Final Progress Report for Research Projects Funded by Health Research Grants

Instructions: Please complete all of the items as instructed. Do not delete instructions. Do not leave any items blank; responses must be provided for all items. If your response to an item is "None", please specify "None" as your response. "Not applicable" is not an acceptable response for any of the items. There is no limit to the length of your response to any question. Responses should be single-spaced, no smaller than 12-point type. The report **must be completed using MS Word**. Submitted reports must be Word documents; they should not be converted to pdf format. Questions? Contact Health Research Program staff at 717-783-2548.

1. **Grantee Institution:** Temple University – of the Commonwealth System of Higher Education

2. **Reporting Period (start and end date of grant award period):** 01/1/2009 -12/31/2012

3. **Grant Contact Person (First Name, M.I., Last Name, Degrees):** Germaine Calicat, MLA

4. **Grant Contact Person's Telephone Number:** 215.204.7655

5. **Grant SAP Number:** 4100047651

6. **Project Number and Title of Research Project:** Project 29 **-** Biology Department Core Facility Infrastructure

7.

8. **Start and End Date of Research Project:** 12/1/2010-7/31/2011

9. **Name of Principal Investigator for the Research Project:** Joel Sheffield, Ph.D.

10. **Research Project Expenses**.

9(A) Please provide the total amount of health research grant funds spent on this project for the entire duration of the grant, including indirect costs and any interest earned that was spent:

$ 202,100.00

9(B) Provide the last names (include first initial if multiple individuals with the same last name are listed) of **all** persons who worked on this research project and were supported with health research funds. Include position titles (Principal Investigator, Graduate Assistant, Post-doctoral Fellow, etc.), percent of effort on project and total health research funds expended for the position. For multiple year projects, if percent of effort varied from year to year, report in the % of Effort column the effort by year 1, 2, 3, etc. of the project (x% Yr 1; z% Yr 2-3).

Last Name	Position Title	% of Effort on Project	Cost
None			

9(C) Provide the names of **all** persons who worked on this research project, but who *were not* supported with health research funds. Include position titles (Research Assistant, Administrative Assistant, etc.) and percent of effort on project. For multiple year projects, if percent of effort varied from year to year, report in the % of Effort column the effort by year 1, 2, 3, etc. of the project (x% Yr 1; z% Yr 2-3).

Last Name	Position Title	% of Effort on Project
Sheffield, Joel	Professor	5

9(D) Provide a list of **all** scientific equipment purchased as part of this research grant, a short description of the value (benefit) derived by the institution from this equipment, and the cost of the equipment.

Type of Scientific Equipment	Value Derived	Cost
Scanning Electron Microscope	Research enhancement in Biology, Physics, Engineering, Chemistry, Bioengineering	$130,011.19
Spectrofluorimeter	Research Enhancement in Biochemistry	$36,307.00
Glassware Washer	Facilitation of Research in Biology, Biochemistry	$31,675.00
Ice Machine	Facilitation of Research in Biology, Biochemistry	$4,567.17

10. Co-funding of Research Project during Health Research Grant Award Period. Did this research project receive funding from any other source during the project period when it was supported by the health research grant?

Yes_X_____ No_____

If yes, please indicate the source and amount of other funds:

University Funds. $179,762.00

11. Leveraging of Additional Funds

11(A) As a result of the health research funds provided for this research project, were you able to apply for and/or obtain funding from other sources to continue or expand the research?

Yes_____ No__x_____

If yes, please list the applications submitted (column A), the funding agency (National Institutes of Health—NIH, or other source in column B), the month and year when the application was submitted (column C), and the amount of funds requested (column D). If you have received a notice that the grant will be funded, please indicate the amount of funds to be awarded (column E). If the grant was not funded, insert "not funded" in column E.

Do not include funding from your own institution or from CURE (tobacco settlement funds). Do not include grants submitted prior to the start date of the grant as shown in Question 2. If you list grants submitted within 1-6 months of the start date of this grant, add a statement below the table indicating how the data/results from this project were used to secure that grant.

A. Title of research project on grant application	B. Funding agency (check those that apply)	C. Month and Year Submitted	D. Amount of funds requested:	E. Amount of funds to be awarded:
None	☐NIH ☐ Other federal (specify:_____) ☐ Nonfederal source (specify:_)		$	$

11(B) Are you planning to apply for additional funding in the future to continue or expand the research?

Yes__x_____ No_____

If yes, please describe your plans:

Individual investigators who use the renovated facilities will be applying for research grants.

12. Future of Research Project. What are the future plans for this research project?

The facilities improvements have allowed us to hire new faculty and expand our contribution. These faculty have, in turn, attracted other faculty, so the department has been able to expand significantly. We expect this process to continue. The equipment that was purchased will become part of the departmental infrastructure, to be used as needed by departmental and university researchers and students.

13. New Investigator Training and Development. Did students participate in project supported internships or graduate or post-graduate training for at least one semester or one summer?

Yes_____ No__x_____

If yes, how many students? Please specify in the tables below:

	Undergraduate	Masters	Pre-doc	Post-doc
Male				
Female				
Unknown				
Total				

	Undergraduate	Masters	Pre-doc	Post-doc
Hispanic				
Non-Hispanic				
Unknown				
Total				

	Undergraduate	Masters	Pre-doc	Post-doc
White				
Black				
Asian				
Other				
Unknown				
Total				

14. **Recruitment of Out-of–State Researchers**. Did you bring researchers into Pennsylvania to carry out this research project?

