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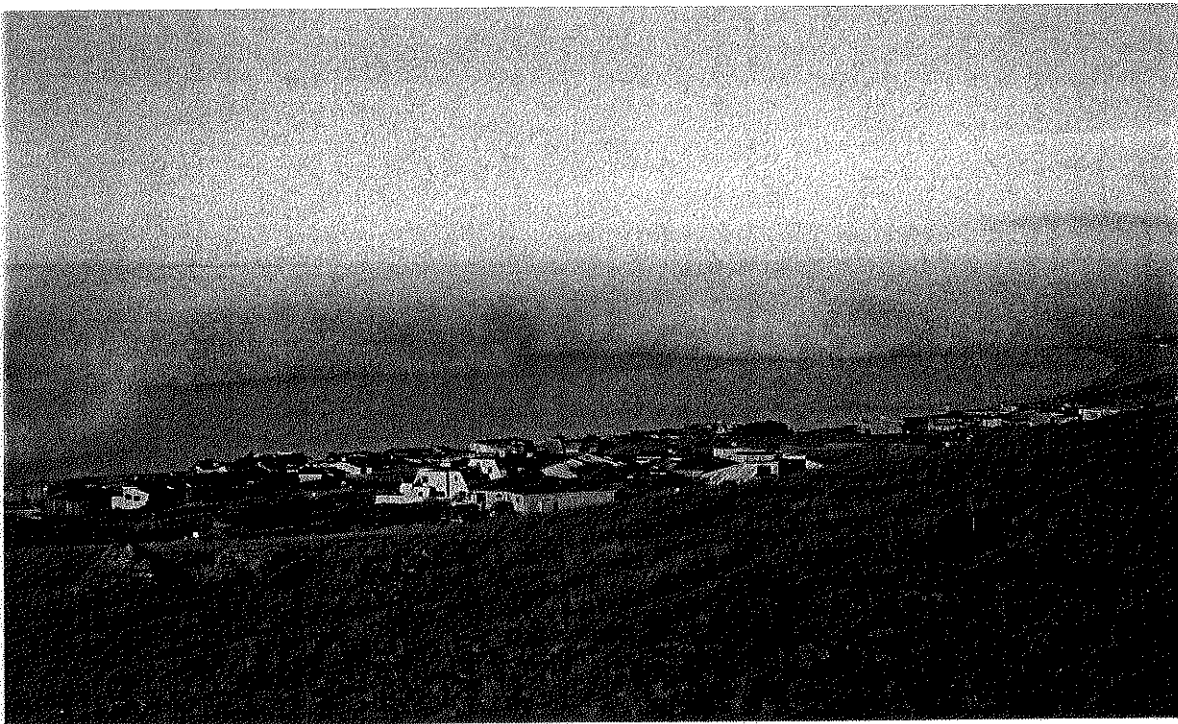
# California Marine Research and Cooperative Extension Conference

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Radisson Hotel  
Sacramento, CA  
May 31 - June 1, 2001

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Conference Organizers

Ted Grosholz: Department of Environmental Science and Policy, Cooperative Extension, UC Davis  
Paul G. Olin: University of California Cooperative Extension, Sea Grant Extension Program

Editors

Paul G. Olin: University of California Cooperative Extension, Sea Grant Extension Program  
Victoria C. Wilson: University of California Cooperative Extension

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The California Sea Grant College System is a statewide, multi-university program of marine research, education, and extension activities, administered by the University of California. Sea Grant-sponsored research contributes to the growing body of knowledge about our coastal and ocean resources and, consequently, to the solution of many marine-related problems facing our society. Through its Marine Extension Program, Sea Grant transfers information and technology developed in research efforts to a wide community of interested parties and actual users of marine information and technology, not only in California but throughout the nation. Sea Grant also supports a broad range of educational programs so that our coastal and ocean resources can be understood and used judiciously by this and future generations.

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Cover Photo: Fog bank moving onshore at Bodega Bay.

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## **Acknowledgements**

I would like to express my appreciation to the Marine and Coastal Workgroup of the University of California Division of Agriculture and Natural Resources, and to the California Sea Grant College Program for their support in providing this opportunity for information exchange and collaboration. It is hoped that this will stimulate future efforts to increase cooperation and strengthen the research and extension continuum to effectively provide "Science Serving California's Coast"

Significant support in organizing this conference was provided by Kim Beard and Felicia Bradshaw from the University of California Sea Grant Extension Program. Their assistance is highly valued and appreciated.

## Introduction

California's remarkable 1,200 mile coastline supports an unparalleled wealth of natural resources being used for commerce and recreation by the State's 34 million residents. Research to better understand, utilize and protect California's coastal and marine resources is crucial in a state where 80 percent of the people live in coastal areas. Equally as important, are close linkages between the research and extension communities in order to provide timely information to legislators, resource managers and the public. To insure that marine research continues to be valued and supported by the public, it is important that applied research addresses priority needs, and that the public has an understanding and appreciation for basic research.

As California's population increases there will be growing demands placed on marine and estuarine resources, and a tremendous need for science based information to develop sound management policies. Research is crucial to insure that in the future we will have stable fisheries, sustainable aquaculture, and functioning estuarine and marine ecosystems that support abundant fish and wildlife, and the commercial and recreational opportunities afforded by them. Research can create the knowledge that will provide solutions to problems facing our natural resources today and in partnership with extension programming, this information can be applied to insure the sustainable use and management of these resources in the future.

