

NASA Mission Involvement

In our research activity, we are using data from various martian spacecraft and a variety of instruments. In particular, we are using images of the martian surface plus lidar data from the current Mars orbiter (Mars Global Surveyor).

Future Directions

- Investigate streams in Greenland as analogs for martian streams

- Determine relations of martian streams to slopes, compute their possible lifetime

- Work with Penn State and JPL groups to compute the hydrogen escape rate from early Earth
- Publish paper on the role of impacts in generating rivers on Mars

RNA World and Origin of Life

Project

Senior Project Investigator(s):
M. Yarus

Roadmap Objectives

#2
Origin of Life's Cellular Components

#3
Models for Life

#17
Planetary Protection

Accomplishments

We explored fluor systems for use in the DICE (Direct Isolation of Catalysts or Enzymes) ribozyme isolation procedure, concluding that the initial methods proposed are very difficult and might require development of new single-molecule-detection technology. Therefore, we have switched to the immobilization of RNAs to be tested on agarose beads, along with a substrate that can be converted to a new product if the nearby RNAs include a ribozyme activity. The amplification needed to detect a few turnovers by the RNA enzyme is achieved by linking a second highly fluorescent bead specifically to areas where the product has been created. The bead carrying the active RNA is now a multi-bead system and highly fluorescent, and can be sorted out on a Fluorescence Activated Cell Sorter (FACS) machine, which is a type of standard biomedical lab technology.

A new project has been added: we have synthesized new Transition State Analogues (TSA) for the ribosomal peptidyl transferase ribozyme, and begun a set of selections for RNAs that bind these compounds. Use of previous TSAs synthesized in our laboratory has led to the identification of the peptidyl transferase (enzyme that forms peptide bonds in proteins) on the ribosome as an RNA enzyme and provided a substantial argument that the peptidyl transferase has been an RNA since the origin of protein biosynthesis in the RNA world. These new selections will hopefully show that the peptidyl transferase itself (or some close relative) can emerge directly from randomized RNA sequences. This would strongly confirm one of the primary predictions of the RNA world hypothesis, and accordingly support the hypothesis that RNA organisms were our immediate ancestors on Earth.

NASA Mission Involvement

This project work provides guidance for any NASA mission concerned with early stages of the origin of life. For example, if an RNA world existed (or exists) on Mars, this work can help recognize it.

Future Directions

- We are hoping to carry out our first DICE.

- We are hoping to create and characterize congeners of the biological peptidyl transferase ribozyme on our lab benches in Boulder, paralleling the appearance of this ribozyme at the dawn of Earth's biota.

Roadmap Objectives

Project

Setting the Stage for the Origin of Life on Earth

Senior Project Investigator(s):
S. Mojzsis

#1
Sources of Organics on Earth

#3
Models for Life

#5
Linking Planetary & Biological Evolution

#7
Extremes of Life

#8
Past and Present Life on Mars

#9
Life's Precursors & Habitats in the Outer Solar System

#12
Effects of Climate & Geology on Habitability

#13
Extrasolar Biomarkers

Accomplishments

This effort to investigate the origin of life on Earth was initiated at the CU-Boulder Center for Astrobiology in January, 2001, with the arrival of a new investigator (Mojzsis) from UCLA. We have been laying the foundation for the Astrobiology Materials Research Laboratory (AMRL) centered at the Department of Geological Sciences. The tasks completed so far include: refurbishment of laboratory space, transport of samples from the PI's UCLA collection to Colorado, purchase of equipment and supplies for the laboratory, establishment of a microscopy system for sample characterization, sample curation, purchase and installation of sample preparation devices (crushing systems, embedding media, etc.). Our current emphasis is on ramping up the laboratory for work following the summer field season.

Highlights

Progress in our understanding of the nature of Earth's earliest crust was made with the report of oxygen isotope compositions of ancient terrestrial zircons, up to 4.3 Ga in age. These zircons are the only direct record we have of Hadean-era surface environments of the Earth at the time when life emerged on this planet. We reported evidence for extensive liquid water and weathering at or near the Earth's surface before 4.3 Ga ago.

Developments in analytical techniques in the last year have made it possible to measure minute quantities of platinum-group elements (e.g. iridium) in ancient sediments to trace signatures of the late heavy bombardment in rocks older than 3.8 Ga.

Field studies in the Itsaq Gneiss Complex of southern West Greenland have confirmed that the ages of the oldest known sedimentary components are in excess of 3.8 Ga.

Development of a new multi-collection ion microprobe technique to explore non-mass-dependent sulfur isotope anomalies in ancient terrestrial and extraterrestrial sulfides confirm previous work by Farquhar and colleagues at UCSD (UCLA-NAI affiliation).

Field Trips

Southern West Greenland, Itsaq Gneiss Complex, June-July 2000.

UCLA-NAI led fieldwork to the Itsaq Gneiss Complex, southern West Greenland. The purpose of the expedition was to map and explore the oldest known sedimentary rocks on Earth and to trace their ages, origin and preservation. This work is ongoing. Members participating: S.J. Mojzsis (leader), T.M. Harrison, C.E. Manning.

Western Australia, Narryer Gneiss Complex, June-July 2001.

CU-NAI led fieldwork to the Narryer Gneiss Complex, Western Australia. Participants: S.J. Mojzsis (trip leader), T.M. Harrison (UCLA), M. Humayun (Univ. Chicago), J. Kirschvink (CalTech), B. Weiss (CalTech), T. Ireland (ANU), R. Pidgeon (Curtin), A. Nemchin (Curtin). This expedition is one of two pilot-projects of the Mission to Early Earth (MtEE) focus group of the NASA Astrobiology Institute. The other project, led by A.D. Anbar (Univ. Rochester) and R. Buick (Univ. Washington) will be visiting the Pilbara, also in Western Australia. The two groups will overlap for two days in the field.

NASA Mission Involvement

All of the work described in this report is directly applicable to planning NASA missions. Our work also provides the necessary background knowledge for better evaluation of geological data returned from other planetary surfaces, as well as for planning Mars sample return.

In this case, it may be argued that study of the ancient terrestrial rock record is akin to the study of "Mars on Earth."

Future Directions

- Establishment of an FTIR microscopy system for measurement of mineral compositions (e.g., nitrogen concentration) and calibration of nitrogen standards for FTIR microscopy in muscovite, biotite and plagioclase for subsequent nitrogen isotope measurement with collaborators

- Large-scale separation work on samples from Western Australia and western Greenland for zircon, apatite, mica, feldspar, Cr-spinel, sulfides and other mineral species for isotope and trace-element measurements

- Development of new analytical techniques at CU-Boulder and with collaborators to explore mineral inclusion chemistry of the oldest known terrestrial materials – zircons from Western Australia. These inclusions could provide more detailed information about the environment of formation of >4 Ga old zircons and thereby information about the origin and evolution of the first crust.

- Field programs in southern Wyoming to explore the preservation of Archean and Proterozoic crust in the Medicine Bow Mountains that contain detrital zircon populations up to 4 Ga in age. How do these compare with what is found in West Greenland and Western Australia?

- Trace the co-evolution of nitrogen and sulfur isotopes in ancient sediments to investigate changes in the composition of the atmosphere over time (particularly of oxygen). This will be accomplished by examining how non-mass-dependent signatures of Archean sulfides that record atmospheric chemical effects and changes in the $^{15}N/^{14}N$ of micas in the same rocks relate to each other. Emphasis will be placed on investigating the earliest Archean to Proterozoic rock record.

Roadmap Objectives

#4
Genomic Clues to Evolution

#6
Microbial Ecology

#10
Natural Migration of Life

Project

Symbiosis and the Origin of Multicellularity in Photosynthetic Organisms

Senior Project Investigator(s):
W. Friedman

Accomplishments

Symbioses, the mutually beneficial relationships between two organisms, have evolved numerous times over the course of the evolutionary history of life on Earth. Different forms of symbiosis have resulted in some of the most profound evolutionary radiations; e.g., the origin of the mitochondrion from a symbiosis of a proteobacterium with early eukaryotes, the origin of the chloroplast from a symbiosis of a cyanobacterium with an originally heterotrophic eukaryote resulting in the evolution of all photosynthetic lines of eukaryotes, and the establishment of multicellular photosynthetic organisms (plants) in terrestrial environments from a symbiosis of a fungus and a green alga.

We have begun to study, for the first time, the actual fungal symbiotic partners that may have been critical to the colonization of terrestrial environments on Earth by photosynthetic organisms. Using DNA extraction and amplification techniques, we have begun to identify the fungal lineages that currently play a central role in the widespread symbiosis between plants and fungi (mycorrhizal association). We have chosen to work with phylogenetically basal land plant lineages so that we can reconstruct the evolutionary history of this key symbiosis.

During the last year, a synthesis of the developmental biology of cells that conduct water within the body of terrestrial plants was completed and published in the Philosophical Transactions of the Royal Society. The goal of this work was to provide developmental insights for the paleontological community in their analysis of the earliest phases of the origin of terrestrial photosynthetic life and the microfossil record that was left behind.

Highlights

- Beginning to understand one of the most important symbioses in the evolutionary history of life on Earth, the mycorrhizal (plant-fungus) association in early land plant lineages

- Analyzing whether plants radiated into terrestrial environments or whether fungi in association with plants were the key to the one time (in the 4 billion year history of life on Earth) colonization of land by photosynthetic organisms

- First DNA sequence based identification of fungal symbionts of a phylogenetically basal land plant

- First developmental integration of information on water-conducting cells from extant early land plant lineages for the purpose of assisting with interpretation of the microfossil record of early multicellular terrestrial photosynthetic organisms

Future Directions

- Continue to identify the fungal partners of phylogenetically basal land plants based on amplification of rDNA sequences

- Perform BLAST and phylogenetic analyses to begin to formulate hypotheses about the number of origins of plant-fungal symbioses in the evolutionary history of life on Earth

- Continue developmental analyses to determine how phylogenetically basal lineages of terrestrial plants are infected with fungal symbionts – this will include confocal microscopy and other techniques

Roadmap Objectives

#4
Genomic Clues to Evolution

#6
Microbial Ecology

Project

Toward a Molecular Phylogeny of a Metabolic Enzyme, Maleylacetoacetate Isomerase

Senior Project Investigator(s):
S. Copley

Accomplishments

Maleylacetoacetate (MAA) isomerase is an enzyme involved in degradation of phenylalanine and tyrosine and certain other aromatic compounds. This enzyme provides an ideal model system for studies of the evolution of metabolic pathways and the spread of metabolic genes because of its puzzling phylogenetic distribution. Glutathione-dependent MAA isomerases occur in some proteobacteria, some plants, a nematode, mice and humans. In many other organisms (the Archaea, *Drosophila*, other proteobacteria), MAA isomerase genes cannot be identified in genome databases. It is not clear whether the distribution of the enzyme is patchy, as might be expected if the gene has been distributed by infrequent lateral transfer events or lost selectively in certain lineages, or whether the genes in these organisms are very dissimilar and are simply not recognized by search algorithms.

Our initial efforts have been directed toward identifying additional MAA isomerases in the unfinished microbial genome databases and developing a genetic system that will allow us to easily identify MAA isomerase genes from a variety of organisms. We have identified the MAA isomerase gene in *Pseudomonas putida* strain KT2440 and are working on knocking it out using pKnock. The knock out strain will be used for cloning MAA isomerase genes from other microorganisms.

We have also made progress on developing a convenient and quantitative assay for MAA isomerase activity. We have amplified a homogentisate dioxygenase gene and are working on cloning it into pQE30 (a vector that adds a His6-tag to the N-terminus of the protein). This protein will be used to prepare MAA from homogentisate. We have also amplified the gene for fumarylacetoacetate hydrolase from *P. putida* and are working on cloning it into pQE30 for expression in *E. coli*. This enzyme will be used to couple the isomerization of MAA to fumarylacetoacetate to the hydrolysis of fumarylacetoacetate to fumarate and acetoacetate (two stable products). This procedure will allow us to use a continuous UV/vis assay to quantitate MAA isomerase activity in proteins from various sources.

Highlights

Work on this project began in January, 2001 and all aspects are still in progress.

Future Directions

- Prepare libraries of genomic DNA from various organisms , transform the libraries into the knock-out strain, and select colonies that have regained the ability to grow on tyrosine

- Sequence the MAA isomerase genes identified using the selection procedure

- Express and purify suspected MAA isomerase proteins and quantify their enzymatic activity

Education & Public Outreach

Is There Intelligent Life Elsewhere?

Every fall the University of Colorado Center for Astrobiology puts on a public symposium relating to astrobiology. The symposia are well-attended by the public as well as students, and the auditorium is generally filled to capacity. This year the topic was intelligent life and the possibility it might exist elsewhere extraterrestrially. Four speakers were invited to give brief (20 minute) presentations, and opened the floor to audience Questions & Answers for an additional 15 minutes at the end. Afterwards an informal reception allowed attendees to meet the speakers.

