Merck/AAAS Undergraduate Research Program 2008

Loyola Marymount University has received an award from the Merck/AAAS Undergraduate Research Program* to support five Research Associates for ten weeks during the summer of 2008. Each Research Associate will work with at least two faculty advisors on a interdisciplinary project that spans biology and chemistry, field and laboratory research and centers on the Ballona Wetlands.

The summer program will extend from May 19 through July 25. Research Associates will receive a stipend of \$3000 and housing will be provided, if needed. In addition, each Research Associate will be expected to present results at an LMU departmental seminar and at a regional or national conference. Research associates are encouraged to continue their research in the 2008-2009 academic year by enrolling for independent research credit with their advisor.

If you wish to apply, please submit the attached application form to the Biology office (Seaver 204) by **4:30PM on Friday, February 22.**

These project descriptions are based on the research abstracts prepared by Merck/AAAS Research Associates during the first two years of the program. The 2008 Research Associates will continue work on these projects.

Project 1: Developing *Salicornia virginica* as a Bio-monitor for Heavy Metals (Dr. Pippa Drennan, Dr. Jim Landry)

The coastal Ballona wetlands are a part of the highly urbanized Los Angeles watershed. Studies performed on the water quality of the channel feeding the wetlands have shown high levels of heavy metals. The widely distributed halophyte *Salicornia virginica* (commonly known as pickle weed) was used to establish trends in heavy metal pollution for Ballona wetlands. It is hypothesized that *S. virginica* in a contaminated wetland will take up heavy metals along with the salts. Plant tips and soil samples, 85 for each, were collected from 16 sites throughout the Ballona wetlands. Five plant tips were taken from the Sunset Beach Back Bay for comparison. The methods used were oven drying to establish wet and dry weights for water content, flame photometry to determine sodium and potassium levels in the tissues, and atomic absorption spectroscopy for zinc (flame analysis) and cadmium (graphite furnace) levels. The amount of each metal in the plants was correlated with the amount of metal in the soil. Correlations were positive and significant for cadmium and zinc. With *S. virginica* established as a reliable biomonitor, areas of constant elevated cadmium and zinc pollution were identified.

For consideration, individuals interested in a fellowship must interview with Dr. Drennan (pdrennan@lmu.edu) or Dr. Landry (jlandry@lmu.edu).

Project 2: Biomagnification of Wetland Contaminants in Garden Spiders (Dr. Martin Ramirez, Dr. Jim Landry, Dr. Jeremy McCallum)

The purpose of this study was to examine spatial heterogeneity in the heavy metal accumulation in garden spiders, *Argiope trifasciata*, and to assess the impact of heavy metal loads on spider fitness. During October 2006, adult females were collected from three sites at the Ballona Wetlands, a highly degraded urban wetland in Los Angeles. Spiders were measured (carapace width, mm) and weighed (mg) prior to analysis with atomic absorption spectroscopy to yield whole-body metal concentrations (Cd, Cr, Cu, Pb, Zn). Spider excretia was analyzed via HPLC. Size and weight values were natural logarithm transformed and the procedures of Jakob et al. (1996) were used to generate the residual index (RI, the residuals of body mass on body size), a non-destructive measure of body condition, for each spider. ANOVA tests were used to assess spatial heterogeneity in whole-body metal concentrations among the three sites. Of the five metals, cadmium and chromium were homogeneous among sites, whereas copper, lead, and zinc varied significantly by site with different patterns in each case. Regression analysis for each

metal using various groupings of the samples showed significantly negative relationships between dry metal concentrations and spider bodily parameters in four cases (Cd: size, weight; Cd, Cu: residual index). Further analyses must be completed on the HPLC excretia samples. As major invertebrate predators, spiders may bioaccumulate materials from the bodies of their prey. Our results are consistent with such a possibility and suggest that web-building spiders residing in polluted areas may be compromised in terms of fitness.