# California Marine Research and Cooperative Extension Conference

## Plenary Session

**Welcome and Introduction: Ted Grosholz** – Department of Environmental Science and Policy and ia Cooperative Extension, University of California, Davis

**The Extension-Research Continuum: A Blending of Two Success Stories: Russell Moll**, Director, California Sea Grant

**Update from the Office of Research: Larry Coleman**, Interim Vice-Provost for Research, Office of the President.



## Workshop Abstracts

### The University Of California Coastal Environmental Quality Initiative

Gary B. Griggs, Chair, University of California Marine Council

California is a coastal-dependent state yet the economic value of its marine resources is largely under-appreciated. Recreation and tourism in California are major economic engines, producing ~\$10 billion in revenues annually and supporting over 500,000 jobs. The state has six major ports with a yearly economic impact of \$3.4 billion. Commercial and recreational fisheries generate an additional nearly \$1 billion. A 1994 study concluded that seven ocean-dependent industries contributed more than \$17 billion to the state's economy annually.

To be healthy and productive, coastal oceans and the industries that depend on them need to be healthy and sustainable. It is apparent in California that human activities have led to significant modifications of the coastal zone's ecological systems, seriously impacting their ability to sustain themselves. Near shore waters receive wastewater from domestic, industrial and agricultural drainage, often with significant negative impacts. Many of the state's fisheries have collapsed, and former economically valuable species are now on the endangered list. Contaminated sediments have increasingly begun to restrict dredging of our major ports, through which 95% of our foreign trade must pass. Invasions of non-native species are threatening California's estuaries. We see the warning signs, but we don't have a comprehensive picture of the magnitude of these problems and their cumulative impacts on the economic well being of California.

There is a pressing need to initiate a systematic and long-term assessment of representative environmental health indicators and understand how the coastal environment functions and how human activity has affected this zone. These issues must be addressed in ways that will not only contribute to increasing our scientific understanding of both natural processes and human-induced changes, but that will also be useful to legislators, policy makers and managers who must make the decisions and develop the policies and legislation required for the long-term sustained health of California's coastal ocean.

In response to these concerns the University of California Marine Council developed the Coastal Environmental Quality Initiative as a long term approach that would utilize the collective expertise of the marine scientists within the University of California to focus attention on California coastal problems. A need exists for monitoring, mapping and assessing critical physical, biological and environmental parameters for the entire coastal zone of California. The population trends of threatened marine mammals, trends in catch statistics for commercial species, incidents of beach closures and water quality indicator trends, and incidences of non-native species invasions are examples of potentially useful indicators of ocean health that need to be studied and understood.

The University of California is well recognized for its strength in policy research and has a critical role to play in building bridges between scientific research and responsive policy development. Links are necessary between University natural and social scientists, and state

resource managers and policy makers to ensure that research informs long term policies that lead to recovery or sustaining the state's coastal ocean resources. With \$1,500,000 in annual support from the State of California, the UC Marine Council, working with the Resources Agency and RASGAP, developed a funding program, identified a set of research priorities, developed a Request for Proposals, and initiated the program in early 2001. Three broad research areas identified as priorities in the first year of the Coastal Environmental Quality Initiative include 1) coastal water and sediment quality, 2) fisheries and marine ecosystems, and 3) coastal hazards and shoreline processes.

# **The Center for the Coastal Environment at Scripps Institution of Oceanography**

John Largier, Director, Center for the Coastal Environment, SIO/UC San Diego

Scripps has long been involved in pioneering coastal ocean research. Over the years the organizational structure at Scripps has become increasingly complex and research has become increasingly specialized. The Center for the Coastal Environment (CCE) was initiated last year to integrate scientific research, developing more of a systems approach, and linking research science with societal needs. These twin aims guide the design and development phase of the CCE. In a brief presentation, I will discuss:

- (i) our approach to addressing societal issues while remaining focused on the aims of academic research;
- (ii) difficulties and pleasant surprises encountered in dealing with researchers, university hierarchy, public agencies, NGO's, etc.;
- (iii) our vision of common purpose, requiring a non-exclusive and collaborative relationship with other research institutions.

## **The UC Davis Wildlife Health Center's Marine Ecosystem Health Program: Supporting Science and its Linkage to Stakeholders**

Kirsten V.K. Gilardi, Coordinator, Marine Ecosystem Health Program,  
Wildlife Health Center, School of Veterinary Medicine, UC Davis

The mission of the UC Davis Wildlife Health Center's Marine Ecosystem Health Program (MEHP) is to restore and maintain the health of marine wildlife and ecosystems by supporting conservation, research, and management efforts. The scope of the program encompasses all marine vertebrates, and the biotic and abiotic environments upon which they depend for survival. The program was established in January 2000 with a private gift to the Wildlife Health Center. Presently, the program focuses on the North American Pacific, with emphasis given to the issues facing the Inland Sea of Washington and British Columbia (Puget Sound/Northwest Straits/Georgia Basin) region.