Jakosky public and invited talks

Jakosky & members of his team are regularly invited throughout the year to give lectures and presentations on astrobiology in professional, educational, and public venues. This year, six presentations were given. Highlights included the Lunar and Planetary Institute's Annual Teacher's Workshop, a Biology class at the United States Air Force Academy and the Undergraduate Astrobiology Society on the University of Colorado campus.

Undergraduate Astrobiology Society

The Undergraduate Astrobiology Society in a student-run, Co-Investigator mentored program designed to reach undergraduates interested in astrobiology. The Astrobiology Society holds monthly meetings during the school year with astrobiology-related activities such as planetarium visits, lectures, and informal seminars for undergraduates interested in astrobiology.

Undergraduate Courses in Astrobiology

An undergraduate course entitled "ET Life" is offered on the University of Colorado campus to upper division undergraduates during the fall and spring semesters. The course addresses a broad range of astrobiology issues, including origins of life, planetary system formations, and the sociological implications of extraterrestrial life and the search for it. The class consists of lecture and discussion of the topics, with student participation highly encouraged.

Undergraduate Internship Program

In this program, undergraduate students with an interest in astrobiology are given an opportunity to work in the labs of the Principal Investigator or one of the Co-Investigators, thereby gaining experience in the field of astrobiology. Interns help with compiling data or laboratory/field experiments and receive mentoring. They are supported by the Center for Astrobiology member with whom they work.

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Back to Table of Contents

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Back to Table of Contents

NASA
Astrobiology
Institute

New Lead Teams

The selection of four new NAI Lead Teams was announced in March 2001, and their cooperative agreements were initiated in July 2001.

Reports here for the four new NAI Lead Teams are 'Executive Summaries' that profile their planned astrobiology research projects. Accomplishments, education/public outreach, and publications are not reported here, since the NAI Year 3 period ends with June 2001, before their cooperative agreements were initiated.

Jet Propulsion Laboratory (2)

NASA Astrobiology Institute

Pasadena, California

Principal Investigator

Victoria Meadows

Jet Propulsion Laboratory (2) Team Members

Mark Allen
Jet Propulsion Laboratory

Linda Brown
Jet Propulsion Laboratory

Martin Cohen
University of California Berkeley

David Crisp
Jet Propulsion Laboratory

Amir Fijany
Jet Propulsion Laboratory

Keliann Grasmik
California Institute Of Technology

Wesley T. Huntress
Carnegie Institution Of Washington

James F. Kasting
Pennsylvania State University

Victoria S. Meadows
Jet Propulsion Laboratory

Cherilynn Morrow
Space Science Institute

Kenneth H. Nealson
Jet Propulsion Laboratory

Mark Richardson
California Institute Of Technology

Robert Rye
California Institute Of Technology

Norman Sleep
Stanford University

Michael Storrie-Lombardi
Jet Propulsion Laboratory

Thangasamy Velusamy
Jet Propulsion Laboratory

Yuk Yung
California Institute Of Technology

Kevin Zahnle
Ames Research Center

Executive Summary

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#12
Effects of Climate & Geology on Habitability

#13
Extrasolar Biomarkers

#14
Ecosystem Response to Rapid Environmental Change

Motivated by the recent discoveries of a multitude of extrasolar planets, NASA has initiated a series of studies for space-based observatories that will be able to search for life on these worlds. To optimize the designs of these NASA missions, and to ultimately interpret the data that they return, we need to be able to recognize habitable worlds and to discriminate between planets with and without life.

The principal goal of this research is to learn how to recognize the presence of life on extrasolar planets by identifying the signatures of life in their spectra. To achieve this goal, we are developing a suite of innovative modeling tools to simulate the environments and spectra of extrasolar planets. The modeling tools will constitute a Virtual Planetary Laboratory, which will be used to explore the plausible range of atmospheric compositions and globally-averaged spectra for early Earth, other planets in our solar system, and for extrasolar planets both with and without life. The results of this research will provide an improved understanding of the range of atmospheric compositions that are possible for planets with and without life, and will help to quantify the effect of life on the atmospheric spectrum and composition of a planet. The models will also provide a comprehensive spectral catalog, a “menu” of biosignatures, which will be used to determine the optimum wavelength range, spectral resolution, and sensitivity required to remotely sense the signs of life in the atmosphere or on the surface of another world. This study will also provide recommendations for the design and optimization of the search strategies for future NASA planet detecting and characterizing missions such as TPF and Life Finder.

Field Trips

Feeding into our Task 5, we will collaborate with NAI JPL1 to send some of our members on joint field trips to explore the spectral detectability of the presence or action of microbial life on a planet’s surface, at a number of different spatial scales. This may include a trip to Hawaii to explore the detectability of microbial weathering of basalt using basalts spanning a range of ages.

NASA Mission Involvement

This research program will provide practical, timely, and much-needed recommendations and guidelines for development of future NASA missions that seek to detect and characterize extrasolar terrestrial planets, such as TPF (Terrestrial Planet Finder) and its second generation follow-on, Life Finder. Our work will drive the design and survey strategies of these missions, and it will also provide an initial solid theoretical basis for the interpretation of the results of these missions.

The modeling tools and spectroscopic results that we produce will also be valuable for basic research in many other NASA research programs. These include Planetary Astronomy, Planetary Atmospheres, Sun-Earth Connection and the Origins program. Such programs provide basic research that feeds into mission definition and design for robotic exploration of the solar system. Our tools and results may also be of use in the design of future NASA Discovery proposals that seek to discover or characterize extrasolar planets.

Future Directions

The first year of this research effort is to build a Virtual Planetary Laboratory (VPL), which will see the completion of proposal Task 1, the partial completion of Task 2, and the commencement of Tasks 4 and 5. Specifically:

- Task 1 will determine sensitivity to environmental characteristics in the globally averaged synthetic spectra of terrestrial planets using a radiative transfer model, and will provide the first step for development of the subsequent modeling tools.

- Task 2 is the next step in the development of the modeling tools, and will validate the coupling of the radiative transfer and climate model, and the 1-D globally averaged approach used in our modeling.

- Task 4 will start simultaneously with Task 1. In this task we will design and develop the models for the geological and exospheric processes identified in our proposal. These processes will eventually be implemented in the Abiotic Planet Model component of the VPL on completion of the mechanistic core at the end of year 2.

- Task 5 will start simultaneously with Task 1 and will identify biological sources and sinks of volatiles for parameterization and ultimate inclusion into the VPL to create the Inhabited Planet Model.

Back to Table of Contents

Michigan State University

NASA Astrobiology Institute

East Lansing, Michigan

Michigan State University Team Members

Albert Bennet
University Of California Irvine

Elbert Branscomb
DOE Joint Genome Institute

David Gilichinksy
Michigan State University

Richard Goldstein
University of Michigan

Trevor Hawkins
DOE Joint Genome Institute

Sophie Kathariou
North Carolina State University

Richard Lenski
Michigan State University

David Lubman
University of Michigan

John McGrath
Michigan State University

Paul Predki
DOE Joint Genome Institute

L. Sepulveda
University of Puerto Rico Mayaguez

Michael F. Thomashow
Michigan State University

James Tiedje
Michigan State University

Executive Summary

Roadmap Objectives

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#7
Extremes of Life

#16
Bringing Life with Us Beyond Earth

Low temperature is a predominant environmental characteristic of interstellar space, our solar system, including most of the planets and their satellites, and asteroids and meteors. An understanding of the impact of low temperatures on the responses and evolution of biological organisms is, thus, integral to our knowledge of astrobiology. The research that we propose will explore multiple aspects of microbial adaptation to low temperatures. One major line of investigation will be to conduct structural and functional genomic and proteomic analyses of bacteria that have been isolated from the Arctic and Antarctic permafrost. What genes and proteins enable the permafrost bacteria to inhabit these subfreezing environments? Do they have specific "freezing tolerance" genes and proteins, or "specialized alleles" of commonly found bacterial genes, or both? How is expression of the bacterial genome affected by low temperatures and other conditions that "hitchhiker" bacteria might encounter during travel through space on natural objects or spacecraft? In a second line of investigation, we will directly examine, through "test-tube evolution" experiments, bacterial adaptation to low temperatures. The studies will provide insight into how an organism, with a given complement of genes, can cross niche barriers that are defined by decreasing temperatures. And finally, we will use the information gained to explore the potential development of "signatures" for the presence of life in cold environments including Earth and other bodies such as Mars and Europa.

The proposed studies will provide significant new information relating to multiple goals outlined in the "Astrobiology Roadmap" including: Explore how life evolves on the molecular, organism and ecosystem levels (goal 3); Determine how the terrestrial biosphere has co-evolved with the Earth (goal 4); Establish limits for life in environments that provide analogues for conditions on other worlds (goal 5); Determine how to recognize the signature of life on other worlds (goal 7); and Understand the response of terrestrial life to conditions in space or on other planets (goal 10). Moreover, the proposed studies address one of the perceived gaps in the current NAI research program, namely, "provide understanding of the response of life to the space environment, from gene expression to microbial evolution." Finally, there are significant potential practical applications of the work ranging from the identification of genes that may be used to confer improved environmental stress tolerance in crop plants to the discovery of enzymes uniquely suited to catalysis at low temperature, a characteristic of importance in numerous biotechnology applications.

The proposed lines of interrelated investigations call for a broad range of expertise. Consequently, we have assembled a group of investigators with diverse backgrounds, training and research interests. The areas of expertise include microbial ecology (Tiedje, Sepulveda), the isolation and characterization of permafrost bacteria (Gilichinsky, Tiedje), permafrost geology and geochemistry (Gilichinsky), molecular genetics and gene regulation (Kathariou, Thomashow), evolution and population genetics (Lenski, Bennett), cryobiology and mechanisms of freezing tolerance (McGrath, Thomashow), proteomics and protein evolution (Lubman, Goldstein), and structural genomics (Branscomb, Hawkins, Predki). As the proposed lines of research develop, we anticipate that the Center will evolve, bringing in new investigators to provide expertise to attack the next generation of questions. For instance, the structural genomic and gene expression profiling studies will lead to lines of investigation focused on specific genes with the objectives of establishing their roles in cold tolerance; determining their modes of action; and developing hypotheses as to how the genes evolved. While the current Center investigators are well qualified to conduct these lines of research, it may, perhaps, become evident that adding an investigator with expertise in using X-ray crystallography and/or NMR-spectroscopy to determine protein structure-function relationships would strengthen the research efforts.

We also envision that additional lines of investigation will evolve out of our interactions with researchers in other Centers of the Astrobiology Institute. For instance, among our objectives are to conduct experiments to determine how bacterial gene expression and evolution are affected by conditions that relate to the space environment. Our major focus will be on cold temperatures. However, it seems likely that it would be of interest and importance to other Center members to understand how microorganisms react, in terms of gene expression and evolution, to other environmental conditions of space such as the Martian atmosphere and regolith or perhaps microgravity and solar radiation. We view the defining of mutual research interests and development of collaborative Center research efforts to address fundamental questions in astrobiology to be an underlying goal of our program.

NASA Mission Involvement

Our proposed research studies will provide significant new information relating to multiple goals outlined in the “Astrobiology Roadmap” listing. These include: 1) Explore how life evolves on the molecular, organism and ecosystem levels; 2)

Determine how the terrestrial biosphere has co-evolved with the Earth; 3) Establish limits for life in environments that provide analogues for condition on other worlds; 4) Determine how to recognize the signature of life on other worlds; and 5) Understand the response of terrestrial life to conditions in space or on other planets.

The information from our research project will thus be relevant in a general way to planning future NASA missions and may provide specific information relevant to detection of past (or present) life on other planets and the potential of transmitting "hitchhiker" microbial life to other sites in the solar system.

Future Directions

- Complete general physiological characterization of bacterial permafrost isolates including a determination of whether antifreeze proteins are produced.

- Conduct a general genetic characterization of the permafrost isolates to determine which are best candidates for detailed genetic analysis.

- Initiate genome sequencing and RNA profiling of permafrost bacteria.

- Initiate proteome profiling (of most highly expressed proteins) of permafrost bacteria.

- Initiate "test-tube" evolution studies on microbial adaptation to low temperature.

- Initiate isolation and characterization of tropical bacteria related to permafrost isolates for "field-truth" experiments designed to assess the importance of cold adaptive traits to success in nature.

Back to Table of Contents

Year 3

University of Rhode Island

NASA Astrobiology Institute

Kingston, Rhode Island

Principal Investigator

Steven D'Hondt

University of Rhode Island Team Members

Steven D'Hondt
University of Rhode Island

Kai-Uwe Hinrichs
Woods Hole Oceanographic Institute

David C. Smith
University of Rhode Island

Arthur Spivack
University of Rhode Island

Andreas P. Teske
Woods Hole Oceanographic Institute

Executive Summary

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#13
Extrasolar Biomarkers

#15
Earth's Future Habitability

A better understanding of the Earth's deep biosphere is essential because it can serve as a model for life on other planets and it is a critical component of the Earth's biogeochemical cycles. This research is aimed at gaining a fundamental understanding of the life in deeply buried marine sediments. We are undertaking an interdisciplinary set of projects that take advantage of our considerable expertise in marine sedimentary microbiology, sedimentary biogeochemistry and deep ocean drilling. Our objectives are to understand the subsurface microbial ecosystems of marine sediments, their role in Earth's biogeochemical cycles, and their relevance to the search for life on other planets. We focus on three major projects.

