DOE/ER/61896--1

CONF-9408199--2

Support for NATO Advanced Study Institute on Molecular Ecology of Aquatic Microbes, August 28-September 9, 1994

Final Report on Grant no DE-FG02-94ER61896-1

Report prepared by:

Dr Ian Joint
Plymouth Marine Laboratory
Prospect Place, The Hoe
Plymouth
UK

Tel 44 1752 222772
Fax 44 1752 670637
email I.JOINT@PML.AC.UK

16 June 1995

Prepared for

THE U.S. DEPARTMENT OF ENERGY
AWARD NO. DE-FG02-94ER61896

Abstract

A NATO Advanced Study Institute entitled "The Molecular Ecology of Aquatic Microbes" was held at *Il Ciocco*, Castelvecchio Pascoli, Lucca, Italy from 28 August to 9 September 1994. This DOE grant enabled 5 additional lecturers to attend the ASI and support was also given to a number of young US scientists to enable their participation. The ASI was timely and reviewed progress in the development of molecular ecology. The quality of the lectures was extremely high; all participants found it a most stimulating meeting and were exposed to the very latest developments in the field of molecular ecology. In addition to formal lectures, tutorial sessions were arranged throughout the meeting. Themes for discussion were chosen by the students and 3 or 4 parallel discussion sessions, generally lasting for 2 hours, allowed students to focus on topics of most relevance to their own research. Each discussion was led by one of the lecturers or by one or more of the students who had particularly relevant experience. These tutorials allowed in-depth discussion of the advantages and disadvantages of particular approaches and techniques and the "tricks of the trade" were freely passed on.

Report

Background

The NATO Advanced Study Institute entitled "The Molecular Ecology of Aquatic Microbes" was organised by a steering committee of Dr Ian Joint (ASI Director, Plymouth Marine Laboratory, UK), Dr Paul Falkowski (Brookhaven National Laboratory, USA), Professor Noel Carr (University of Warwick, UK) and Dr Luigi Lazzara (University of Florence, Italy). The ASI was funded by NATO, who gave a grant of 2,800,000 Belgian Francs (*ca* $80,000). The conditions of the NATO grant were that no more than 15 lecturers could be funded from the NATO award. Since the steering committee hoped to attract a large proportion of the best researchers in the field, an application was made to the Department of Energy for a grant to support 5 additional lecturers from the US. Part of the grant was also to give assistance to younger scientists from the US to enable them to attend the ASI. The DOE gave a grant of $15520 to support the ASI (including a small amount to cover the travel costs of Dr Patrick Holligan of the Plymouth Marine Laboratory to attend a DOE meeting in the US).

Report of proceedings

The ASI was held at *Il Ciocco*, Castelvecchio Pascoli, Lucca, Italy from 28 August to 9 September 1994. It was attended by 96 scientists from 22 countries. The ASI was considered to be very timely because of the rapid progress which has been made in the last few years in applying the techniques of molecular biology to some previously intractable problems in aquatic biology. The ASI offered an ideal opportunity to review progress in the development of molecular ecology. The format of the meeting involved 2 or 3 formal lectures per day; these were not constrained by time and lecturers were given the freedom to talk for 90 minutes, or more if required. The quality of the lectures was extremely high; state-of-the-art developments were discussed and participants were exposed to the very latest approaches in the field of molecular ecology. In addition to formal lectures, tutorial sessions were arranged throughout the period of the ASI meeting. Most of the themes for discussion were chosen by the participants. Parallel discussion sessions, generally lasting for more than 2 hours, took place on most days. Each discussion was led by one of the lecturers but much of the liveliest discussion came from participants who had particularly relevant experience. These sessions focused on topics in-depth and allowed discussion of methodological problems, the advantages and disadvantages of particular approaches, novel technique development and the "tricks of the trade" were freely passed on in these discussions and problems were solved.

Financial report

The details of how the grant was spent are attached to this report as Appendix 1.