The MEHP provides a mechanism by which ecosystem health problems in a target area are identified, solutions are made possible through funding and facilitation of scientific investigations, and important information is translated for, and transmitted to, stakeholders. The program encourages and supports cooperation and interaction among scientists, managers and stakeholders through preferential funding of projects that demonstrate strong linkages between project outcome and on-the-ground maintenance or restoration of marine ecosystem health, and through facilitation of strategic convening. Principal components of the MEHP include the following:

- 1) A competitive grants program, awarding up to \$300K annually to applied science and conservation or management projects. Projects funded in 2000 included studies of the population genetics of green sea urchins in the Northwest Straits, assessments of forage and rockfish habitat in Island and San Juan counties, expansion of a beached seabird survey program into the Northwest Straits, a toxicological assessment of orca prey, a study of the impacts of sea otters on nearshore benthic communities in the Strait of Juan de Fuca, the use of plasma metabolites to assess habitat quality for migrating sandpipers, and the spread and impact of introduced Japanese seaweed on native kelp forests of the San Juan Archipelago. Requests for Pre-proposals are issued in April of each year.
- 2) An Advisory Board and Scientific Advisory Committee comprised of scientists and stakeholders, serve to advise the Coordinator on programmatic issues, to select the most meritorious project proposals for funding, and to ensure that the program is responsive to the needs of stakeholders; and
- 3) A staff scientist residing year-round in the San Juan Islands, whose chief responsibilities are to serve as a translational scientist for local, state and federal natural resource managers and stakeholder groups, to coordinate wildlife and ecosystem health-related research or management projects, and to network with researchers and stakeholders.

The MEHP is one of several targeted ecosystem health programs administered by the Wildlife Health Center (WHC) at the UC Davis School of Veterinary Medicine. The WHC is dedicated to balancing the needs of people, wildlife, and the environment, by recruiting scientists, industry,

and conservationists to focus their efforts on targeted issues. One of its major tools is the competitive grants model, in which funds for research and management activities are raised and then awarded. This model ensures stakeholder involvement (fund raising and receiving) and helps provide the objective science necessary for sound decision-making and policy development.

## **Marine Fisheries Opportunities Related to the Marine Life Management Act**

Christopher M. Dewees, Marine Fisheries Specialist  
and Sea Grant Extension Program Leader, UC Davis

The Marine Life Management Act (MLMA) of 1998 is making fundamental changes in how California fisheries are managed. The Act calls for fishery management plans for state managed fisheries and these plans need to incorporate the best available science, peer and constituent review, as well as periodic evaluation. The state is looking for collaboration and partnerships with universities in support of MLMA implementation. Opportunities are opening up for marine fisheries research and extension projects with potential funding from the state, Sea Grant and other sources.

There are a wide variety of possibilities ranging from developing improved cost-effective methodologies of estimating resource abundance, increasing understanding of nearshore ecosystems and predicting likely social and economic effects of management alternatives, to expanding the management capabilities of the Department of Fish and Game. The Sea Grant Extension Program has already become involved with compiling the "Status of the Fisheries" report and setting up a peer-review system for independent review of CDFG marine fisheries research and management activities. I will discuss some of these opportunities.

# **Marine Reserve Design for Conservation and Fisheries Management: A Case Study from the California Channel Islands**

Deborah A McArdle, Marine Advisor, California Sea Grant  
UC Cooperative Extension, Santa Barbara

Eight large islands with a diverse wealth of resources lie off the coast of southern California, USA. Natural resource decisions made at the Channel Islands have traditionally addressed human needs (e.g., fisheries, recreation, mineral extraction) through the management goals of a particular agency, organization, or interest group. Agencies have attempted to conserve and protect the area's scenic, ecological and scientific values with the designation of eight marine protected areas (MPAs/reserves). However, despite their number and sizes, these designations provide limited protection for living marine resources. Only one small marine reserve (Landing Cove, Anacapa Island) prohibits all take of marine organisms year-round (14.9 ha or 0.004% of the Sanctuary). A perceived steady deterioration in the region's biological resources has led many to question the ability of current management approaches to provide long-term environmental and economic viability. To this end, the California Fish and Game Commission requested that a science, economic and stakeholder panel be established to design an ecologically sound reserve network for the Channel Islands. In this paper, I discuss the ecological criteria necessary to evaluate alternative reserve designs, and provide an example of how these criteria were applied in the Channel Islands National Marine Sanctuary. One of the most critical components of the analysis was the data on habitats in the Channel Islands. The analysis is guided by a set of clear goals that reflect the interests of agencies, fishermen, environmentalists, and other members of the community. The dual goals of biodiversity conservation and fisheries sustainability could be met through this habitat-based approach.

## **Antifouling Strategies Program for Recreational Boats**

Leigh Taylor Johnson, University of California Sea Grant Extension Program

The Regional Water Quality Control Board in San Diego is conducting a Total Maximum Daily Load (TMDL) analysis for copper in the Shelter Island Yacht Basin of northern San Diego Bay. They have found that recreational boats are the primary source of the copper that is contaminating the sediments and water column. Dissolved copper in the water column exceeds the federal and state standard of 3.1 parts per billion (micrograms per liter). Samples of water from the basin kill or cause abnormal development in mussel larvae.