The first project explores the taxonomic composition, metabolic activity and geochemical consequences of buried microbial ecosystems in marine sediments with widely different physical and chemical characteristics Environments include hot, deeply buried anoxic sediments where life may exist independently of the photosynthesis-based ecosystem at Earth's surface, and old, deeply buried sediments where life may be limited by the availability of electron donors or key nutrients. The ecosystems of such subsurface habitats are potentially representative of the ecosystems that may exist on other planets.

The second project documents the nature, extent and perturbation-sensitivity of microbial activity in marine sediments and the effect of that activity on Earth's biogeochemical cycles of various chemical species, particularly sulfate and methane. Sulfate, the second most abundant anion in seawater, is the dominant terminal electron acceptor in marine sediments. Methane is climatically active and marine methane deposits comprise the largest untapped hydrocarbon reservoir on Earth. The biogeochemical cycles of these and other chemical species are potentially sensitive to centennial to millenial changes in Earth's surface temperatures, nutrient fluxes to the ocean, and the structure of marine ecosystems.

The third project focuses on identifying signatures of present and past microbial processes in Earth's subsurface as a guide to predicting such signatures in extraterrestrial subsurface environments. The molecular-isotopic based data that will be generated as part of this project will complement the objectives of the first two projects. For example, the study of biomarkers produced by deeply buried methanotrophic microbes give us an indication of both past and present activity of this specific taxonomic group.

We have already obtained extensive sample sets from deep Pacific drill-holes for analysis. We developed procedures for identifying contamination of samples, spearheaded the drive to instrument a microbiology laboratory on the Ocean Drilling Program (ODP) drill-ship JOIDES Resolution, and compiled relevant global geochemical and physical databases. We will participate in the first ODP Leg (Leg 201) dedicated to Subsurface Biosphere research scheduled for 2002. These and other developments provide the framework for collecting, analyzing and interpreting microbiological, biogeochemical and physical data from a wide range of deep subsurface samples. This framework leaves us poised to greatly advance subsurface biosphere objectives.

Our approach is fundamentally interdisciplinary and we will use advanced techniques from various fields, including microbiology, molecular biology, organic and inorganic biogeochemistry, isotope geochemistry, large data set integration and computational modeling. The work will be field-, laboratory-, and model-based.

NASA Mission Involvement

This project is not presently affiliated with any specific missions. However, its results will ultimately be relevant to the search for life on Mars and Europa, as well as to the search for biogeochemical signatures of life on more distant planetary bodies.

Future Directions

Our primary first-year goals will include (1) biogeochemical and microbiological study of a model system for a deep anaerobic biosphere where anaerobic carbon recycling occurs via adjacent methanogenesis and methane oxidation, and (2) field-study of factors that limit microbial activity and presence or absence in low-activity subsurface ecosystems.

In order to address these goals in this year, we will participate in the first deep-sea drilling cruise ever focused on subsurface biosphere [Ocean Drilling Program Leg 201 to the Peru Margin and the eastern equatorial Pacific (from February to April)]. We were the principal proponents of this international drilling project. Both during and after the cruise, we will collaborate with numerous international partners, probably including members of other NAI teams (the cruise team is presently being assembled).

Environments targeted for drilling by this cruise include both cold organic-poor sediments that are likely to contain inactive, dead or slow-growing microbial communities and microbiologically very active organic-rich sediments that are most appropriate for studying the activity and composition of methanogenic and anaerobic methanotrophic subsurface communities.

- In preparation for this field expedition, we will refine and develop a number of microbiological and geochemical techniques for shipboard and post-cruise studies.

- In this year, our studies of biogeochemical cycles will also be advanced by using our global compilations of subsurface sulfate and methane data to develop robust estimates for local and global rates of sulfate reduction and methanotrophy in the subsurface biosphere of oceanic sediments.

- Personnel objectives of special emphasis during the first year will be recruitment and training of graduate students, post-doctoral scholars, and undergraduate research fellows at GSO and WHOI.

- In this year, we will also develop a working and readily accessible website on Astrobiology and Subsurface Biospheres.

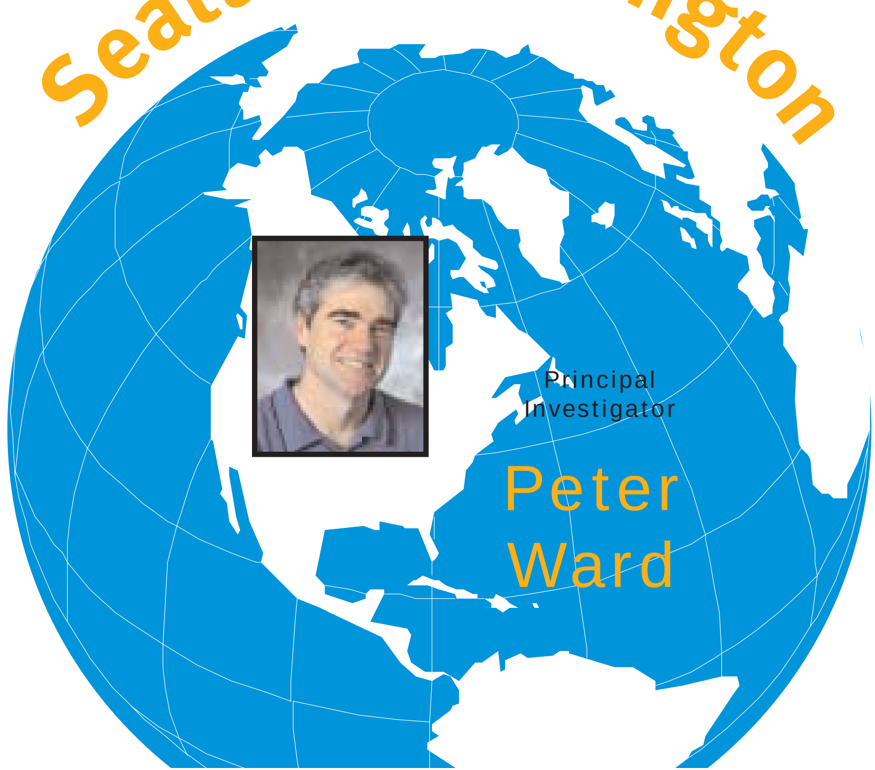
Back to Table of Contents

Year 3

University of Washington

NASA Astrobiology Institute

Seattle, Washington



Principal Investigator

Peter Ward

University of Washington Team Members

John A. Baross
University Of Washington

Donald Brownlee
University Of Washington

Roger Buick
University Of Washington

Jody Deming
University Of Washington

Jesse Dillon
University Of Washington

Ken Farley
California Institute Of Technology

Guillermo Gonzalez
University Of Washington

Linda Khandro
University Of Washington

Monica Kress
University Of Washington

David A. Kring
University Of Arizona

John Leigh
University Of Washington

Conway Leovy
University Of Washington

Jonathon Lunine
University Of Arizona

Thomas Quinn
University Of Washington

Mitchell L. Sogin
Marine Biological Laboratory

Viatcheslev Solomatov
New Mexico State University

David Stahl
University Of Washington

James Staley
University Of Washington

Woodruff Sullivan
University Of Washington

Peter D. Ward
University Of Washington

Stephen Warren
University Of Washington

Executive Summary

Roadmap Objectives

#1
Sources of Organics on Earth

#2
Origin of Life's Cellular Components

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#11
Origin of Habitable Planets

#12
Effects of Climate & Geology on Habitability

#14
Ecosystem Response to Rapid Environmental Change

The surge of interest in astrobiology has resulted in new information about many aspects of early evolution and the range of conditions under which life can exist, based largely on new understandings of extremophilic microbes. Far less understood, however, is the frequency and range of conditions under which more complex organisms, specifically metazoans, might occur and survive for long periods of time. Together, astrophysical and geophysical processes have provided the molecular components of life, the carbon and energy sources to sustain life, and the diverse and changing environments on Earth that favor evolution and biocomplexity. In fact, increasing biocomplexity is one common trend of the evolution of life on Earth and is the result of a co-evolution of organisms with their environment. The interchange between the complex environmental factors and molecular mechanisms that led to the evolution of eukaryotes and metazoans from a common microbial ancestor are not known. However, recent advances in isotopic and organic analyses of rocks, molecular techniques, and astrophysical models have made it possible to begin the task of understanding how life emerged and evolved biocomplexity and what life forms could be expected to exist on other planets that have evolved through geological stages that overlap conditions on Earth during the past 4 Ga.

Our over-reaching assumption is that Earth life is representative of life elsewhere in that it will be carbon based, require liquid water, and use energy sources that are associated with universal astro- and geophysical processes. Our research program is focused on planetary habitability and the evolution of biological complexity. This is integrated multidisciplinary effort with concentration on four broad problems:

1. How often do planets with truly Earth-like properties form?

2. How important is plate tectonics in the formation and maintenance of metazoan life?

3. How important are mass extinctions for the evolution and extinction of complex life: are mass extinctions fertilizer or poison (or both) in the garden of complex organisms?

4. What are the evolutionary pathways by which complex organisms originate from microbes?

NASA Mission Involvement

At the current time, there is no direct link to a planned NASA mission other than Project Stardust. This Discovery Mission project has Dr. Brownlee as the Stardust team PI.

Stardust will gather and return samples of interstellar dust from our Solar System as it flies by a comet called Wild-2 in January 2004 and returns to Earth in 2006.

Future Directions

1. Investigate the Triassic-Jurassic mass extinction in outcrops in Western Canada. We will sample outcrops in the Queen Charlotte islands

2. Investigate the Permian-Triassic mass extinction in South Africa. We will sample and analyze samples from the Karoo of South Africa

3. Model the formation of emerging stellar systems and planets. We will use computer models to examine planetary formation

4. Study the nature of cometary material falling onto the Earth. We will examine the chemistry of interplanetary dust particles.

5. Study the biology of microbes in extreme environments. We will examine the ecology of extremophiles in various environments

6. Study the genetics of microbial lineages showing evidence of accumulating morphological complexity. We will use DNA sequencing to better understand critical microbial lineages in an attempt to better understand the rise of eukaryotes.

Back to Table of Contents

NASA
Astrobiology
Institute

Focus Groups

NAI Focus Groups are research and planning teams formed around topics relevant to specific NAI goals and objectives. Established through proposals submitted to the NAI's Director, Focus Groups contribute to astrobiology space missions, extend long-distance collaborations by exploiting networking technologies, and innovate research directions.

Roadmap Objectives

#1
Sources of Organics on Earth

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#8
Past and Present Life on Mars

#12
Effects of Climate & Geology on Habitability

#17
Planetary Protection

Project

Astromaterials

Focus Group Chair:
D. McKay

Focus Group Members:

Carl Agee
Johnson Space Center

Baruch S. Blumberg
NASA Astrobiology Institute

Henry Bortman
NASA Astrobiology Institute

Arthur Bowman
Hampton University

Monica Byrne
Wellesley College

Simon J. Clemett
Lockheed Martin

Everett K. Gibson
Johnson Space Center

Richard Hoover
Marshall Space Flight Center

Thomas Kieft
New Mexico Institute of Mining and Technology

David J. Lindstrom
Johnson Space Center

Marilyn Lindstrom
Johnson Space Center

John Lisle
Lockheed Martin

Teresa Longazo
Lockheed Martin

David S. McKay
Johnson Space Center

Robert D. Minard
Pennsylvania State University

Jeffrey Plescia
USGS Menlo Park

Luke Probst
Bemidji State University

Rusty Schweickart
California Academy of Sciences

Sean C. Solomon
Carnegie Institution of Washington

Andrew Steele
Johnson Space Center

Karen Stocco

G. Jeff Taylor
University Hawaii

Kay W. Tobola
Johnson Space Center

Kathie Thomas-Keprta
Lockheed Martin

Jan Toporski
Portsmouth University (UK)

Allan H. Treiman
Lunar And Planetary Institute

Malcolm Walter
Macquarie University

Susan J. Wentworth
Lockheed Martin

Accomplishments

Astromaterials include lunar samples, meteorites, interplanetary dust particles, and any sample returned to Earth by a space mission. The objective of the newly created Astromaterials Focus Group is to coordinate and facilitate astrobiological investigations of Mars meteorites, terrestrial analogs of potential Mars samples, and sample acquisition and collection technology and procedures; and in general to advance the development of instruments and techniques for examining all astromaterials for evidence of past or present Life.