Summary of lectures supported by the DOE Grant

The grant supported 5 US lecturers: Dr Farooq Azam (Scripps Institution of Oceanography), Dr Stephen Giovannoni (University of Oregon), Dr Melvin Simon (California Institute of Technology), Dr Bess Ward (University of California, Santa Cruz) and Dr Jonathan Zehr ((Rensselaer Polytechnic Institute, New York). The following gives full title and authorship, with brief summaries, of their chapters.

Bacteria in Oceanic Carbon Cycling as a Molecular Problem: Farooq Azam, David C. Smith, Richard A. Long, Grieg F. Steward, *Marine Biology Research Division, Scripps Institution of Oceanography, University of California, San Diego, La Jolla, California 92093, USA*

This paper is written to facilitate dialogue between molecular biologists and marine microbial ecologists on research strategies in understanding the role of marine microbes in the oceanic carbon cycle. We think molecular biology can revolutionize marine microbial ecology and biogeochemistry, but at this early stage the challenge is to articulate fundamental unsolved problems in a way that shows how molecular approaches might be applied. In this paper we discuss one of the central problems in oceanic carbon cycling, namely bacteria-organic matter coupling. We will explain why bacteria-organic matter coupling is an important problem, stress the need for studying the problem in a suitable ecosystem context, formulate hypotheses for future research, and suggest how molecular approaches might be of value.

Microbial Diversity in Oceanic Systems: rRNA Approaches to the Study of Unculturable Microbes: Stephen J. Giovannoni, Thomas D. Mullins, and Katharine G. Field *Department of Microbiology, 220 Nash Hall, Oregon State University, Corvallis, OR 97331-3804, USA*

Bacterioplankton play important roles in nutrient regeneration and carbon flux in planktonic marine systems. Until recently, most ecological studies regarded bacterioplankton populations as a "black box" because standard bacteriological methods could not accurately resolve community composition. Knowledge of bacterioplankton community structure has advanced significantly with the application of ribosomal RNA gene cloning and sequencing methods to natural populations of bacteria. These studies have proven that numerous novel microbial taxa populate seawater. Many of these taxa branch deeply within phylogenetic trees and have only weak affiliations with known bacteria phyla. Of considerable importance to ecological studies has been the observation that many of the same taxa occur in surface samples from the Atlantic and Pacific Gyres. Thus, ecological studies which focus on the most abundant of these taxa using oligonucleotide probes may provide an important avenue for investigating bacterioplankton population dynamics. The hybridisation of taxon-specific oligonucleotide probes to rRNAs and rDNAs at the Bermuda Atlantic Time Series Station (BATS) in the Sargasso Sea has revealed pronounced spatial and temporal patterns in the distributions of major bacterioplankton groups.

Signal Transduction in Microorganisms: Melvin I. Simon *Division of Biology 147-75, California Institute of Technology, Pasadena, California 91125, USA*

The ability to adapt to an ecological niche requires a variety of sensory mechanisms. Bacteria are equipped with systems that allow them to measure the physical and chemical parameters of their environment and to respond in an ecologically appropriate fashion. Thus, for example, there are clear mechanisms for sensing osmolarity, nutrient, temperature, light, pressure, and surfaces, as well as a variety of other characteristics, including the density of organisms in the environment. All of this information is integrated into a series of complex biochemical circuits. Even simple organisms are

capable of a variety of responses ranging from modifications of biochemical pathways to motility, growth, adhesion and other adaptation.

Many sensory signalling mechanisms have similar characteristics, and these characteristics are mediated by a variety of proteins, including specific receptors, kinases, and methylases that amplify and transduce chemical information into metabolic changes, and effectors that modulate enzyme activity and mediate changes in gene expression and gene function. We will describe some of the circuitry that has been studied in a variety of microorganisms and show how the information processing systems are required by the organism in order for it to efficiently and successfully populate a specific environment.