Copper contamination has created environmental compliance costs for the San Diego Unified Port District. When marinas are dredged to maintain depth, the Port must pay higher disposal fees because the contaminated material is considered hazardous waste. Copper removed from boats during haulout and repainting raises the environmental compliance costs paid by boatyards under their National Pollutant Discharge Elimination System (NPDES) permits. Ultimately, these costs could be passed on to marinas and boaters through leases, slip fees and haulout costs. Because boating is an amenity, it is price sensitive. Thus, environmental compliance costs could affect this industry that contributed \$11 billion to the gross state product in 1995, according to the California Department of Boating and Waterways.

The 2000 Plan for California's Nonpoint Source Pollution Control Program (Vol. I, pp. 139-141) requires the underwater hull cleaning industry to train and certify divers in the use of best management practices. The state will step in if 75% of boat cleanings are not done by certified divers within four years. The plan also mandates a phase-out of toxic hull paints on state and local agency-owned vessels. Finally, it recommends legislation to ban toxic hull paints, if necessary, after a thorough analysis of the situation.

The TMDL assessment is nearly complete and Regional Board staff will soon begin to develop strategies to reduce copper emissions from boat hulls in the basin. One option that has been discussed would be to phase in a reduction of the number of craft that use a copper-based antifouling paint. There are a few, nontoxic coatings available for recreational boats. However, using a nontoxic strategy successfully requires a new paradigm.

The traditional paradigm is to add a toxicant, such as cuprous oxide, to the bottom paint. The toxicant gradually leaches to the surface of the paint, where it slows fouling growth and is ultimately lost to the water. In San Diego the hull must be cleaned in the water monthly to remove early stages of fouling growth. Hull cleaning accelerates copper loss and best management practices are used to keep it to a minimum. Eventually, most of the cuprous oxide is lost, the paint is no longer effective, and the boat must be hauled and repainted.

The nontoxic paradigm does not slow fouling growth. Nontoxic coatings are generally more expensive, require special hull preparation and application procedures and the hull must be cleaned twice a month. However, the life of the paint is not dependent on the life of the toxicant. A durable, nontoxic hull coating could greatly extend the time between haulouts and make up the cost difference.



In order to educate boaters and to track the performance and costs of using nontoxic antifouling coatings, the University of California Sea Grant Extension Program (UC-SGEP), with state/federal funding under the 319h Nonpoint Source Pollution Program, and the San Diego Unified Port District will be conducting demonstrations of nontoxic antifouling coatings during the next two years. They will coordinate their assessments of performance and costs of using nontoxic coatings so a broader range of data can be collected. The UC-SGEP will conduct field days and workshops for boaters, develop a brochure, report to the boating associations, and post information on the internet about the demonstration.

The UC-SGEP will also conduct an economic comparison of traditional and nontoxic coatings for recreational boat bottoms. The UC Davis Center for Pest Management, Research and Education is providing partial funding. The California Legislature is considering a bill (SB 315 – Alpert) that would establish a committee to study and report on how to develop incentives for recreational boaters to use nontoxic, antifouling coatings.

# Assessing the Consequences of the Non-Native Invasive Cordgrass *Spartina alterniflora* in West Coast Estuaries

Edwin Grosholz<sup>1</sup>, Alan Hastings<sup>1</sup>, Don Strong<sup>2</sup>,  
Susan Ustin<sup>3</sup>, David Layton<sup>1</sup>, Lisa Levin<sup>4</sup>, Andy Cohen<sup>5</sup>

- 1) Department of Environmental Science and Policy, UC Davis
- 2) Section of Evolution and Ecology, UC Davis,
- 3) Department of Land, Air, and Water Resources, UC Davis
- 4) Marine Life Research Group, Scripps Institution of Oceanography, UC San Diego
- 5) San Francisco Estuary Institute

Among the most important threats to coastal marine habitats is the invasion of non-native species. In western North America, the invasive Atlantic cordgrass, *Spartina alterniflora* has already begun to change community, food web, and ecosystem processes in coastal bays and estuaries. As this plant invades and overgrows the naturally unvegetated mud flats of west coast bays and estuaries, it can reduce light levels, increase sedimentation rates, and shift primary production away from algal dominance to a detritus-based system with altered nutrient abundance and decomposition rates. Through a new project funded by the NSF Biocomplexity Program, we will develop this integrated approach to understanding the changes brought about by this species including the dynamics of an invasive species, a core mathematical/conceptual model, physical and biological feedbacks, and a careful, justified study of impacts on non-commercial human values. We predict that changes in the base of the food web will propagate throughout the food web as the result of trophic alterations as well as physical and chemical changes brought on by this invasion. In addition, we predict that the *Spartina* invasion will have consequences for endangered plants, birds, and mammals, and other species of concern as well as have impacts on the human commercial, traditional, non-commercial, and esthetic values of Pacific estuaries.