The initial efforts of the Astromaterials Focus Group have been directed towards the fifteen known Mars meteorites which are available for scientific analysis. These meteorites, together with other examples of astromaterials such as carbonaceous chondrites and interplanetary dust particles (IDP), contain organic compounds which may be the result of abiotic synthesis. The study of such materials can provide basic information on extraterrestrial prebiotic chemistry, addressing such questions as; what compounds may have been available before life began, what was their relative abundance, and where were they most abundant? Other major areas of investigation which can be advanced by the examination of these samples include, 1) evidence for the present or past occurrence of water on Mars, 2) age dating and the history of Mars, 3) possible microfossils in the Martian environment, and 4) the development of new life detection techniques.

Highlights

- The Astromaterials Focus Group includes studies of Mars meteorites, studies of terrestrial analogs of potential Mars samples, studies of sample acquisition and collection technology and procedures, and development of instruments for examining astromaterials for any evidence for life.

- The Astromaterials Focus Group can, therefore, act as a bridge between the astrobiology community and the astromaterials community. Its efforts are improving communication and understanding between these two groups of researchers.

Field Trips

Although the Astromaterials Focus Group has not initiated any field expeditions to date, this Focus Group can serve to track and coordinate the use of astromaterial samples collected by others in the astrobiology community. The Astromaterials Focus Group can also take responsibility for the description of meteorites, IDPs, and other astromaterials which might be of interest to the broader astrobiology community.

NASA Mission Involvement

The NAI Astromaterials Focus Group will have responsibility for monitoring research on Mars meteorites that bears on the question of possible life on Mars. Such information should feed into NASA Mars mission planning as part of the NASA Astrobiology Institute contribution to site selection, instrument requirements, operational requirements, and returned sample requirements. This focus group should have strong recommendations to make to mission planners for the 2007 lander mission and for the 2011 sample return mission, and it should also be the main bridge between the NAI and the Planetary Protection community.

Future Directions

The Astromaterials Focus Group is planning to work with personnel at the NASA Ames Research Center to prepare a workshop for mid-November, 2001 which will review and examine data related to biogenic magnetite in Mars Meteorite ALH84001. The workshop of invited speakers will cover the evidence for and against biogenic magnetite in ALH84001. A questionnaire is also being formulated by the Astromaterials Focus Group to be sent to all of its members and to the NAI Executive Council which will solicit opinions on, and arguments for and arguments against, maintaining future Mars samples cold during collection, packaging on Mars, return to Earth, and in the sample receiving and processing lab.

Back to Table of Contents

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#7
Extremes of Life

#9
Life's Precursors & Habitats in the Outer Solar System

#12
Effects of Climate & Geology on Habitability

Project

Europa

Focus Group Chair:
R. Greeley

Focus Group Members:

Louis Allamandola (ARC)
Amy Barr University of Colorado Boulder
Deborah Bass SWRI
Max Bernstein (ARC)
David Blake (ARC)
Baruch S. Blumberg (NAI)
Henry Bortman (NAI)
Jerome Borucki (ARC)
Bob Carlson (JPL)
Christian Caron Springer-Verlag (Germany)
Frank Carsey (JPL)
Frank Chuang USGS Flagstaff
Chris Chyba (ARC)
Ben Clark Lockheed Martin
David Cole USA CRREL
Max Coon Northwest Res. Assoc.
John Cooper GSFC Raytheon
Andrew D. Czaja (UCLA)
Steven D'Hondt University of Rhode Island
Brad Dalton (ARC)
Leonard David Space.com
Wanda Davis SETI Institute
Jody Deming (UoW)

Jason P. Dworkin (ARC)

Hajo Eicken
University of Alaska

Lisa Faithorn (NAI)

Jack D. Farmer (ASU)

Michael Flynn

Marilyn L. Fogel (CIW)

Christian Fritsen
Desert Research Institute

Gian Gabriele Ori
In't Res. School of Planetary Sciences

Rebecca J. Gast
Woods Hole Oceanographic Institute

Zann Gill
NASA

James Granahan
BAE Systems

Ronald Greeley (ASU)

Rose A. Grymes (NAI)

Tracey Herrera (UCLA)

Paul Holland
Throleaf Research Inc.

Christopher House (PSU)

Louis Irwin
University of Texas at El Paso

Robert Johnson
University of Virginia

Torrence Johnson (JPL)

Jeffrey Kargel
USGS Menlo Park

Jozef Kazmierczak
Polish Acad. Sciences

Stephan Kempe
Geol. Paleontol Institute

Bishum Khare (ARC)

Krishan Khurana (UCLA)

R.M. Killen
SWRI

Kenneth Klaasen (JPL)

Dan Kojiro

A. Lonne Lane (JPL)

Robin Lloyd
Space.com

James Lyons (UCLA)

Nicholas Makris
Massachusetts Institute of Technology

Giles Marion
Desert Research Institute

Tom McCollom
Woods Hole Oceanographic Institute

Tom McCord
University of Hawaii

Henry McDonald (ARC)

Christopher McKay (ARC)

Bruce Moommaw

Jeff Moore (ARC)

William B. Moore (UCLA)

David Morrison (NAI)

Rafael Navarro-Gonzalez
UNAM (Mexico)

Susanne Neuer (ASU)

Doug O'Handley (ARC)

Robert Pappalardo
Brown University

Nikia Paraham

Cynthia Phillips
SETI Institute

Carl Pilcher
NASA Headquarters

Ogla Prieto
Centro de Estudios Universitarios

Daniel Prieur
University de Bretagne Occidentale

Louise Prockter
John Hopkins University

David S. Ross
USGS Menlo Park

Bruce N. Runnegar (UCLA)

Scott Sandford (ARC)

Paul Schenk

Greg Schmidt (ARC)

Erland Schulson
Dartmouth College

Dirk Schulz-Makuch
University of Texas at El Paso

Nicole Spaun
Brown University

John Spencer
Lowell Observatory

Timothy Swindle
University of Arizona

Lesile Tamppari (JPL)

Aaron Thode
Massachusetts Institute of Technology

Luis Vazquez-Martinez
University Complutense de Madrid

Brian Wade (ASU)

Kimberly Warren-Rhodes (ARC)

Chuck Weisbin (JPL)

Llyd E. Wells (UoW)

Kevin Zahnle (ARC)

Daniel Zeigler
Ohio State University

Accomplishments

The objective of the Europa Focus Group (EFG) is to foster scientific studies and joint investigations of Europa and related research in order to understand the exobiological potential of this satellite of Jupiter. This goal is to be met through various video conferences, workshops, and information exchanges sponsored by the NAI. The study of Europa requires a multidisciplinary approach using spacecraft data analyses, laboratory simulations, terrestrial analog studies, and other techniques to understand the complex history and present environments on and in Europa. Participation in the EFG is, therefore, open to all interested members of the relevant communities, including (but not restricted to) planetary scientists, biologists, and terrestrial ice scientists.

Europa has been identified by NASA and the National Academy of Sciences as a high priority for exploration. This priority is established largely because Europa appears to be one of the few objects in our Solar System having conditions favorable for life. Europa is a rocky object slightly smaller than Earth's moon. It has an outer shell of water composition, modeled to be ~150 km thick, the surface of which is frozen. Several lines of evidence suggest that liquid water existed below the ice crust in the recent geologic past, and that liquid water might be present today. Friction generated by tides within Europa through its interaction with Jupiter and neighboring satellites might generate sufficient heat to fuel silicate volcanism at the base of the water layer. These factors, coupled with remote sensing observations of various salts on the surface and the likely implantation of organic compounds through cometary impact lead to the presence of the essential ingredients for exobiology; an energy source, water (possibly liquid), and organic chemistry. The Europa Focus Group provides a forum to bring the relevant interests together to exchange information and share ideas regarding Europa, including plans for its future exploration.

Highlights

- An organizing workshop was held at NASA Ames Research Center in early February, 2001, involving about 65 participants, including invitees who have not previously been involved with planetary science. The workshop was organized around the following topics: 1) Europa and terrestrial analogs (sea-ice, etc.), 2) Aqueous (liquid and ice) organic chemistry relevant to Europa, 3) Strategies in the search for life at Europa, and 4) Future exploration: needs, priorities.

- The EFG also met as a special breakout session during the Year 2001 General Meeting of the NAI, held in Washington, D.C.

NASA Mission Involvement

The results of this Focus Group are likely to influence the Europa Orbitor mission, currently scheduled for 2008; and potentially follow-on missions including landers. Although the Galileo mission is now in the final stages of operation, the Europa Focus Group also relates directly to this active mission.

Future Directions

The next EFG workshop is scheduled for September, 2001, at the U.S. Geological Survey Astrogeology facility in Flagstaff, Arizona. This workshop will provide the opportunity for participants to share their new research findings through one or two focused questions, and to provide a venue to foster collaborations.

Back to Table of Contents

EvoGenomics

Project

Focus Group Chairs:
B. Hedges and J. Lake

Focus Group Members:

Steven A. Benner
University of Florida

Jack D. Farmer
Arizona State University

James R. Garey
University of South Florida

J. Peter Gogarten
University of Connecticut

S. Blair Hedges
Pennsylvania State University

Christopher H. House
Pennsylvania State University

David K. Jacobs
University of California Los Angeles

Joseph L. Kirschvink
California Institute of Technology

Sudir Kumar
Arizona State University

James Lake
University of California Los Angeles

Charles Marshall
Harvard University

Monica Riley
Marine Biological Laboratory

Lynn J. Rothschild
Ames Research Center

Bruce N. Runnegar
University of California Los Angeles

Mitchell L. Sogin
Marine Biological Laboratory

Roadmap Objectives

#2
Origin of Life's Cellular Components

#3
Models for Life

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#7
Extremes of Life

#8
Past and Present Life on Mars

#12
Effects of Climate & Geology on Habitability

#13
Extrasolar Biomarkers

#14
Ecosystem Response to Rapid Environmental Change

Accomplishments

The purpose of the Evolutionary Genomics (EvoGenomics) Focus Group is to coordinate, combine, and enhance research efforts involving evolutionary genomics across the multiple disciplines and institutions represented in the astrobiology community. Evolutionary analysis of the complete genomes of organisms has greatly advanced our understanding of how life originated, adapted to diverse environments, and increased in complexity on this planet; and in turn, these studies will lead to a better understanding of life elsewhere in the Universe. The EvoGenomics Focus Group is a unique collaboration of astrobiologists combining expertise in molecular evolutionary analysis, organic chemistry and biochemistry, earth history, and paleontology. Our unifying goal is to compare the early evolutionary history of life, as revealed through analyses of genomic sequence data, with changes in Earth's environment through time, providing the basis to identify biomarkers for habitable planets.

During the past year the EvoGenomics Focus Group has engaged in a wide variety of activities including: collaborative research, an EvoGenomics Workshop held at UCLA (March 9-11, 2001), a breakout session at the Year 2001 NAI General Meeting, videoconferences, and the development of an EvoGenomics Focus Group website (www.evogenomics.org).

A focal point of much of the discussions during the videoconferences and workshop concerned research in the area of Neoproterozoic evolution and the impact of global glaciations. Benner (University of Florida) and Riley (MBL) have also made significant progress in their collaborative studies of the evolution of gene function. This first year of interaction as a group has been rewarding and has shown that complete collaboration should not be expected but rather a balance of independent and team research seems to be desirable. There will be joint publications resulting from inter-team collaborations, but the success of this Focus Group will be in bringing together researchers with diverse views so they can be heard and discussed. Thus, the research publications of individuals in this Focus Group, even if not co-authored with members of other teams, have directly benefited by Focus Group activities.

Highlights

- Organized productive workshop on Neoproterozoic/animal evolution (March 9-11, 2001).

- The discovery of the early colonization of land by eukaryotes and possible impact on global environment in the Proterozoic. (Heckman, D. S., D. M. Geiser, B. R. Eidell, R. L. Stauffer, N. L. Kardos, and S. B. Hedges. 2001. Molecular evidence for the early colonization of land by fungi and plants. *Science* 293:1129-1133)

NASA Mission Involvement

The inter-team research on evolutionary genomics conducted by this NAI Focus Group will directly or indirectly affect many of the near-term NASA space missions related to astrobiology; including those to Mars and Europa, as well as the Terrestrial Planet Finder. This is because the detection of life elsewhere relies on biomarkers, the interpretation of which is based on the evolution and characteristics of life on Earth. The results of the EvoGenomics Focus Group can be applied to the identification and refinement of these biomarkers by clarifying the relationship between biotic and environmental evolution. An additional, mission–related aspect of this work is studying the persistence of life (inferred through evolutionary analyses) during environmental extremes in the past history of the Earth, such as the global glaciations of the Neoproterozoic.

Future Directions

• The EvoGenomics Focus Group plans to further discussions of the Animal Origins project, and to seek funding for this project.

• Plans are also underway for a workshop to take place at the Penn State Lead Team Institution.

• There will be continued travel and additional collaboration between members of the individual teams; e.g., Benner-Riley, Marshall-Hedges, Lake-Riley.

• It is anticipated that evolutionary genomics will be one of the themes at the Astrobiology Science Conference II meeting to be held in April of 2002.