Functional and Taxonomic Probes for Bacteria in the Nitrogen cycle: Bess B. Ward *Marine Sciences Program, University of California, Santa Cruz, CA 95064, USA*

The contrasting physiology and phylogeny of nitrifying and denitrifying bacteria require different methods for detection and quantification in the environment. rRNA probes are appropriate for nitrifiers. Denitrifiers are more diverse; the conserved enzymology of denitrification provides a functional approach for probes. Although quantification is still rudimentary, comparison among immunofluorescence enumeration, PCR amplification and quantitative hybridization allow an assessment of genotype vs phenotype abundances. Nitrifiers are a very small fraction of the total bacterial population, but a few serotypes and rRNA groups are ubiquitous in natural waters. The denitrification genotype is apparently abundant, but individual denitrifying strains are present in very low numbers.

Nitrogen Fixation in the Sea: Why Only *Trichodesmium*? Jonathan P. Zehr *Biology Department, Rensselaer Polytechnic Institute, Troy, New York 12180, USA*

The relative importance of different nutrients in limiting primary production in the sea continues to be the subject of debate, but it is clear that several nutrients are often in short supply in many regions of the oceans. Nitrogen fixation capabilities should provide an ecological advantage to microorganisms in the oceanic environment, regardless of the primary nutrient limiting productivity. *Trichodesmium* is a filamentous nonheterocystous nitrogen-fixing cyanobacterium which is a conspicuous component of tropical and subtropical oceans, and appears to play a major role in carbon and nitrogen-fixation in regions where it is found. It is not intuitively obvious which characteristics of *Trichodesmium* confer an ecological advantage such that it is the predominant organism to exploit nitrogen fixation as a mechanism to obtain nitrogen in nitrogen-deficient oligotrophic oceans. Furthermore, there still remains the question of whether *Trichodesmium* is truly one of only a few species to fix nitrogen, or whether other nitrogen fixing organisms exist in the open ocean, but have yet to be cultivated. Molecular approaches have provided a way to examine both of these issues: what is the distribution of nitrogen fixing microorganisms in the marine environment, and what are the molecular and biochemical features that determine the ecological success of *Trichodesmium* in the open ocean environment?

Publication

The proceedings of the ASI are in press. A copy of the book will be sent to DOE as soon as it is published (publication is expected in September 1995). Appendix 2 is a copy of the preface to the book, which formally acknowledges the DOE Grant. Appendix 3 is a list of chapters from the book.

The full reference of the book is:

***Molecular Ecology of Aquatic Microbes* (ed I Joint)**
Springer Verlag, NATO ASI Series G, 38: 1-415

The price of the book is not yet known.

Financial summary removed

APPENDIX 2

Copy of Preface page from book, with Acknowledgement of DOE Grant

Preface

A NATO ASI on "Molecular Ecology of Aquatic Microbes" was held at *Il Ciocco,* Lucca, Italy from 28 August - 9 September 1994. The aims of the ASI were to evaluate the potential for molecular biology to solve some important questions in aquatic microbiology, particularly in relation to biogeochemical cycling and microbial physiology.

Techniques developed by molecular biologists have now been adopted by a wide range of scientific disciplines. In the last 5 years, aquatic microbial ecologists have begun to incorporate these methods into their research and, as a result, are developing a much clearer understanding of phylogenetic diversity, the molecular basis of physiological acclimations and the transduction of environmental signals and organism responses. The aim of this ASI was to assess progress in this new field of research, to compare and describe techniques and experimental approaches, and to foster communication between disciplines. The ASI offered an excellent opportunity to bring together aquatic ecologists with molecular biologists and to encourage efficient technology transfer. The meeting provided a forum for detailed and broad exchange of information on the status and trends of aquatic molecular ecology and to assess how emerging molecular techniques might solve some important problems in ecology which have prove intractable because of lack of appropriate methodologies.