# **Understanding Food Web and Contaminant Effects on Fish Populations: An Interdisciplinary Approach for the Western Center for Estuarine Ecosystem Indicator Research**

William A. Bennett<sup>1</sup>, Susan L. Anderson<sup>2</sup>, Swee J. Teh<sup>3</sup>, and Kathryn M. Kuivila<sup>4</sup>

1) John Muir Institute of the Environment, 2) Bodega Marine Laboratory, UC Davis

3) Department of Anatomy and Physiology, UC Davis, 4) U.S. Geological Survey, Sacramento

We present an interdisciplinary approach for the development of indicators of anthropogenic effects on western estuaries which exemplify the objectives of the new Western Center for Estuarine Ecosystem Indicator Research (WCEEIR) funded as one of the U.S. EPA STAR Estuarine and Great Lakes Programs. Fishes of the San Francisco Estuary have been subjected to rapid changes in the abundance and composition of zooplankton food resources due to invasive species and exposure to pesticides in urban and agricultural run-off. These factors can act alone or synergistically to affect growth rates, year-class success, and future reproductive success. Based on earlier work in which we quantified the mechanisms underlying larval fish "condition" using morphometry and histopathology, we are currently tying in evaluations of fish otoliths to assess growth rate and pattern, as well as genotoxic responses to contaminant exposure for individual larval and juvenile delta smelt, a threatened species. For individual specimens, histopathological analyses of hepatic tissue indicate site-specific abnormalities consistent with a contaminant etiology. Comparison of growth rates and DNA strand breaks as measured by an electrophoretic assay (the Comet Assay) among sites indicates lower growth rates as well as higher percentages of damaged DNA at the sites in which abnormal histological condition was also identified. In addition, water chemistry evaluations indicate elevated, yet sublethal, concentrations of pesticides at the time of specimen collection. Collectively, these integrated studies suggest an approach for quantifying biological indicators that can be translated from individuals to populations, as well as readily incorporated into monitoring programs. In addition to expanding this approach, the WCEEIR will be developing a variety of interdisciplinary approaches, including integrative laboratory and field experiments using wetland plants, amphipods, and gobies at common sites ranging from northern California to Mexico. By focusing heavily on indicator integration with common data management, modeling, and remote sensing, the WCEEIR will provide new and quantitative approaches for evaluating "ecosystem health" in western estuaries.

## **Coho Salmon in California: How Long To Extinction?**

Peter B. Moyle and Ronald M. Yoshiyama. UC Davis

California is the southern end of the range of coho salmon, which is largely coincident with that of coastal rain forests. Historically coho were once found in nearly 600 streams along the California coast, numbering in the hundreds of thousands and contributing to sport and commercial fisheries. A study we published in 1994, using agency data, indicated that about half of historical coho streams had lost their coho populations. It also indicated that there were less than 5,000 wild coho left in the state, along with about 25,000 coho of hatchery origin. Just as remarkable was the poor quality of the data available because so little monitoring of coho populations was being done. Partly as a result of this study, coho salmon in California and southern Oregon were listed by NMFS as a threatened species. The coho fishery was shut down in 1994. Although our study has not yet been repeated, studies on a few streams and anecdotal evidence indicates that coho have continued to decline in abundance. The long-term decline of coho salmon is primarily the result of degradation of spawning and rearing streams, by urbanization and agriculture in the south and by logging and road building in the north. Poor conditions for coho survival in the ocean have apparently contributed to the decline but such conditions in the past have not led to wide-spread extinction. We predict that native wild strains of coho salmon will be extirpated from California within 20 years unless fairly dramatic measures are taken to maintain populations in selected streams. Long-term recovery will depend on restoration of forested watersheds to conditions that will support coho spawning and rearing. With the increasing human population in California, increased pressure on limited land and resources, and lack of fundamental change in public attitudes towards wild systems, we see little reason to be optimistic about the future of coho salmon in California.

## Population Dynamics and Management of Nearshore Fisheries

Louis W. Botsford, Department of Wildlife, Fish, and Conservation Biology, UC Davis

Two recent laws, the MLPA and the MLMA provide vehicles for needed improvements in California's declining fisheries, but even with them, better management depends critically on appropriate information. The development of the red sea urchin fishery over the past 15 years is an example of some of the needs for more information and better understanding. As a new fishery develops the "fishing up" effect provides unrealistic expectations of high catch and catch-per-unit-effort. As that transient effect subsides, there is an attempt to establish size limits and effort levels that provide the maximum long-term yield. As the fishery continues to decline, there is an attempt to prevent recruitment failure. Doing this requires knowing how fishing affects lifetime reproduction, and the minimum level of lifetime reproduction required for that species. Size limits and controls on effort can be used to maintain that minimum. A new method of maintaining minimum lifetime reproduction is the establishment of marine reserves. As we become more aware of the spatial aspect of exploited population's dynamics, we are realizing the need for better information on movement of: 1. larval stages in response to varying ocean currents and biological productivity, 2. juvenile and adult fish as regards reserve boundaries, and 3. fishermen and fisherwomen in response to spatially explicit management. We also need a better understanding of new methods of spatial management, such as reserves and rotating spatial harvest.

## **Seymour Intertidal Monitoring Program: Monitoring the Rocky Intertidal of Monterey Bay by High School Students**

John Pearse, Dawn Osborn, and Christy Roe. UC Santa Cruz.

We are developing site-specific protocols for long-term monitoring of the rocky intertidal of central California to be used by high school students and other volunteers. Current high schools involved are Aptos, Harbor, Watsonville, and the Monterey Academy of Ocean Sciences. Other people, including home-school students and individuals in volunteer groups are also participating. Counts of prominent species in random quadrats within permanent plots at Natural Bridges continue a monitoring program begun in 1972. Vertical transects at Natural Bridges, Soquel Point, and Point Pinos establish zonation patterns that will change if sea level changes.