Back to Table of Contents

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

#16
Bringing Life with Us Beyond Earth

#17
Planetary Protection

Project

Mars

Focus Group Chair:
J. Farmer

Focus Group Members:

C. Alexander
Carnegie Institution of Washington

Brian Beard
University of Wisconsin

Steve Benner
University of Florida

Bob Blakenship
Arizona State University

Baruch Blumberg
NAI Central

Gary Coulter
Colorado State University

Wanda Lorraine Davis
SETI

David Des Marais
Ames Research Center

Eilleen Dunn
Arizona State University

Tom Foltz
Arizona State University

A. Gimenez
Centro de Astrobiologia

Julia Goreva
Arizona State University

Ronald Greeley
Arizona State University

John Holloway
Arizona State University

Clark Johnson
University of Wisconsin

Andrew Knoll
Harvard University

Dana Kovaric
UCLA

Robin Lloyd
Space.com

Isela Maldonado
Arizona State University

David McKay
Johnson Space Center

Mark Messerli
Marine Biological Laboratory

Michelle Minitti
Arizona State University

Paul Niles
Arizona State University

Sandra Pizzrello
Arizona State University

Jeff Plescia
USGS

Jim Rice
Arizona State University

Valeria Routt
Arizona State University

Steve Ruff
Arizona State University

Tom Sharp
Arizona State University

Leigh Anne Smith
UCLA

Mitchell Sogin
Marine Biological Laboratory

Sean Solomon
Carnegie Institution of Washington

Valeria Souza
UNAM Ecology

Christopher Staples
Arizona State University

Mike Tice
Stanford University

David Wynn-Williams
British Antarctic Survey

Veronia Ann Zabala
Arizona State University

Accomplishments

The Mars Focus Group was chartered to provide a forum within the NAI for discussing the scientific goals, objectives and measurement requirements for ongoing and future Mars missions involved with the exploration for past or present martian life, and/or pre-biotic chemistry. The products of this Focus Group are basic science recommendations and advice on implementation of astrobiology missions provided to mission planning groups.

During the past year the primary aim of the Mars Focus Group (MFG) has been to develop landing site recommendations for presentation to mission planners for the 2003 landed mission. Two NAI-wide videocons were presented in January (1-8 & 1-16, 2001) which: 1) reviewed the 2003 mission architecture, science payload, and engineering/landing site constraints, 2) discussed potential landing sites with a high priority for astrobiology and which could meet first order engineering constraints, and 3) sought a consensus recommendation from the NAI regarding science and site priorities for the 2003 mission. NAI Mars Focus Group discussions were made available to the entire Institute, but also included targeted invitees from outside of the NAI to broaden its base of expertise. Site reviews were presented by members of the ASU team (Farmer, Greeley, Hamilton, Nelson), and two members (Cabrol and Gulick) outside of the NAI who represented the Center for Mars Exploration (CMEX) at the NASA Ames Research Center. Presentations were followed by open discussions organized around several high-level questions that dealt with science and mission priorities for the 2003 mission. Presentation materials and a summary of the discussion were archived on the ASU Astrobiology and CMEX web sites. Results of the landing site discussions were presented by the MFG Chair at a landing sites workshop held at NASA Ames at the end of January, 2001. Based on inputs from that Workshop, the 2003 Landing Sites Steering Committee headed by John Grant of NASA Headquarters developed a shortlist of approximately 10 sites (from ~40 presented by the community at the workshop) for additional engineering studies and targeted high resolution imaging by the Mars Orbiter Camera now in Mars orbit. About half of the sites short-listed were on the NAI MFG list of recommendations.

The Mars Focus Group also organized a break-out session in conjunction with the Year 2001 Meeting of the NAI held at the Carnegie Institution of Washington in April. The group met, under standing-room only conditions, to review the results of the landing site workshop and to discuss future directions. One new goal identified was to broaden the scope of the group's present activities, which are primarily directed towards the development of future space missions, to include a basic research component. Several potential directions for basic research initiatives were proposed at the meeting, including the areas of life detection and technology development for astrobiology.

Highlights

- The NAI Mars Focus Group presented recommendations for science and landing site priorities at the 2003 Landing Sites Workshop. Among the ten sites short-listed by the Landing Sites Steering Committee for 2003, more than half had been identified as high priority sites for astrobiology by the MFG.

- Among the highest rated landing sites now short-listed for the 2003 mission is the so-called hematite site at Terra Meridiani. This site has been given high priority by the NAI MFG because of the potential for sampling aqueous sedimentary deposits of importance in the search for fossil biosignatures.

- On the basis of recommendations presented by the NAI MFG, the southern latitudinal limit for the 2003 mission has been extended several degrees to accommodate a site in Gusev Crater identified as having a high priority for astrobiology research.

NASA Mission Involvement

Our primary goal has been to provide science recommendations to help in the planning of Mars missions that will explore for past or present life, and/or prebiotic chemistry. This past year our efforts were focused on developing recommendations for landing site selection and science priorities for the 2003 mission.

Future Directions

Near-term plans of the NAI Mars Focus Group include efforts to review the existing plans for 1) the 2005 Mars mission, including the recommendations of the ‘05 Science Definition Team (which included several NAI members), 2) the summary of recent Mars Exploration Payload Advisory (MEPAG) activities, and 3) the upcoming instrumentation workshop. This group is also exploring opportunities to develop a summer institute focused on Mars Astrobiology. This new program would be aimed primarily at education and cross-training to develop a shared experience base among individual NAI investigators and students to enhance interactions between MFG members from different disciplines and to strengthen overall NAI participation in missions.

Back to Table of Contents

Mission to Early Earth

Project ▶ Roadmap Objectives

Focus Group Chairs:
A. Anbar, S. Mojzsis, R. Buick

Carlton C. Allen
Johnson Space Center

Jan Amend
Washington University

Ricardo Amils
Centro de Astrobiologia

Ariel D. Anbar
University of Rochester

G. Arnold
University of Rochester

Huiming Bao
University of California San Diego

Ekaterina Bazilevskaya
Pennsylvania State University

Brian L. Beard
University of Wisconsin Madison

Carrine Blank
University of California Berkeley

Baruch S. Blumberg
NASA Astrobiology Institute

Henry Bortman
NASA Astrobiology Institute

Tanja Bosak
Cal Poly State University

Arthur Bowman
Hampton University

Sam Bowring
Massachusetts Institute of Technology

Susan Brantley
Pennsylvania State University

Roger Buick
University of Washington

Sherry L. Cady
Ames Research Center

Rosemary Capo
University of Pittsburgh

David Catling
Ames Research Center

Deneyse J. Churchill
Pennsylvania State University

Carol E. Cleland
University of Colorado Boulder

Albert Colman
Yale University

Andrew D. Czaja
University of California Los Angeles

Steven D'Hondt
University of Rhode Island

Tzanko Doukov
Pennsylvania State University

Jennifer L. Eigenbrode
Pennsylvania State University

James J. Elser
Arizona State University

Douglas H. Erwin
Smithsonian Institution

William Fagan
Arizona State University

Jack D. Farmer
Arizona State University

James Farquhar
University of California San Diego

David Fernández Remolar
Centro de Astrobiologia

Eric Gaidos
University of Hawaii

Nathalie Grassineau
RHUL (UK)

David Grinspoon
University of Colorado Boulder

Rose A. Grymes
NASA Astrobiology Institute

T. Mark M. Harrison
University of California Los Angeles

Robert M. Hazen
Carnegie Institution of Washington

Tracey Herrera
University of California Los Angeles

Kai-Uwe Hinrichs
Woods Hole Oceanographic Institute

Andrew N. Hock
University of California Los Angeles

Christopher House
Pennsylvania State University

Munir Humayun
University of Chicago

Bruce M. Jakosky
University of Colorado Boulder

Charles C. James
Carnegie Institution of Washington

Emmanuelle Javaux
Harvard University

Clark M. Johnson
University of Wisconsin Madison

Alan J. Kaufman
University of Maryland

Roadmap Objectives

#5
Linking Planetary & Biological Evolution

#8
Past and Present Life on Mars

#16
Bringing Life with Us Beyond Earth

#17
Planetary Protection

Thomas Kieft
New Mexico Institute of Mining and Technology

Joseph L. Kirschvink
California Institute of Technology

Paul Knauth
Arizona State University

Andrew H. Knoll
Harvard University

Lee R. Kump
Pennsylvania State University

Donald R. Lowe
Stanford University

James Lyons
University of California Los Angeles

Timothy Lyons
University of Missouri Columbia

Craig Manning
University of California Los Angeles

Mary McHale
Johnson Space Center

Lucas J. Mix
Harvard University

Steve Mojzsis
University of Colorado Boulder

Penny A. Morris-Smith
University of Houston

David Morrison
NASA Astrobiology Institute

Brett Nielan
U. New S. Whales

Euan Nisbet
RHUL (UK)

Nora Noffke
Harvard University

Hiroshi Ohmoto
Pennsylvania State University

Shuhei Ono
Pennsylvania State University

Victor Parro Garcia
Centro de Astrobiologia

Alexander Pavlov
Pennsylvania State University

Juan Perez Mercader
Centro de Astrobiologia

Randall Perry
University of Washington

Robert Pidgeon
Curtain University

Jeffrey Plescia
USGS Menlo Park

Susannah Porter
Harvard University

Luke Probst
Bemidji State University

Nina D. Rosenberg
Lawrence Livermore National Laboratory

Bruce N. Runnegar
University of California Los Angeles

Robert Rye
California Institute of Technology

William Schopf
University of California Los Angeles

Rusty Schweickart
California Academy of Sciences

Mukul Sharma
Dartmouth College

Yanan Shen
Harvard University

Norm Sleep
Stanford University

Alexei Smirnov
University of Rochester

David W. Smith
Massachusetts Institute of Technology

Leigh Ann Smith
University of California Los Angeles

Sean C. Solomon
Carnegie Institution of Washington

Sherry Stafford
University of Pittsburgh

Brian W. Stewart
University of Pittsburgh

Roger E. Summons
Massachusetts Institute of Technology

Carol Tang
California Academy of Sciences

John Tarduno
University of Rochester

Mike Tice
Stanford University

Akiko Tomitani
Harvard University

Allan H. Treiman
Lunar And Planetary Institute

Luis Vasquez
Centro de Astrobiologia

Malcolm Walter
Macquarie University

Benjamin Weiss
California Institute of Technology

Llyd E. Wells
University of Washington

Susan J. Wentworth
Lockheed Martin

Frances Westall
Lunar And Planetary Institute

George W. Wetherill
Carnegie Institution of Washington

Kosei E. Yamaguchi
Pennsylvania State University

Thomas Yulsman
University of Colorado Boulder

Kevin Zahnle
Ames Research Center

Accomplishments

The rational search for life beyond Earth requires some concept of the conditions under which life originates and begins to evolve, and of the environmental "fingerprints" of primitive biospheres. This concept must be informed by a solid understanding of the only planet on which life is known to exist - the Earth. Hence, study of life and the environment on the early Earth is a critical component in developing mission plans for astrobiology space missions. This is the underlying rationale of the Mission to Early Earth (MtEE) Focus Group.

The geologic record is increasingly sparse as one examines the condition of the early Earth further back in time. Furthermore, the quality of material easily available is not high - particularly when interest in environmental and biological history leads to a focus on biogeochemical signatures that are not robust against oxidation and other alteration processes at the Earth's surface. From discussions early in the formation of the MtEE Focus Group, a consensus emerged that progress in this new area of "bio-environmental reconstruction" is fundamentally sample limited. This consensus in the community led directly to the concept that the MtEE Focus Group might be most useful as a means to promote, provide justification for, and identify support for the acquisition and distribution of samples from the Precambrian, with a special focus on deep drilling to acquire pristine sediment samples from the near subsurface (< 1000 m). This activity has the potential to stimulate participation and collaboration from across the Institute, one of the benefits of the NAI Focus Groups as originally conceived by the Director.

The first MtEE FG meeting was held in conjunction with the Year 2000 AGU conference, and the second meeting was held as a breakout session at the Year 2001 General Meeting of the NAI, where approximately 75 participants attended.

Highlights

- The Mission to Early Earth Focus Group sees the study of ancient life and the early environment of Earth as critical in assessing the potential for life beyond Earth.

- The study of ancient life and the early-Earth environment is regarded by the Mission to Early Earth Focus Group as being sample-limited. While it is relatively easy to find ancient rocks, it is difficult to find ancient materials that are well-enough preserved to provide the desired information. It is also difficult to find materials that provide information about the Earth near the time of life's emergence. Such materials exist, but are scarce or exist only below the surface.

- The Mission to Early Earth Focus Group recommends initiation of subsurface drilling projects to acquire well-preserved geological materials with information about ancient life and environmental conditions.

- With the support of the NAI, the Mission to Early Earth Focus Group has completed a successful excursion to W. Australia during the Summer of 2001 to develop plans for such a pilot drilling project.

