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6. AUTHOR(S) Rebecca Van Beneden				
7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES) Duke University School of the Environment Marine Laboratory 111 Pivers Island Road Beaufort, NC 28516			8. PERFORMING ORGANIZATION REPORT NUMBER	
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13. ABSTRACT (Maximum 200 words) The ONR sponsored grant entitled "Advanced Research Training in Marine Molecular Biology and Biotechnology" founded a training course to teach students and established scientists the basic principles of modern recombinant DNA technology. The emphasis was on the development of marine models and problems in environmental and ocean sciences. Students were trained in the isolation and characterization of DNA and RNA which included Southern and Northern blotting, DNA sequencing and cDNA libraries. Other techniques taught were the use of the polymerase chain reaction (PCR), <u>in situ</u> hybridization, gene transfer, mtDNA analysis and immunoblotting. These techniques were applied to problems in environmental toxicology, marine microbiology, the development of transgenic organisms, evolution and population studies, developmental biology and gene expression analysis.				
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FINAL TECHNICAL REPORT

Grant#: N00014-92-J-1958

R&T Code: 4412130

PRINCIPAL INVESTIGATOR: Rebecca J. Van Beneden, Ph.D.

INSTITUTION: Duke University School of the Environment
Marine Laboratory
Beaufort, NC 28516

GRANT TITLE: Advanced Research Training in Marine Molecular Biology
and Biotechnology

AWARD PERIOD: May 1, 1990 - April 30, 1992

I. Summary of all work accomplished

Objective: To train undergraduate, graduate and post-graduate students in the basic principles of modern molecular biology and recombinant DNA techniques. The emphasis was on the application of these methods to study processes in biological oceanography and the development of marine organisms as model systems.

Approach: Students were trained in basic techniques of molecular biology, which included: (1) DNA ANALYSIS: (DNA isolation, Agarose gel electrophoresis, Southern blotting, hybridization, plasmid preparation and DNA sequencing); (2) MARINE MICROBIOLOGY: (taxonomy of *Vibrio spp.* using molecular techniques, immunoblotting, antibiotic production, PCR); (3) GENE TRANSFER AND MUTAGENESIS: (cell culture, electroporation, calcium phosphate precipitation, use and recovery of shuttle vectors); (4) RNA ANALYSIS: (RNA isolation, preparation and screening of cDNA libraries, Northern blots); (5) DEVELOPMENTAL BIOLOGY OF SEA URCHINS: (*in situ* hybridization, micromanipulation); (6) mtDNA ANALYSIS: (mtDNA isolation, PCR).

Staff members:

Dr. Rebecca Van Beneden, Duke University (year 01/02)
Dr. James Oliver, Univ. of North Carolina at Charlotte (year 01)
Drs. James and Barbara Burkhart, NIEHS, Research Triangle, NC
(year 01/02)
Dr. Patricia McClellan-Green, Duke University (year 01/02)
Dr. Gary Wessel, Brown University (year 01)
Dr. Robert Chapman, East Carolina University (year 01)
Dr. Chuck Ettensohn, Carnegie-Mellon Institute (year 01)
Dr. C.M. Lin, Center of Marine Biotechnology, Baltimore, MD
(year 02)
Dr. Mark Martindale, University of Chicago (year 02)
Dr. Jonathan Henry, University of Illinois (year 02)
Dr. Jeff Hardin, University of Wisconsin (year 02)

Students (year 01):

Robin Huff, Graduate Student, Duke University
Katherine Reagan, Graduate Student, Duke University
Kevin Fielman, Graduate Student, University of Delaware
Karen Rogowski, Masters Student, Brooklyn College, NY
Dr. Kevin Kleinow, Assistant Professor, Louisiana State
University
Dr. Richard DiGiulio, Associate Professor, Duke University
Dr. Tony Clare, Assistant Research Professor, Duke University
Dr. Peter Thomas, Professor, University of Texas, Marine Science
Institute, Port Aransas
V. Sasikumar, Graduate Student, University of Madras, India
Sandy Blake, Graduate Student, VA Institute of Marine Science
Gretchen Colon, Graduate Student, University of Puerto Rico