Abundance and sizes of important species (e.g., owl limpets and sea stars) at all three sites will detect recruitment and mortality patterns. An interactive website with the data will be maintained by the Seymour Marine Discovery Center. Data collected to date at Natural Bridges compare favorably with those collected by professional researchers, giving us confidence that the program will not only introduce students to the rich biota of the rocky intertidal and how to study it, but will provide data that can be used by scientists and resource managers to track long-term changes in this spectacular habitat. Supported by California Sea Grant and the Monterey Bay National Marine Sanctuary.

## Anthropogenic Contaminants in the Sediments of Santa Monica Bay, California

M. I. Venkatesan, Institute of Geophysics and Planetary Physics UCLA

Santa Monica Bay (SMB) is adjacent to the highly urbanized Los Angeles region that receives contaminant inputs from multiple sources including treated municipal wastewaters, storm runoff and atmospheric deposition. The Sanitation Districts also discharged DDTs through submarine outfalls from 1937 to early 1970s in southern California coastal waters. Remobilization of the DDT-contaminated sediments, now buried in deeper strata below the sediment-water interface, could be a significant problem. The current levels of DDTs and their metabolites, polychlorinated biphenyls (PCBs), polycyclic aromatic hydrocarbons (PAHs), linear alkylbenzenes (LABs) and coprostanol were determined in surface sediments of SMB. LABs found in household laundry detergents and coprostanol present in human (and also other mammalian) fecal waste, formed from reduction of cholesterol by intestinal microbes, are considered as sewage tracers. A correlation (or lack of correlation) between such tracers and other contaminants was examined to discern the possible sources of the latter.

Over eighty surface grab sediments were collected from 1997-1999 from the SMB from a water depth of 30 to 100 m. Sediments were extracted with organic solvents and fractionated into compound classes. Target compounds were quantitated by gas chromatography (GC) or GC/mass spectrometry.

The concentration levels, spatial distribution trends and the intercorrelation of various contaminants help resolve their sources to the Bay. For example, sediments near the outfalls are clearly influenced by the continuing wastewater discharge, as indicated by the elevated ...LABs (sewage markers). A negative gradient in their content is generally observed with increasing distance from the source. LABs and PAHs appear to be highly localized near the outfalls at the 30m water depth contour and they co-vary with each other ( $r=0.877$ ), thus confirming their major source in the modern day municipal discharges. Poor to moderate correlation of DDTs and PCBs respectively with the sewage markers suggests that DDTs in the Bay sediments originate from the historic deposits and that PCBs are derived from historic discharges to a much less extent. Further, considering that the modern day effluents from the Hyperion Plant (City of Los Angeles Sanitation Districts) contain DDTs and metabolites below detection levels (effluent loading being  $<0.005$  lb/day), DDTs in the Bay sediments obviously originate from the historic deposits. Apparently, very low levels of historic DDTs are still being chronically remobilized from the sediments near the discharge point from the outfalls termini, especially from the 7-mile sludge disposal site, which was terminated in 1987. Preferential accumulation of DDTs is observed between 60 and 100m water depth and there are hot spots of other contaminants accumulation as well in the Bay at a water depth of 100 meters. Source of these hot spots farther from the outfalls termini will be discussed. Concentration levels of some contaminants will be examined in the light of sediment quality objectives to assess potential adverse biological effects and the Baywide inventories of the contaminants discussed.

# **Hormonal Regulation of Crustacean Growth, Development, Reproduction, And Response to Stress**

Ernest S. Chang Bodega Marine Laboratory UC Davis

Crustaceans comprise an important group of animals in marine environments because of their value for fisheries and aquaculture. In addition, crustaceans are key members of aquatic ecosystems and hence have intrinsic interest in the study of food webs and as invasive species. Much of the crustacean's physiology and biochemistry are regulated by hormones.

I will present an overview of the isolation and characterization of three hormones (or hormone families) important for the growth and development of decapod crustaceans. These hormones include the ecdysteroids (steroid molting hormones), the crustacean hyperglycemic hormone (CHH) neuropeptide family, and the terpenoids methyl farnesoate. I describe work on these hormones using various life stages of the shrimp (*Sicyonia igentis*), the crab (*Cancer magister*), and the lobster (*Homarus americanus*) as the principal models. I will also present recent research on the possible role of CHH in various stress responses.



## Applications of Pantropic Retroviral Vectors in Oyster Disease Research and Control

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Management of shellfish disease requires sensitive tools for diagnosis and pathogen characterization. While tools developed for and applied in mammalian disease research are often quickly adapted for use with shellfish, one conspicuous exception is the failure to generate permanent cell lines from marine shellfish tissue. The absence of cell lines has particularly impeded research on viruses and obligate intracellular bacteria. We are attempting to create cell lines from tissues of the American oyster, *Crassostrea virginica* using a highly efficient method to integrate oncogenes into the genome of cells in primary culture. Heart tissue primary cultures were infected with replication-defective retroviral vector(s) containing viral promoters driving expression of the gene(s) of interest. These retroviral vectors contain the envelope glycoprotein of vesicular stomatitis virus that binds to phospholipid components of the cell membrane, thus allowing entry of the virus particle into a wide range of cell types. Conditions for gene transfer and expression were optimized using a vector construct containing the luciferase reporter gene, followed by application of retroviral vectors encoding the SV40 large T antigen and ras oncogenes. Results from these studies indicated that our in vitro culture conditions were suboptimal and we conducted a thorough analysis of the chemical composition of oyster hemolymph. Another large barrier hampering oyster disease research is the inability to stably transfer genes into embryos or gametes, which is essential for the genetic engineering of disease resistance. Studies currently in progress employ pantropic retroviral vectors to attempt introduction and expression of the luciferase gene in oysters via infection of eggs or sperm prior to fertilization.