Field Trips

With support from the NAI, the first major activity of the MtEE Focus Group has been to organize an Australian field excursion for the summer of 2001 to Western Australia. This general area of Western Australia is one of the few places on Earth with well-preserved sediments (including biosignatures) from the Archean, i.e., the period before 2.5 billion years ago - the first half of Earth history. As a result, a number of exciting, astrobiologically-relevant publications have emerged recently as a result of research into this region. One project organized by the MtEE Focus Group traveled to the Jack Hills area under the guidance of Steve Mojzsis (University of Colorado Team). The primary goal of this effort was to become familiar with the local stratigraphy and astrobiologically-relevant localities in order to develop concepts for a pilot astrobiology drilling project. A second team visited the Pilbara, under the guidance of Roger Buick (University of Washington Team). The primary goal of this group was to obtain samples from the earliest geologic record. Both of these excursions were scheduled to begin after the NAI-sponsored Earth Systems conference in Edinburgh in late June, 2001 and to flow into the Astrobiology Workshop at Macquarie University scheduled for mid-July, 2001. The latter meeting was sponsored by the Australian Centre for Astrobiology, an NAI Affiliate Member, and coordination for all of these events was possible by collaboration with Malcolm Walter at Macquarie University.

NASA Mission Involvement

The rational search for life beyond Earth requires some concept of the conditions under which life originates and begins to evolve, and of the environmental "fingerprints" of primitive biospheres. This concept must be informed by a solid understanding of the only planet on which life is known to exist - the Earth. Hence, study of early life and the environment on the early Earth is a critical component in developing mission plans for astrobiology space missions. This is the underlying rationale of the Mission to Early Earth Focus Group. The projects under development by the MtEE Focus Group should, in particular, provide information useful to development of the Terrestrial Planet Finder mission, by elucidating the history of oxygenic photosynthesis and the oxygenation of the atmosphere.

Future Directions

- Future plans for the MtEE FG include the development of a pilot drilling project concept, informed by the Summer 2001 Australian excursion, to present at the Fall Year 2001 GSA or AGU meetings.

- A second MtEE field excursion is also being planned for the Summer of 2002, possibly in North America to minimize costs and maximize participation.

Back to Table of Contents

Mixed Microbial EcoGenomics

Project

Roadmap Objectives

#4
Genomic Clues to Evolution

#5
Linking Planetary & Biological Evolution

#6
Microbial Ecology

#7
Extremes of Life

#10
Natural Migration of Life

#11
Origin of Habitable Planets

#12
Effects of Climate & Geology on Habitability

#14
Ecosystem Response to Rapid Environmental Change

Focus Group Chairs:
M. Sogin and D. Des Marais

Brad M. Bebout
Ames Research Center

Michael P. Cummings
Marine Biological Laboratory

David Des Marais
Ames Research Center

Jesse Dillon
University of Washington

Virginia P. Edgcomb
Marine Biological Laboratory

Jack D. Farmer
Arizona State University

R. Feldman
Molecular Dynamics

Ferran Garcia-Pichel
Arizona State University

Tori M. Hoehler
Ames Research Center

Linda L. Jahnke
Ames Research Center

Richard M. Keller
Ames Research Center

Andrew McArthur
Marine Biological Laboratory

Scott Miller
Ames Research Center

H. Morrison
Marine Biological Laboratory

Norman R. Pace
University of Colorado Boulder

C. Palacios
Marine Biological Laboratory

Chris Potter
Ames Research Center

Mitchell L. Sogin
Marine Biological Laboratory

John Spear
University of Colorado Boulder

David Stahl
University of Washington

Andreas P. Teske
Woods Hole Oceanographic Institute

Pieter T. Visscher
University of Connecticut

Accomplishments

The over-arching objective of the Mixed Microbial EcoGenomics (EcoGenomics) Focus Group is to define the relationship between microbial diversity, complex gene expression patterns, and biogeochemical processes that shape planetary environments. Using the hypersaline cyanobacterial mats of Guerrero Negro, Baja California, Mexico as a field site, this inter-team effort is characterizing biogeochemical patterns in the microbial mats with special emphasis on gradient location and shape. These measurements are being coupled with molecular-based assessments of microbial population structures and DNA micro-array measurements of gene expression. The gene expression data will identify important biochemical activities that play key roles in formation and shifts in biogeochemical gradients in response to transient and periodic perturbations imposed by diel cycles. The ultimate objective is to establish models that can predict the behavior of these complex systems.

"Where did we come from, how did we get here, and where are we going?" embodies the principle objectives of astrobiological research. Answers to the first question are rooted within the microbial world, which represented the only form of life during the initial three billion years of our evolutionary history. Ever since the origin of life, complex interactions of microorganisms with each other and with all other components of the biosphere have dominated the course of evolution in our biosphere. Even the earliest biogenic fossils (microbial mats) display integrally layered patterns of organization. This spatial structuring likely reflects functional interdependence of different microbes in early communities. Microbial creatures of untold diversity continue to dominate every corner of our biosphere, and they are likely to be the only life forms that might be encountered in other parts of our solar system, if not the entire cosmos. Yet, there is only sparse information about the true diversity of microorganisms, including their capability to orchestrate and drive key biogeochemical cycles that shape our ever-changing planet. A more complete understanding of microbial diversity, descriptions of ecosystem-wide patterns of gene expression, and detailed analyses of biogeochemistry would provide a new foundation for interpreting paleontological and geological studies that describe Earth's early history.

The NASA Astrobiology Institute (NAI) is uniquely positioned to initiate an interdisciplinary project to address these questions. With modern technology developed by the genome community, is is now possible to assess microbial diversity and the total genetic coding capacity of any particular environment. This would be based on phylogenetic surveys of ribosomal RNAs and high-throughput DNA sequencing. The EcoGenomics Focus Group has formulated a bold strategy for linking microbial gene expression patterns with particular metabolic activities that underlie central biogeochemical processes. Community DNA extracted from natural microbial populations of a selected site will be treated as a complex mixed genome. Members of the microbial community will be surveyed by analysis of rRNAs, and descriptions of potential metabolic diversity will be inferred from database analyses of several hundred thousand randomly selected DNA sequences. This large database of DNA sequences from the "mixed environmental genome" will be used to design DNA microarrays that can detect mRNA transcription patterns. With this mixed environmental genome array technology (MEGAT), tens of thousands of distinct genes of known sequence and function can be efficiently monitored. MEGAT will define changes in gene expression from a large number of environmental samples with known spatial and temporal distribution patterns in a well-structured microbial community, e.g., microbial mats or stromatolites. Results of these studies will be correlated with detailed measurements of biogeochemical gradients throughout the studied environment. The initial objective is to define biological complexity within the studied microbial system. The ultimate objective is to model how coordinated gene expression patterns observed in microbial consortia shape the environment.

Highlights

- The NAI EcoGenomics Focus Group has initiated a comprehensive sampling of biogeochemical processes and microbial diversity in the hypersaline mats of the Guerrero Negro.

- The W.M. Keck Foundation awarded funds to the MBL for the construction of an advanced laboratory of ecological and evolutionary genomics.

Field Trips

EcoGenomics Focus Group meetings were held in November, 2000 and April 2001 to plan for a major field excursion to take place during the Summer of 2001. Five NAI research teams, as well as colleagues from other institutions, took part in this EcoGenomics FG-sponsored field trip to the hypersaline microbial mats at Exportadora de Sal, S. A. de C.V. Guerrero Negro, BCS, Baja California, Mexico June 2-11, 2001. Participants in the field trip included David Des Marais, Tori M. Hoehler, Scott Miller, and Brad Bebout, Ames Research Center; Kendra Turk, University of California at Santa Cruz; Steven Carpenter, Orbital Corporation; Jesse Dillon and David Stahl, University of Washington; Feran Garcia-Pichel and Jack Farmer, Arizona State University; Norman Pace, John Spear, and Ruth Ley, University of Colorado; Pieter Visscher, University of Connecticut; R. Castenholz, University of Oregon, Miguel Angel Huerta-Diaz, Instituto de Investigaciones Oceanológicas, Universidad Autónoma de Baja California, Baja California, Mexico; Bo Thamdrup, Odense University, Denmark.

Five primary goals were defined for this field trip: 1) establishing the microbial mat carbon and oxygen budgets, 2) characterizing populations of sulfate reducing bacteria, 3) measuring the lateral distributions of cyanobacterial populations within subtidal microbial mats, 4) conducting total microbial diversity surveys throughout the mat, and 5) characterizing the biogeochemistry of sulfur in photosynthetic microbial mats.

During the field trip, the Ames team lead by Dave Des Marais examined exchange of carbon and oxygen across the interface between the microbial mats and the overlying water column over a full 24-hour day-night cycle. They obtained depth profiles of oxygen (O_2) at several locations in order to assess the uniformity of distribution of photosynthesis. The University of Washington team lead by David Stahl acquired a series of samples in association with the diel experiment in order to identify and determine the distribution of various species of sulfate reducing bacteria. These results are to be correlated with population studies of cyanobacteria that were conducted by Feran Garcia-Pichel of Arizona State University. More than 100 core samples of mat were acquired, as grids of replicate samples ranging in size from centimeters to kilometers. Norm Pace's group from the University of Colorado collected samples for sequence analyses of ribosomal RNA genes. These data will provide a broad survey of phylogenetic diversity throughout the subtidal mat. Finally, Pieter Visscher from the University of Connecticut collected mat sediments in order to enrich for sulfate reducing and sulfide oxidizing bacteria through various enrichment techniques. Visscher also collected samples for characterizing volatile sulfur species (dimethylsulfide and dimethylsulfoxide) and sulfate reduction rates.

The MBL group (M. L. Sogin) in collaboration with F. Garcia-Pichel has initiated construction of genomic libraries from two cyanobacterial strains that represent major species in these hypersaline cyanobacterial mats. We will also construct libraries for two of the most prominent isolates of sulfate reducing bacteria as determined by the initial microbial population structure surveys described above. These libraries will be sequenced and used to construct DNA micro-arrays for monitoring changes in gene expression patterns during different stages of the diel cycle. The MBL was also successful in a proposal submitted by Sogin, Cummings and Wernegreen to the W.M. Keck Foundation. This foundation award provides resources to build and equip an advanced laboratory for high-throughput genomics and post-genomics related to evolutionary and ecological studies. This equipment will provide a full range of technical capabilities normally found only within industrialized genome centers. The NAI activities played an important role in the success of this Keck proposal.

NASA Mission Involvement

Although this work is not directly related to a specific NASA space mission, it does provide information that will be important in the search for extraterrestrial life. The field studies carried out by the EcoGenomics Focus Group will help us to understand the range of conditions that were present on early Earth. It is clear that the discovery of life on other solar system bodies would most likely be microbial, and the EcoGenomics Focus Group seeks to understand how microbial ecosystems affected the early atmosphere and the biological processes that left traces of early life in ancient sedimentary rocks. These studies will help in the design of life detection experiments and to interpret geological studies of samples returned to earth.

Future Directions

Each of the teams in the EcoGenomics Focus Group will use the field samples to carry out the molecular based analyses for characterizing population structures. These will be used to design the sampling strategies for subsequent field trips to the Guerro Negro, where we will carry out finer scale measurements of biogeochemical processes and microbial population structures. These results will also be correlated with changing patterns of gene expression during different stages of the diel cycle. For these experiments we will employ DNA microarray measurements based upon the major cyanobacterial and sulfate reducing bacterial isolates.

Back to Table of Contents

NAI Central Education & Public Outreach

The NAI Central Education and Public Outreach (EPO) Program works to coordinate and complement efforts by each of our Lead Teams in the area of EPO. For details on the EPO efforts of the Lead Teams, refer to the individual team reports.

NAI Central Education & Public Outreach

EPO Manager:
Krisstina Wilmoth

Karen Dodson
EPO Lead

Julie Fletcher
Graphic Design

Darlene Gadd
Materials and Support

John Han
Graphic Design Intern

Jennifer Kwong
Technical Writing

Jaron Ross
Science Writing Intern
(Term complete August 2001)

Daniella Scalice
Video Project Development

Kim Winges
Science Writing Intern
(Term complete August 2001)

Annual Science Report Year 2 (2000)

The NAI Central Outreach Team edited and produced the *NAI Annual Science Report Year 2*. Two thousand five hundred CD-ROMs were distributed to members, relevant science committees, other scientists, and interested individuals internal and external to NASA. A pdf of the document was also posted on the NAI website for download.