The organising committee was Dr Paul Falkowski (Brookhaven National Laboratory, USA), Professor Noel Carr (University of Warwick, UK) and Dr Luigi Lazzara (University of Firenzi, Italy) I would like to express my appreciation for the efforts they made to ensure a successful meeting.

I would also like to acknowledge financial support from a number of organisations. NATO provided the majority of the funding which allowed this meeting to take place. The US Department of Energy gave generous assistance through grant number DE-FG02-94ER61896, which enabled us to invite 5 additional lecturers. The UK Natural Environment Research Council also gave a grant from the Special Topic on Molecular and Genetic Advances, which supported 2 lecturers from the UK. The generous support of all these organisations is gratefully acknowledged.

Ian Joint
ASI Director

APPENDIX 3
Copy of contents pages from book

CONTENTS

The Potential of Molecular Ecology
IAN JOINT. 1

Towards Understanding the Molecular Ecology
of Phytoplankton Photosynthesis
PAUL G. FALKOWSKI. 17

Bacteria in Oceanic Carbon Cycling as a Molecular Problem
FAROOQ AZAM, DAVID C. SMITH, RICHARD A. LONG, GRIEG F. STEWARD. . . 39

The Role and Regulation of Microbes in Sediment Nitrogen Cycle
HENRY BLACKBURN. 55

Functional and Taxonomic Probes for Bacteria in the Nitrogen Cycle
BESS B. WARD. 73

The Role of Mixotrophy in Pelagic Environments
BO RIEMANN, HARRY HAVSKUM, FREDE THINGSTAD, AND
CATHERINE BERNARD. 87

Successional Change in the Planktonic Vegetation:
Species, Structures, Scales
COLIN S. REYNOLDS. 115

Can Molecular Techniques Change Our Ideas
About the Species Concept?
LINDA K. MEDLIN, MARTIN LANGE, GARY L.A. BARKER, AND
PAUL K. HAYES. 133

How Do Cyanobacteria Perceive and Adjust to Their Environment?
JEAN HOUMARD. 153

How Do Cells Express Nutrient Limitation at the Molecular Level?
NICHOLAS H. MANN. 171

The Problem of Excess and/or Limitation of the Habitat Conditions:
Do Natural Assemblages Exist?
RICARDO GUERRERO AND JORDI MAS-CASTELLÀ. 191

APPENDIX 3 (cont)
Copy of contents pages from book

Signal Transduction in Microorganisms
MELVIN I. SIMON. 205

Microbial Diversity in Oceanic Systems:
rRNA Approaches to the Study of Unculturable Microbes
STEPHEN J. GIOVANNONI, THOMAS D. MULLINS, AND KATHARINE G. FIELD. 217

Viruses - the New Players in the Game; Their Ecological Role
and Could They Mediate Genetic Exchange by Transduction?
GUNNAR BRATBAK AND MIKAL HELDAL. 249

Molecular Analysis of Plastid Evolution
WOLFGANG LÖFFELHARDT. 265

Can We Estimate Bacterial Growth Rates from Ribosomal RNA Content?
PAUL F. KEMP. 279

The Cell Cycle of Phytoplankton:
Coupling Cell Growth to Population Growth
DANIEL VAULOT. 303

Response of Photosynthetic Microorganisms
to Changing Ambient Concentration of CO_2
MICHAL RONEN-TARAZI, RAKEFET SCHWARZ, ANNE BOUEVITCH,
JUDY LIEMAN-HURWITZ, JONATHAN EREZ AND AARON KAPLAN. 323

Nitrogen Fixation in the Sea: Why Only *Trichodesmium*?
JONATHAN P. ZEHR. 335

Molecular Ecology of Marine Methanotrophs
J COLIN MURRELL AND ANDREW J HOLMES. 365

Microbial Cultures and Natural Populations
NOEL G CARR. 391

List of Participants. 403

Subject Index. 411