# Geographic Distribution of Withering Syndrome and Development of an Oral Administration of Oxytetracycline to Control Associated Losses in Cultured Red Abalone *Haliotis rufescens*

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Withering syndrome (WS) is a chronic and fatal disease of wild and cultured abalone, *Haliotis spp.* causing significant losses to commercial farmers and wild populations. The etiological agent of WS is an obligate intracellular bacterium (RLP), recently described as "*Candidatus Xenohaliotis californiensis*". There is great concern over the introduction or presence of this bacterium on the north coast, where the last remaining health abalone (*Haliotis rufescens*) populations remain. It is unknown whether the bacterium is endemic or if it was inadvertently introduced into north coast red abalone populations by out-planting of cultured red abalone, or release from commercial abalone farms or research laboratories (prior to the identification of the RLP). In this study, eight point source locations and five reference locations north of Carmel were examined for the RLP. Point source locations (PSL) were chosen by their proximity to either a commercial abalone farm, out-plant location or research laboratory. Reference locations were not associated with any point sources. Up to 60 abalone were randomly collected at each location and examined for clinical and microscopic signs of WS. This research revealed "*Candidatus Xenohaliotis californiensis*" and WS north to San Francisco, at both PSL and RL. In addition, the bacterium has been observed at two PSL north of San Francisco, suggesting a possible link between anthropogenic influences and the present distribution of this bacterium.

Withering syndrome has resulted in devastating losses of cultured abalone during warm water events. In order to assess the controlled application of a therapeutant, we investigated the ability of several antibiotics to reduce RLP infection intensities. Intramuscular injections of oxytetracycline effectively alleviated RLP infections and reduced or prevented associated losses. In order to control losses due to this disease in abalone culture facilities, we are assessing the development of an oral administration of oxytetracycline. WS-positive, red abalone were placed into tanks (6 control and 6 treatments) and held at ~15 °C. Experimental animals were fed a diet containing oxytetracycline for 2 wk, while control animals were fed kelp. After the treatment, an El Nino event was simulated for two months and then animals were returned to ambient conditions. Two weeks after the final feeding of the medicated diet, five abalone were sampled from each tank for histology and drug residue analysis. In addition, moribund animals from various tanks were sampled after 3 months. We have observed significant reductions in the intensity of RLP infections in both the postesophagus and digestive gland 2 wk and 4 mo after a 2 wk treatment ( $p < 0.05$  and  $p < 0.001$ , respectively). Losses of treated abalone (6.45%) were also significantly less than those in the unmedicated, control treatment (36.30%;  $p < 0.0001$ ). The yield of treated animals exceeded those of control animals by 48% 10.6 months after a single treatment. Tissue residue levels were below the 2 ppm FDA allowable level by day 15 after medication. All trials were conducted under the guidance of the FDA/CVM through their Aquaculture Investigational New Animal Drug program (INAD #9332).

## Protozoal Brain Infections in California Sea Otters and Harbor Seals: A Case of Pathogen Pollution?

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Fatal protozoal brain infections were first recognized in southern sea otters (*Enhydra lutris nereis*) and pacific harbor seals (*Phoca vitulina richardsi*) in California in 1992. Two distinct species of parasites have been implicated in these infections. The first parasite is similar or identical to *Toxoplasma gondii*. Molecular, antigenic and ultrastructural characterization of isolates from sea otters and a harbor seal reveal no significant differences between the *T. gondii*-like parasites obtained from sea otters and seals, and those isolated from terrestrial animals and humans. Interestingly, *T. gondii* is a significant human pathogen, whose only recognized definitive, or egg-shedding hosts are wild and domestic felids. The second parasite isolated from sea otters and harbor seals with fatal encephalitis is molecularly, antigenically and ultrastructurally indistinguishable from *Sarcocystis neurona*. *Sarcocystis neurona* is an important pathogen of horses. To date the only recognized definitive host for *S. neurona* is the opossum (*Didelphis virginiana*), which was introduced to central California about 100 years ago. Frequent infections of coastal marine mammals by these parasites, whose only recognized definitive hosts are terrestrial mammals, suggest flow of protozoal pathogens from the terrestrial to the nearshore marine environment. We are currently investigating routes by which these pathogenic protozoa may be efficiently concentrated and transmitted to the threatened southern sea otter in California. This research may have important implications for human health, as otters and humans share many of the same coastal marine prey species.