Ask An Astrobiologist

Residing within the NAI website, the *Ask An Astrobiologist* site serves the student community and the public by providing a forum for their questions about astrobiology. This year, the site underwent a major revision to present five categories of previously asked questions and answers for users to look through and learn from, with space provided for users to ask new questions. In addition to the NAI Central writing staff, each quarter a new NAI Lead Team is featured as the astrobiology experts, and new questions are sent directly to them for answers. All responses are posted on the website in the appropriate categories and users are notified when the answers are posted and given information on how to access the them. The *Ask An Astrobiologist* site also features questions with longer, more in-depth answers and video supplements and has special sections that address the two most frequently asked questions: “What is astrobiology?” and “How can I become an astrobiologist?” *Ask An Astrobiologist* is available on the NAI website at http://nai.arc.nasa.gov

Astro-Venture

Astro-Venture is an interactive educational, multimedia web environment highlighting NASA careers and astrobiology research in the areas of astronomy, geology, biology and atmospheric sciences. Students in grades 5-8 are transported to the future where they role-play NASA occupations and using scientific inquiry, search for and build a planet with the necessary characteristics for human habitation. Experts in multi-media pedagogy and middle school education have utilized a two-step project where modules are field-tested and revised prior to release on the website. Supporting activities include chats with NASA scientists, online collaborations, classroom lessons, a student publishing area and occupation fact sheets and trading cards. NAI Central EPO provided funding, science expertise and content for the development of the astronomy module in year 3. The project is available at http://astroventure.arc.nasa.gov

Astrobiology Pathfinder

The *Astrobiology Pathfinder* is being designed to answer the question, "How can I become an astrobiologist?" A survey of astrobiology-related courses offered at universities throughout the country is under development and will be provided to users in a searchable database that includes listings of internships and science programs at local venues. Research scientist biographies and interview video clips will also be available as part of *Astrobiology Pathfinder*. The project can be found under the *Ask an Astrobiologist* section of the NAI website: http://nai.arc.nasa.gov

Astrobiology Workshop

NAI Education and Public Outreach Manager, Krisstina Wilmoth, participated in the “Astrobiology Workshop” July 12-13, 2001 at Macquarie University in Sydney, Australia to present “Education and public outreach: the next generation of astrobiologists.” The workshop was the first meeting of the Australian Centre for Astrobiology and offered an opportunity to discuss education and outreach partnerships. Information regarding the NAI Lead Teams’ research, the *NAI Annual Science Report Year 2*, and the Focus Groups was disseminated to the 100 attendees. Information regarding the meeting is available at
http://www.aao.gov.au/local/www/jab/workshop.html

Educational and Informational Products

NAI Central EPO develops educational products for use by the entire NAI community. During 2001, 160,000 planetary cards highlighting 8 different planets and their environments and advertising the NAI website were produced along with 25,000 bookmarks listing astrobiology readings and the NAI url for distribution at teacher conferences, educational centers, science museums, and to each of the NAI EPOs for their communities. Copies of the *Educator Resource Guide: Astrobiology in Your Classroom* are also available in pdf and hard copy formats. Posters detailing the Astro-Venture website and general information regarding NAI’s Lead Teams, research goals, Focus Groups, Affiliate and Associate Members, and NRC Postdoctoral Fellows Program are also available upon request. Contact the NAI Central EPO office for more information.

Exhibits and Conferences

NAI Central EPO supports, presents at, and supplies materials to several different educational, scientific, and general interest conferences and meetings throughout the year. In Year 3, NAI Central supported five major events.

Earth System Processes: A Global Meeting

The Geological Society of London and the Geological Society of America held a joint meeting in Edinburgh, Scotland, June 24-28, 2001. This meeting brought together scientists from all over the world to discuss the present state of knowledge of earth system processes, both the linkages between systems and how these systems have evolved through time, in order to advance our understanding of planetary processes. In addition to NAI's sponsorhip of two scientific sessions, NAI Central EPO sent a 10 foot exhibit and materials including the Annual Science Report Year 2, as well as information on our Focus Groups, international partnerships, and NRC Postdoctoral Fellows Program. Two staff members interacted with the 100 attendees and assisted the speakers in the NAI-supported sessions as necessary. Information on the meeting is available at:
http://www.geolsoc.org.uk/template.cfm?name=gsa_edinburgh

Moffett Field Airshow

NAI Cental EPO provided and staffed 10 foot exhibit at the Moffett Field Airshow in California. Astrobiology materials including a bookmark detailing general astrobiology suggested readings and planet cards highlighting the NAI website were distributed to the attendees. Staff members interacted with the audience and answered their questions about Astrobiology. Seventy thousand people attended the event.

National Association of Biology Teachers (NABT) Annual Meeting

Two astrobiology sessions and workshops were presented at the NABT National Convention in Orlando, Florida in October 2000. A 10 foot exhibit was displayed and educational materials including 500 CD-ROMs with the *Educator Resource Guide* and the PSU astrobiology publication were disseminated.

National Council of Teachers of Mathematics (NCTM) Annual Meeting

NAI Central EPO Manager, Krisstina Wilmoth, and EPO Lead, Karen Dodson attended NCTM annual meeting in April 2001 as a first step in the development of astrobiology math curriculum. One thousand one hundred sixtry CD-ROMs were distributed along with materials from other NASA Office of Space Sciences Origins. Attendance at NCTM fostered collaborations with other NASA educational groups and with curriculum developers.

National Science Teacher's Association Annual Convention (NSTA)

Four astrobiology-related sessions and one workshop were given by NAI members to teachers attending NSTA in April 2001. NSTA is one of the largest gatherings of teachers interested in the sciences. The 2001 national convention in St. Louis, Missouri drew 19,000 teachers, administrators, and others interested in science education. The convention consisted of four days of workshops and exhibits offering the opportunity for NAI Education and Outreach members to have direct contact with one of our largest audiences. In addition to participating in the NASA Office of Space Sciences Origins booth, NAI Central EPO developed and disseminated 1500 CD-ROMs of Origins materials including astrobiology and NAI Lead Team contributions. Trading cards advertising *Astro-Venture* and 400 *Educator Resource Guides* were also distributed.

Indiana School for the Deaf presentation

Twelve students from the Indiana School for the Deaf visited Ames Research Center in June 2001. NAI EPO Manager, Krisstina Wilmoth, presented astrobiology concepts to them and distributed our *Educator Resource Guide* activity packets for their use.

Mysteries of Fieldwork: Fascinating Microbes

The NAI Central EPO Team and the Quest Educational Group at Ames developed a series of classroom activities and webcasts about the exciting fieldwork involved in studying microbes. Teachers were provided with background material about microbes and given curriculum and hands-on science activities to perform in their classrooms. A series of webcasts featuring the NAI Ames Research Center Lead Team were produced to enhance the curriculum. In the fall of 2001, a three part series on preparing for fieldwork featuring Dr. Tori Hoehler, conducting fieldwork with Dr. David Des Marais, and processing the data with Dr. Brad Bebout and Mary Hogan offered students the opportunity to see the day-to-day work of our researchers and to understand the scientific method. During the webcasts, footage from Dr. Des Marais' fieldwork was used and students were able to interact live with the research team via an internet chat room. In addition, students were able to read biographies of the researchers and to study career fact sheets about them to gain a better understanding of the lives of astrobiologists. An archive of the project is available at http://quest.nasa.gov/projects/astrobiology/fieldwork/

NAI General Meeting - Video studio

A digital video studio was set up at NAI's General Meeting in April 2001. Attendees were invited to be interviewed on camera about the work they presented at the conference. Twenty-five people were interviewed in all, for a total of four hours of footage. The response was very positive and future plans are underway to re-create the video studio at upcoming conferences. The video interviews are being used on the NAI website in a variety of ways as well as in presentations for various audiences. One example featuring Dr. David Wynn-Williams of the UK Astrobiology Forum is available on the NAI website at http://nai.arc.nasa.gov

NAI Central Website

The NAI Central website contains three EPO sections: *Ask an Astrobiologist* (see entry), *For Students*, and *For Teachers*. The teacher section contains a resource catalog of astrobiology related materials, hands-on activities, and curricula to be used in the classroom and the laboratory. Both the Teacher and the Student sections provide background about astrobiology, in depth articles about the science of astrobiology, and educational products. *For Students* also highlights *Astrobiology Pathfinder* (see entry) and interactive projects on searching for and developing habitable worlds. See the website at: http://nai.arc.nasa.gov

NAI Education and Public Outreach Working Group Retreat

NAI EPO Leads were invited by NAI Central to participate in a working group retreat held at Johnson Space Center in February 2001. EPO leads from several teams met at JSC to discuss and plan activities and products for the upcoming year including ways to leverage efforts and activities for the 2001 NAI General Meeting. Attendees toured the JSC Astrobiology Facilities and were given presentations about the Team's research.

NASA Office of Space Science EPO Representation

NAI Central EPO represents the entire NAI EPO team to the NASA Office of Space Science at NASA Headquarters through participation in monthly teleconferences with the Origins Forum, participation in OSS EPO events such as exhibits at the National Science Teachers' Association (NSTA) annual meeting, and input to the NASA Office of Education reporting database known as EdCats. NAI Central EPO also produced a CD-ROM of material from several Origins missions, including specific products contributed by the NAI Lead Teams, for dissemination at NSTA in April 2001. Information regarding funding opportunities, relevant EPO products, and OSS policies and guidelines is distributed to the NAI Lead Team EPOs.

Space Biology Museum Network

NAI Central EPO partnered with NASA's Fundamental Biology Outreach Program and Microgravity Outreach Program to support a network of informal education and museum partners called the Space Biology Museum Network. The Network is comprised of 11 members, including eight domestic and two international museums. It focuses on educating the Network members in areas of NASA research related to space biology and best practices in museum exhibit development. In November 2001, a three day workshop was held at Johnson Space Center with presentations from both NASA and museum members. NAI Central EPO Manager, Krisstina Wilmoth, presented a general astrobiology tutorial as well as information regarding NAI's educational products and programs. As a result, special activities and events incorporating astrobiology topics were developed museum members and presented to their public audiences.

Toward Other Planetary Systems (TOPS)

This 5-year NSF-sponsored Teacher Enhancement program's goals are to initiate systemic reform in science education in Hawaii by enabling science and math teachers to implement astronomy in their classrooms. Teachers participate in an intensive 3-week summer workshop held in part on Oahu and in part on the big island of Hawaii at the Hawaii Preparatory Academy. Teachers learn basic astronomy content, participate in hands-on activities using exemplary materials, begin to integrate state and national science/astronomy standards into their classrooms methods and learn evaluation and assessment techniques. In addition, a privately funded student component of the program hosts local high school students with interests in astronomy. NAI Central EPO Manager, Krisstina Wilmoth, and EPO Lead, Karen Dodson, teamed with MBL Lead Team Member, John Stolz, to conduct a two-day astrobiology curriculum workshop at TOPS in June 2001. During this workshop, teachers were given an overview of astrobiology research, trained in using the *Educators Resource Guide* and *Astro-Venture*, and participated in hands-on microbiology lab activities. Each participant received a copy of the Guide along with other astrobiology materials. Information regarding TOPS may be found at: http://www.ifa.hawaii.edu/tops/

Voyages Through Time

The SETI Institute, with support from the National Science Foundation, the California Academy of Sciences, the NASA Astrobiology Institute, and San Francisco State University, is developing standards-based curriculum materials for a one-year high school integrated science course centered on the unifying theme of evolution and delivered on CD-ROM. Scientists, teachers, curriculum writers, and media specialists have created six modules that integrate astronomical, geological, and biological sciences. The sequence of lessons in each module is designed to promote students' understanding and skills as defined by the National Science Education Standards and Benchmarks for Science Literacy. The six modules, *Cosmic Evolution, Planetary Evolution, Origin of Life, Evolution of Life, Hominid Evolution,* and *Evolution of Technology*, use the constructivist approach of engage, explore, explain, elaborate, and evaluate (BSCS, 1996) as an instructional framework. NAI Central EPO has funded a CD-ROM sampler of the curriculum for distribution at various teacher conferences. More information regarding VTT can be found at: http://www.seti.org/education/vtt-bg.html

Back to Table of Contents

NAI/NRC Postdoctoral Fellows

In Year 2, the NAI initiated a competitive NAI Postdoctoral Fellowship Program administered by the National Research Council of the National Academy of Sciences and the National Academy of Engineering. In Year 3, six NAI Fellows where selected (August 2000) in the first application opportunity and an additional six where chosen (April 2001).