For consideration, individuals interested in a fellowship must interview with Dr. Ramirez (mramirez@lmu.edu), Dr. Landry (jlandry@lmu.edu), or Dr. McCallum (jmccallu@lmu.edu).

Project 3: Freely Dissolved Concentrations of Selected PCBs, PAHs, and Chlorinated Pesticides in Ballona Wetland and Creek (Dr. Rachel Adams, Dr. Gary Kuleck)

The Ballona Wetlands are designated as an ecological preserve with environmental assessment and restoration currently underway. The Ballona Creek Estuary is a contaminated body of water for which the Total Maximum Daily Loads (TMDLs) for several hydrophobic organic contaminants (HOCs) have already been established. However, traditional methods for dissolved chemical measurements (e.g., liquid-liquid extraction) are difficult to use for HOCs at the current TMDL-regulated levels. In this study, passive samplers (polyethylene devices, PEDs) were deployed in the wetlands in June '07 to measure the dissolved concentrations of selected TMDL-regulated contaminants: polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), and chlorinated pesticides. These newly-developed passive samplers allow for the measurement of contaminants at trace levels. The PEDs were deployed in metal paint buckets fastened to stakes in the wetland channels. These buckets ensured that the PEDs were submerged and sampling water throughout the experiment. The PEDs were deployed at four locations in the wetland and at one location in the Creek for 14 days. Blank PEDs were used to correct for any contamination during deployment and recovery. Upon recovery, the PEDs were cleaned and extracted. The extracts were analyzed using a gas chromatograph-mass spectrometer (GC-MS). The dissolved chemical concentrations will be calculated using water equilibrium partitioning coefficients and corrected for cases in which equilibrium is not reached. The measurements will allow scientists and regulators to assess the current HOC levels in the Wetland and Estuary so that these HOCs may be evaluated with respect to the current TMDLs.

For consideration, individuals interested in a fellowship must interview with Dr. Adams (radams@lmu.edu) or Dr. Kuleck (gkuleck@lmu.edu).

Project 4: Survey of Microorganisms in the Ballona Wetlands (Dr. John Dorsey, Dr. Gary Kuleck)

The objective of the research was to establish a method to be able to identify ecological samples of microbes through Automated Ribosomal Intergenic Spacer Analysis. Initial experimentation involved instituting a method of operation to be able to identify a specific genetic fingerprint for each clade or species. This was done through repeated isolation of bacteria via plating to obtain one colony which then underwent PCR to amplify the 16S, ITS, and a part of the 23S region. The PCR product then underwent gel electrophoresis and a series of bands were obtained. Alongside this experiment Vitek analysis was performed on the isolates to establish a correlation between the bands and the species. In most cases a 95% or higher confidence was attributed to the identification of the bacteria. Through repeated testing and a gradient PCR (to ensure actuality of bands) unique "fingerprints" were obtained for various microbes. These "fingerprints", based on initial testing, seem to be species specific. These findings allow the continuation of the research unto uncultured ecological samples. A method of filtration needed to be established prior to PCR so that only bacteria were being identified. This involved a two step vacuum filtration using durapore and a/e glass filters to obtained workable samples. These samples were then either frozen for later experimentation or DNA was extracted and then amplified by PCR for electrophoresis. A series of bands is the target at which point the

bands will be integrated into a library which will be expanded upon. All water samples are from the Ballona wetlands.

For consideration, individuals interested in a fellowship must interview with Dr. Dorsey (jdorsey@lmu.edu) or Dr. Kuleck (gkuleck@lmu.edu).