Students (year 02):

Dr. Richard Lee, Professor, Skidaway Institute, Georgia
Seng Chen, Graduate Student, University of Texas, Marine Science
Institute, Port Aransas
Dr. Yolanda Brady, Assistant Professor, Auburn University
Michael Duvall, Graduate Student, Duke University
Howard Kator, Assistant Professor, Virginia Institute of Marine
Science
Mark Barrett, Graduate Student, Duke University
Julie Olsen, Graduate Student, University of North Carolina,
Chapel Hill
Jesus Paningua, Postdoctoral Fellow, D.S. Ciecese, Ensanada,
Mexico
William Wallace, Graduate Student, SUNY, Stony Brook
Gregory Street, Graduate Student, University of Texas, Marine
Science Institute, Port Aransas

Accomplishments:

The course for the last two years was highly successful, well organized and updated annually. Student evaluations indicated that they both enjoyed this course and were challenged by its content. The course was limited to ten students. Lectures focused on basic molecular genetics and theory of recombinant techniques. The majority of the time was spent in the laboratory where the students received invaluable hands-on experience. Students were exposed to numerous techniques, as described above, and to the unique aspects of marine organisms as models for a wide variety of studies.

Both Robin Huff and Katherine Reagan, graduate students in the Pharmacology Department at Duke University continue to use techniques learned in the course in their graduate research. Kevin Fielman and Karen Rogowski entered Ph.D. programs in the fall of 1993 using molecular techniques to address problems in marine sciences. Karen's thesis advisor is Dr. Peter Thomas, who was her classmate in the 1993 class. Dr. Kevin Kleinow used the knowledge he gained in this course to develop a similar course. Dr. Richard DiGiulio and I have initiated a collaborative effort to examine molecular mechanisms of

carcinogenesis in a sensitive and a resistant catfish species. These examples illustrate the far-reaching impact of this course and the flow of information from molecular biology to problems in the biological ocean sciences. Jose Gomez, my first independent study student (class of 1990) graduated in 1993 from Scripps. His studies were on mtDNA, the analysis of which he learned in my course and during his independent project in my laboratory. Dr. Robert Cashon (class of 1991) is now Assistant Professor at the University of Maine and is using techniques he learned in the class to study free radical damage in fish cell lines.

II. Index of all publications

1. Course Bulletins (years 01 and 01)

III. Patents Pending or Filed

not applicable

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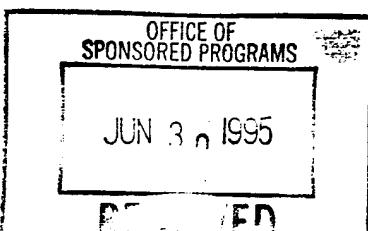
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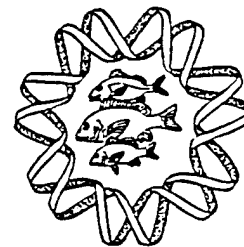
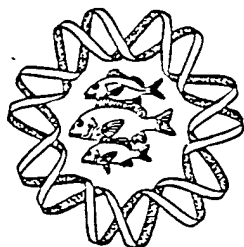
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ADVANCED RESEARCH TRAINING IN MARINE MOLECULAR BIOLOGY AND BIOTECHNOLOGY



Department of Cell Biology
and

DUKE UNIVERSITY SCHOOL OF THE ENVIRONMENT
at the Marine Laboratory

Sponsored by the Office of Naval Research

WHEN

Session I: Lecture/Lab, May 18-June 19, 1992
Session II: Independent Study, June 22-July 24, 1992

WHERE

Duke University Marine Laboratory

FACULTY (Teaching Specialties)

Rebecca Van Beneden, Duke Univ. (DNA Analysis)
James Oliver, UNC-Charlotte (Marine Microbiology)
Jim and Barbara Burkhart, NIEHS (Eukaryotic Gene Transfer and Mutagenesis)
Patricia McClellan-Green, Duke Univ. (Gene Expression)
Robert Chapman, East Carolina Univ. (mt DNA and PCR)
Gary Wessel, Brown University, and Charles Etensohn, Carnegie Mellon (in situ Hybridization and Micromanipulation in Sea Urchins)