August 2000 Fellows

- **Eric Gaucher**

Title: Using Functional Genomics to Infer the Biology and Chemistry of the Last Common Ancestor
Advisor: Steven Benner, University of Florida
NAI Lead Team: Scripps Research Institute

- **Yanan Shen**

Title: Environmental Changes in the Context of Biological Evolution During Neoproterozoic on the Yangtze Platform: A Snowball Earth?
Advisor: Andrew Knoll
NAI Lead Team: Harvard University

- **Mark Messerli**

Title: Physiological Regulation of Cytosolic pH in a Eukaryotic Acidophile
Advisor: Mitchell Sogin
NAI Lead Team: The Marine Biological Laboratory

- **Marc Kramer**

Title: Linking Earth Science and Astrobiology: Surface Hydrology and Microbial Ecology for Global Semi-Arid Ecosystems
Advisor: David Peterson
NAI Lead Team: NASA Ames Research Center

- **Virginia Edgecomb**

Title: Hyperthermopohiles of the Hydrothermal Vent Subsurface: Limits of Life and Extraterrestrial Analogs
Advisor: Andreas Teske, Woods Hole Oceanographic Institute
NAI Lead Team: The Marine Biological Laboratory

- **David Warmflash**

Title: Immunoassay Life Detection Test
Advisor: David McKay
NAI Lead Team: NASA Johnson Space Center

April 2001 Fellows

- **Michelle Minitti**

Title: Chasing Water on Mars: A Geochemical Approach Utilizing Martian Meterorites
Advisor: Laurie Leshin
NAI Lead Team: Arizona State University

- **Alexander Pavlov**

Title: Organic and Sulfur Hazes in the Archean Atmosphere: Climate and Photochemical Consequences
Advisor: Owen Toon
NAI Lead Team: University of Colorado, Boulder

- **Charles Boyce**

Title: The Use of Living Plants and Fossil Chemistry to Study the Morphological Patterns and Developmental Processes of Land Plant Evolution
Advisor: Andrew Knoll
NAI Lead Team: Harvard University

- **Henry Scott**

Title: Stability of Organic Material in Icy Satellites
Advisor: Russell Hemley
NAI Lead Team: Carnegie Institution of Washington

- **Ruth Ley**

Title: Diversity and Function of the Sulfur-Cycling Community in Hypersaline Microbial Mats
Advisor: Norman Pace
NAI Lead Team: University of Colorado, Boulder

- **Sara-Eva Martinez-Alonso**

Title: Identification and Study of Hydrothermal Systems on Mars Through Remote Sensing
Advisor: Bruce Jakosky
NAI Lead Team: University of Colorado, Boulder

Back to Table of Contents

Appendices

NAI Director's Science Council

The NAI Director's Science Council consists of distinguished scientists, many of them Nobel Laureates, who advise the NAI Director. The Science Council is asked to assist the Institute by commenting on research directions of the NAI and related astrobiology issues.

Sidney Altman
Yale University

Philip W. Anderson
Princeton University

Sydney Brenner
Molecular Sciences Institute

Murray Gell-Mann
Santa Fe Institute

Ronald Greeley
Arizona State University

Robert B. Laughlin
Stanford University

Joshua Lederberg
The Rockefeller University

Elliott C. Levinthal
Stanford University

Richard J. Roberts
New England BioLabs

Anneila I. Sargent
California Institute of Technology

Maxine F. Singer
Carnegie Institution of Washington

Jonathan Lunine
University of Arizona, Tucson

Back to Table of Contents

Astrobiology Roadmap

The Roadmap provides guidance for research and technology development across several NASA Enterprises: Space Science, Earth Science, and the Human Exploration and Development of Space. The recommendations were formulated in terms of 3 basic questions and 17 specific science objectives, which have been translated into NASA programs and integrated with NASA strategic planning.

The Roadmap is the product of efforts by more than 400 scientists and technologists, spanning a broad range of disciplines and organizations. More than 100 of these participated in a 3-day Roadmapping Workshop held in July 1998 at NASA Ames Research Center, while others attended previous topical workshops.

How does life begin and evolve?

1. Sources of organics on Earth. Determine whether the atmosphere of the early Earth, hydrothermal systems or exogenous matter were significant sources of organic matter.

2. Origin of life's cellular components. Develop and test plausible pathways by which ancient counterparts of membrane systems, proteins and nucleic acids were synthesized from simpler precursors and assembled into protocells.

3. Models for life. Establish replicating, catalytic systems capable of evolution, and construct laboratory models of metabolism in primitive living systems.

4. Genomic clues to evolution. Expand and interpret the genomic database of a select group of key microorganisms in order to reveal the history and dynamics of evolution.

5. Linking planetary and biological evolution. Describe the sequences of causes and effects associated with the development of Earth's early biosphere and the global environment.

6. Microbial ecology. Define how ecophysiological processes structure microbial communities, influence their adaptation and evolution, and affect their detection on other planets.

Does life exist elsewhere in the Universe?

7. The extremes of life. Identify the environmental limits for life by examining biological adaptations to extremes in environmental conditions.

8. Past and present life on Mars. Search for evidence of ancient climates, extinct life and potential habitats for extant life on Mars.

9. Life's precursors and habitats in the outer solar system. Determine the presence of life's chemical precursors and potential habitats for life in the outer solar system.

10. Natural migration of life. Understand the natural processes by which life can migrate from one world to another.

11. Origin of habitable planets. Determine (theoretically and empirically) the ultimate outcome of the planet-forming process around other stars, especially as it relates to habitable planets.

12. Effects of climate and geology on habitability. Define climatological and geological effects upon the limits of habitable zones around the Sun and other stars to help define the frequency of habitable planets in the universe.

13. Extrasolar biomarkers. Define an array of astronomically detectable spectroscopic features that indicate habitable conditions and/or the presence of life on an extrasolar planet.

What is life's future on Earth and beyond?

14. Ecosystem response to rapid environmental change. Determine the resilience of local and global ecosystems through their response to natural and human-induced disturbances.

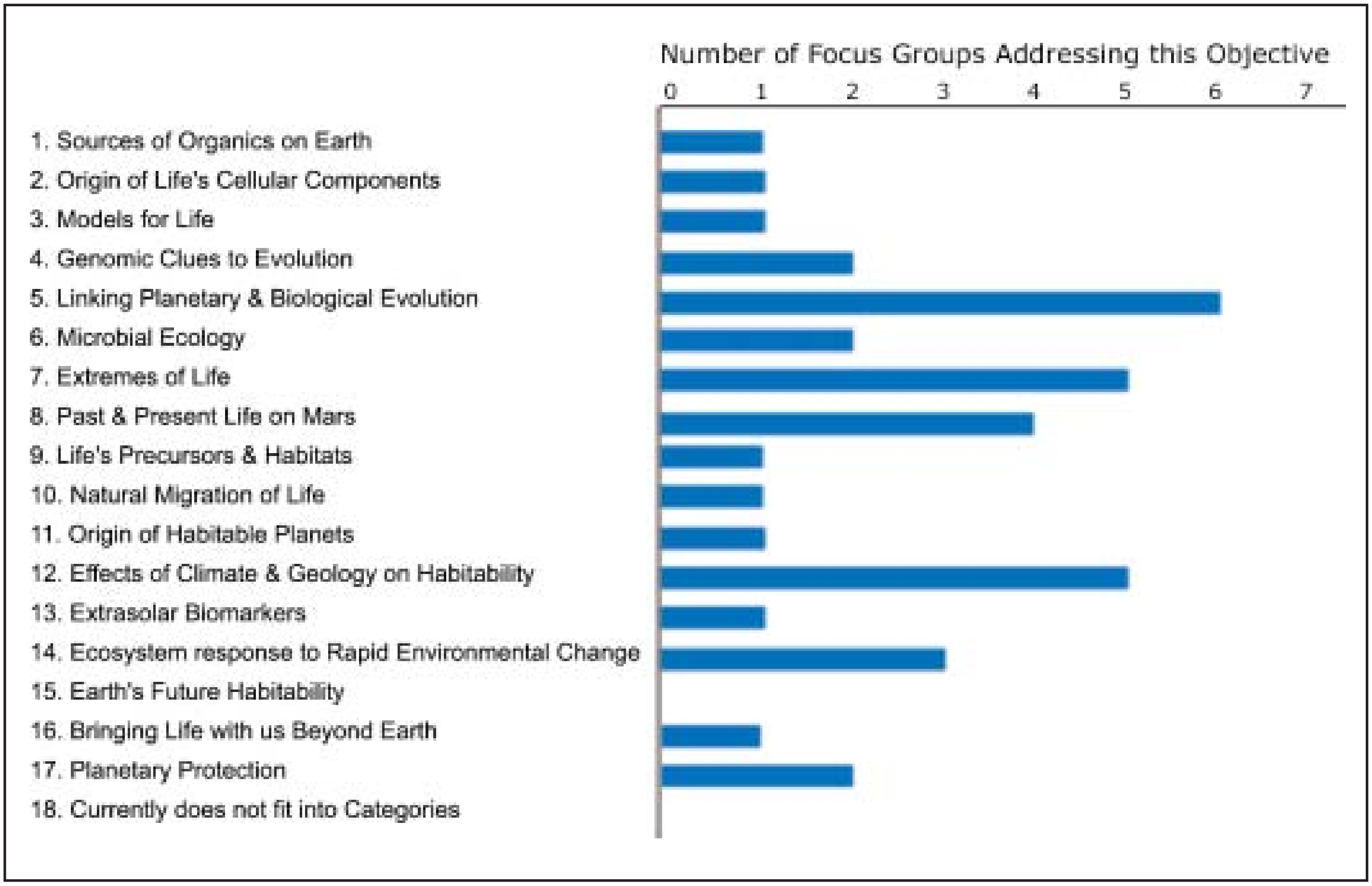
15. Earth's future habitability. Model the future habitability of Earth by examining the interactions between the biosphere and the chemistry and radiation balance of the atmosphere.

16. Bringing life with us beyond Earth. Understand the human-directed processes by which life can evolve beyond Earth.

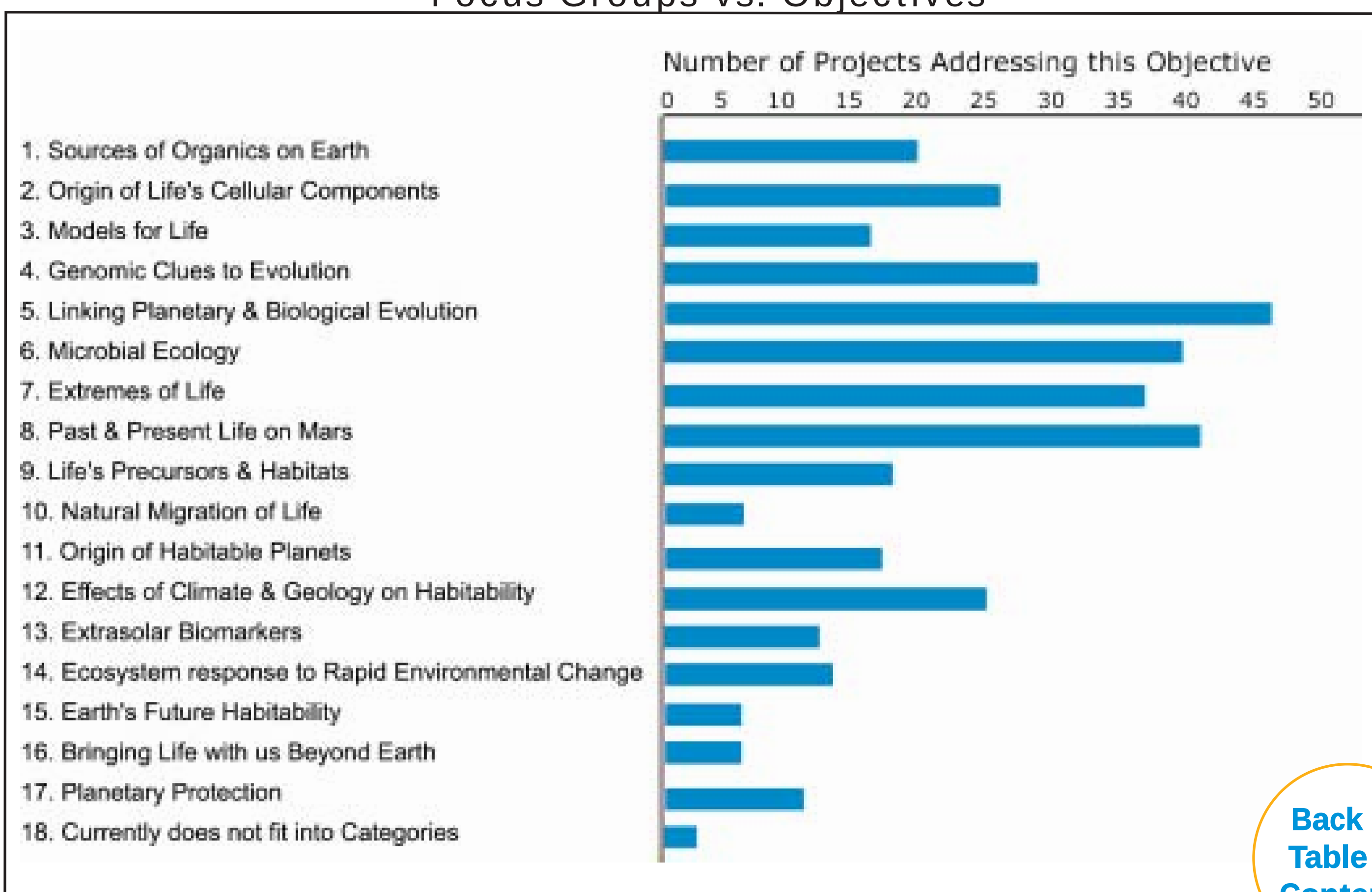
17. Planetary Protection. Refine planetary protection guidelines and develop planetary protection technology for human and robotic missions.

Back to Table of Contents

Lead Team Projects vs. Objectives



Focus Groups vs. Objectives



Back to Table of Contents