Project 5: Identifying Soil Bacteria and Biochemical Pathways in the Ballona Wetlands for the Bioremediation of Organic Pollutants and Analysis of Organic Compounds in the Ballona Atmosphere (Dr. Kam Dahlquist, Dr. Lambert Doezema, Dr. David Moffet, Dr. Carl Urbinati)

The Ballona Wetlands in Los Angeles County are contaminated with organic pollutants including polychlorinated biphenyls (PCBs), polyaromatic hydrocarbons (PAHs), and single-ring aromatics (e.g. toluene) from urban run-off. Initially, to determine whether biochemical pathways exist in the wetlands to degrade toluene we attempted to enrich bacteria from the soil that could metabolize toluene. We obtained a single, pure environmental isolate from media with citrate as a sole carbon source but not from media with toluene as a sole carbon source. Genomic DNA was isolated from the environmental strain and a variable region of the 16S rRNA gene was subcloned. DNA sequencing identified the environmental isolate as *Pseudomonas putida* F1. *Pseudomonas* species, including *Pseudomonas putida* F1, are known to degrade toluene. Currently we are attempting to clone the todC1 gene which encodes one subunit of toluene dioxygenase, the first enzyme in the toluene degradation pathway from the isolated Pseudomonas putida F1. In order to assess the diversity of soil bacteria in the wetlands we isolated genomic DNA from soil samples, amplified a variable region of the 16S rRNA gene and constructed sub-genomic libraries. To date, 51 clones from one library have been sequenced and identified by BLAST database searching. Thus far, the most abundant bacterial species identified is Pseudomonas putida F1; other species identified include Actinobacteria and Sphingomonas spp. Additionally, we are using Length Hetereogeneity-PCR (LH-PCR) to assess diversity in the soil samples. LH-PCR results will be compared to results from direct sequencing of 16S rDNA sub-genomic library clones. To assess volatile hydrocarbons, chemical precursors to ozone, in the Ballona atmosphere, ambient air samples will be collected in evacuated 2 liter stainless steel canisters and analyzed for hydrocarbons using GC-FID.

For consideration, individuals interested in a fellowship must interview with Dr. Dahlquist (kdahlquist@lmu.edu), Dr. Lambert Doezema (ldoezema@lmu.edu) Dr. David Moffet (dmoffet@lmu.edu), or Dr. Urbinati (curbinati@lmu.edu).

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MERCK/AAAS RESEARCH ASSOCIATES APPLICATION FORM RETURN BY 4:30 PM, FRIDAY, FEBRUARY 22*

*Please fill out this application form and return it to the Biology Office (Seaver 204). Use additional pages if necessary. We will notify those who are selected the week of March 10.

Name	Class
Student ID #	
Address	Phone
E-mail address	
Do you need on-campus housing for the summer?	
Major (or prospective major)	

CHOICE OF RESEARCH PROJECT: Please indicate with a number in the blank to the left of the project title, your first, second and third choice. Details of the projects are found on pages 1 - 3 of this document.

Project 1: Developing Salicornia virginica as a Bio-monitor for Heavy Metals (Dr. Pippa Drennan, Dr. Jim Landry)

Project 2: Biomagnification of Wetland Contaminants in Garden Spiders (Dr. Martin Ramirez, Dr. Jim Landry, Dr. Jeremy McCallum)

Project 3: Freely Dissolved Concentrations of Selected PCBs, PAHs, and Chlorinated Pesticides in Ballona Wetland and Creek (Dr. Rachel Adams, Dr. Gary Kuleck)

Project 4: Survey of Microorganisms in the Ballona Wetlands (Dr. John Dorsey and Dr. Gary Kuleck)

Project 5: Identifying Soil Bacteria and Biochemical Pathways in the Ballona Wetlands for the Bioremediation of Organic Pollutants and Analysis of Organic Compounds in the Ballona Atmosphere (Dr. Kam Dahlquist, Dr. Lambert Doezema, Dr. David Moffet, Dr. Carl Urbinati)

Use as much space as you like in answering the questions below; attach additional pages if necessary

Please list all courses taken in Biology and Chemistry and any other science, engineering, or math courses that are relevant to your application for a MERCK/AAAS Fellowship.

Please briefly describe below any previous research experiences you have had.

Please state what you expect to gain from your experience as a Merck/AAAS Research Associate.