# Natural Peptide Antibiotics from Marine Invertebrates

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For over half a century, antibiotics that could cure almost any bacterial infection have been no further away than the nearest hospital or pharmacy. However, the past decade has witnessed the increasing resistance of many microbes to antibiotics. As a result, new sources and new types of antibiotics are urgently needed. In recent years, antimicrobial peptides have become recognized as fundamental components of the innate immune system of animals, ranging from protozoans to humans. Perhaps because invertebrates lack adaptive, lymphocyte-based immune responses, their host defense cells produce a wide variety of these peptides, many of which have unusual structures or properties.

Although tunicates ("sea squirts") are small and not especially exciting to look at, they are protochordates and belong to the same lineage as humans and other vertebrates. Their blood cells (hemocytes), like ours, have numerous cytoplasmic organelles ("granules") that contain antimicrobial peptides. We have worked with hemocytes from two species of tunicates, *Styela clava* and *Styela plicata*, both of which are abundant in California's coastal waters. From *S. clava* hemocytes, we purified two families of antimicrobial peptides: styelins and clavans. Styelins A, B and D had 32 amino acid residues and were highly effective against a panel of gram negative and gram positive human bacterial pathogens, and marine bacteria, such as *Psychrobacter immobilis* and *Planococcus citreus*. One of these peptides, Styelin D, showed unusually extensive post-translational modifications, with two novel amino acids (dihydroxyarginine and dihydroxylysine) and two very uncommon ones (6-bromotryptophan and 3,4-dihydroxyphenylalanine). By comparing native Styelin D with its (synthetic) unmodified counterpart, we found that these modifications enhanced activity at low pH and/or high salinity. Styelins appeared to be homologous to cecropins, a family of antimicrobial peptides widely distributed in insects. We also found that *S. clava* hemocytes contained a family of four alpha-helical antimicrobial peptides that were named clavans A, B, C and D. These histidine-rich peptides, which contained 23 amino acid residues and were C-terminally amidated, were most active at an acidic pH.

The leukocytes of *S. plicata* contain an unusual octapeptide, plicatamide, whose sequence was Phe-Phe-His-Leu-His-Phe-His-deDDO, where deDDO signifies decarboxy-(E)-alpha,beta-dehydro-3, 4-dihydroxyphenylalanine (Tincu, Craig and Taylor, BBRC 270:421-4, 2000). Plicatamide induced a massive loss of potassium from susceptible *E. coli*, without causing a general increase in membrane permeability. We are constructing synthetic counterparts of plicatamide to identify the function of its unusual C-terminal residue, and to aid in structure:activity studies. Because of its small size and unusual mechanism of action, plicatamide provides an attractive template for designing novel antimicrobial peptides.

## The Physiology of Wound Plug Formation in *Dasycladus vermicularis*

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*Dasycladus vermicularis* is a siphonous green algae that contains a natural product (3,6,7-trihydroxycoumarin) unique to the marine environment. The genus *Dasycladus* belongs to the order Dasycladales, a group of primitive marine unicellular green algae that are members of the kingdom Protocista. Despite the fact that a large number of chemically diverse coumarins are available from higher plant extracts, it is worth noting that *D. vermicularis* represents a living fossil and has been shown to exist over 570 million years ago. It should be noted that 3,6,7-trihydroxycoumarin has not been reported in any organisms (plant or otherwise) except in *D. vermicularis*. Reports indicate that the biosynthetic machinery differs from that of terrestrial plants [which are regarded as infantile on geologic time scales when compared to many marine protists] and therefore the elucidation of biosynthetic intermediates would yield a potentially wide array of new coumarin compounds. In this study, we propose to investigate the endogenous roles of 3,6,7-trihydroxycoumarin and its biosynthetic precursors within the host organism itself. This study is aimed at analyzing the unique process of wound repair within *D. vermicularis*. Injury and wounding to multicellular organisms may not be fatal because of the gradual formation of protective and regenerative tissues adjacent to the wound. However, an injured, unicellular organism such as *D. vermicularis* does not have this option and its survival clearly depends on the instant activation of a wound repair mechanism. When the siphon of *Dasycladus vermicularis* is injured, a complex, molecular response is triggered which encompasses an instant repair of the damaged area followed by a transient sealing of the wound by a plug. These plugs appear as gelatinous protrusions appearing from the wounded area.

The formation of wound plugs, in selected chlorophytes, has been documented in several preliminary studies. Unfortunately, the availability of fine structural and histochemical data does not contribute to our understanding of the various biochemical and physiological components of plug formation. Furthermore, the biochemical basis of plug formation is still poorly understood. Depending on the nature of the precursor material, one such mechanism might be based on protein-protein interaction, a second mechanism may involve interactions between polysaccharides, and yet a third may involve proteins and polysaccharides.

Cross linkage of polymeric substrates by low molecular weight enzyme mediated cross-linkers has been hypothesized as a possible mechanism of plug formation and hardening in siphonous green algae. It is believed that in the majority of siphonous green algae, either the plug precursors or the plugs contain peroxidase. There are many examples in higher plants describing a peroxidase/phenylpropane interaction leading to cross-linking that involves protein or polysaccharides. The chemical structures of simple coumarins are very similar to these phenylpropane molecules. The assumption that a similar mechanism may also be operating in siphonous green algae has become of interest after the discovery of unique hydroxy coumarin analogs in the dasycladalean algae.

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