FEES

Tuition [1.5 UG course or 6 grad units (6 sem. hrs.)
\$2,256]
Room & Board at DUML [\$925]; Health Fee [\$44]

ONR and DUKE FELLOWSHIPS

Tuition, Room & Board available on a competitive basis

FOR APPLICATION & INFORMATION, CONTACT

Admissions Office
Duke University Marine Laboratory
Beaufort, North Carolina 28516
(919) 728-2111

THE BEAUFORT SETTING

The Duke University Marine Laboratory is situated on Pivers Island, within the Outer Banks of North Carolina, and adjacent to historic Beaufort. The area's system of barrier islands, sounds, and estuaries is well known for its rich flora and fauna, making the area an excellent resource for those interested in the pursuit of marine science.

COURSE DESCRIPTION

The course is an intensive training program at the graduate level in molecular biology laboratory techniques. By using marine models and the tools of molecular biology, trainees will obtain direct knowledge of indigenous marine organisms and discover the unique properties that allow them to be used as models in environmental and human health problems. Research methodology includes: Week 1 - DNA isolation, Southern blotting, gene mapping and DNA sequencing; Week 2 - RNA isolation, cDNA cloning; Week 3 - mt DNA isolation, cell culture, gene transfer techniques; Week 4 - basic bacteriology, characterization of marine bacteria, PCR DNA amplification; Week 5 - in situ hybridization and micromanipulation. Session I is listed as Cell Biology CBI 235 (4 grad units, lecture only) or Cell Biology CBI 235L (6 grad units, laboratory included). Session II, Independent Study, requires CBI 235/235L as a prerequisite and prior approval of instructor.

DESIGNED FOR BEGINNERS

We invite applications from **graduates, postgraduates, and qualified undergraduates**. No previous experience in molecular biology is required or expected, but basic background in genetics and biochemistry is assumed. To insure a close, small-group learning experience, class size will be limited to 10 in laboratory sessions and 20 in lectures.

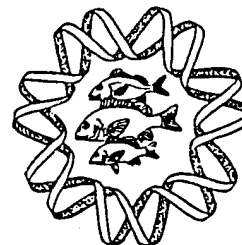
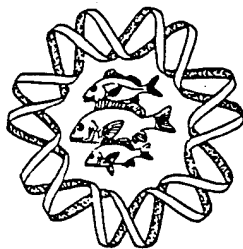
FOR ADDITIONAL INFORMATION ON COURSE CONTENT, CONTACT

Dr. Rebecca Van Beneden and
Dr. Patricia McClellan-Green, Course Directors
(919) 728-2111, ext. 271, 263

APPLICATION DEADLINE

March 20, 1992

ADVANCED RESEARCH TRAINING IN MARINE MOLECULAR BIOLOGY AND BIOTECHNOLOGY



*Department of Cell Biology
and*

*Duke University School of the Environment
Marine Laboratory*

Sponsored by the Office of Naval Research

WHEN

Session I: Lecture/Lab, May 17-June 18, 1993

Session II: Independent Study, June 21-July 23, 1993

WHERE

Duke University School of the Environment Marine Laboratory
Beaufort, North Carolina 28516

FACULTY (Teaching Specialties)

C.M. Lin, Center of Marine Biotechnology, Univ. MD. (DNA Analysis)

James and Barbara Burkhardt, NIEHS (Eukaryotic Gene Transfer and Mutagenesis)

Patricia McClellan-Green, Duke Univ. (Gene Expression)

T.B.A. (Marine Microbiology)

T.B.A. (Developmental Models)

FEES

Tuition [1.5 UG course or 6 grad units (6 sem. hrs.) \$2,412]

Room & Board at DUML [\$950]; Health Fee [\$44]

ONR and DUKE FELLOWSHIPS

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Dr. Patricia McClellan-Green, Course Directors

(919) 728-2111, ext. 271, 263

APPLICATION DEADLINE

April 12, 1993