    Yes_____ No___x_____

    If yes, please list the name and degree of each researcher and his/her previous affiliation:

15. **Impact on Research Capacity and Quality**. Did the health research project enhance the quality and/or capacity of research at your institution?

    Yes___x_____ No_____

    If yes, describe how improvements in infrastructure, the addition of new investigators, and other resources have led to more and better research.

    As described below (section 17), we carried out extensive renovations in laboratories on the northeast wing of the second floor, and the south east and southwest wings of the third floor of Biology-Life Sciences Building. As a result, we were able to attract two new faculty members who have instituted research programs in genomics and biomechanics that have broadened our coverage of the field of Biology. We have also been able to install additional

equipment that supports their research. Thus, it was possible to install a new glassware washer and ice machine have become essential components of our infrastructure, providing clean glassware, and sufficient ice for experiments for many faculty, students and postdoctoral fellows. We were also able to install a scanning electron microscope and spectrofluorimeter, which have been used by a number of graduate students, doctoral students, postdocs and faculty in the department of Biology and department of Chemistry. In addition, the spectrofluorimeter was used for instruction in a laboratory course that is part of the Professional Science Masters Program in Biotechnology.

16. Collaboration, business and community involvement.

16(A) Did the health research funds lead to collaboration with research partners outside of your institution (e.g., entire university, entire hospital system)?

Yes__x_____ No_____

If yes, please describe the collaborations:

Since its installation the spectrofluorimeterhas been used to support a new research project involving the Department of Biology at Temple University, and CRO-Aviano Cancer Research Center in Aviano, Italy. The Principal Investigators of the collaborative project "Molecular Nanoarrays for Life Science Applications" are Allen W. Nicholson (Department of Biology, Temple University) and Matteo Castronovo (CRO-Aviano), who also holds an adjunct appointment in the Department of Biology. The results of experiments using this equipment item are forming the basis of an NIH grant application to be submitted in October 2013.

16(B) Did the research project result in commercial development of any research products?

Yes_____ No___x_____

If yes, please describe commercial development activities that resulted from the research project:

16(C) Did the research lead to new involvement with the community?

Yes___x_____ No_____

If yes, please describe involvement with community groups that resulted from the research project:

Some of the images that were obtained with the scanning electron microscope were exhibited at an art exhibit at a local arts organization.

17. Progress in Achieving Research Goals, Objectives and Aims.

List the project goals, objectives and specific aims (as contained in the grant agreement). Summarize the progress made in achieving these goals, objectives and aims for the period that the project was funded (i.e., from project start date through end date). Indicate whether or not each goal/objective/aim was achieved; if something was not achieved, note the reasons why. Describe the methods used. If changes were made to the research goals/objectives/aims, methods, design or timeline since the original grant application was submitted, please describe the changes. Provide detailed results of the project. Include evidence of the data that was generated and analyzed, and provide tables, graphs, and figures of the data. List published abstracts, poster presentations and scientific meeting presentations at the end of the summary of progress; peer-reviewed publications should be listed under item 20.

This response should be a DETAILED report of the methods and findings. It is not sufficient to state that the work was completed. Insufficient information may result in an unfavorable performance review, which may jeopardize future funding. If research findings are pending publication you must still include enough detail for the expert peer reviewers to evaluate the progress during the course of the project.

Health research grants funded under the Tobacco Settlement Act will be evaluated via a performance review by an expert panel of researchers and clinicians who will assess project work using this Final Progress Report, all project Annual Reports and the project's strategic plan. After the final performance review of each project is complete, approximately 12-16 months after the end of the grant, this Final Progress Report, as well as the Final Performance Review Report containing the comments of the expert review panel, and the grantee's written response to the Final Performance Review Report, will be posted on the CURE Web site.

There is no limit to the length of your response. Responses must be single-spaced below, no smaller than 12-point type. If you cut and paste text from a publication, be sure symbols print properly, e.g., the Greek symbol for alpha (α) and beta (ß) should not print as boxes (□) and include the appropriate citation(s). DO NOT DELETE THESE INSTRUCTIONS.

Project Overview

The objective of the infrastructure project was renovation of space and installation of equipment in the Department of Biology at Temple University, which is housed in Biology-Life Sciences Building. Renovation involved removal of old, outdated equipment, demolition of selected existing interior walls, removing existing sinks and plumbing, constructing and finishing new interior walls, installing new, additional electrical power capacity, internet drops, distilled and regular water supply lines, drain lines, ventilation, and built-in tables/shelving for equipment. The original project included purchase and installation of a scanning electron microscope (SEM), 40X plan Apo lens, tabletop centrifuge, glassware washer, and ice maker.

The outcome of the proposed project is to provide the physical space and related services

(electric, plumbing, ventilation, internet connectivity) necessary to accommodate modern research equipment necessary to enhance the capacity of Temple University researchers to conduct a wide range of biological research involving biochemical analysis. This includes research related to the cause and treatment of disease, species-specific reproductive innovations, and neural and cardiac development.

As described in section 15, the total cost of the project, overall, was $379,762, of which $200,00 was provided by the Health Research Grant Award, and the balance from University funds.

Physical Renovation:

Extensive renovation was carried out on the Northwest corridor of the second floor, and the Southeast and Southwest corridors of the third floor of Biology-Life Sciences Building. On the second floor, two laboratories, of total square footage of 2385 sq.ft., were renovated by removal of casework; refurbishing a Fume Hood; replacing plumbing fixtures; installing new casework, flooring and electrical outlets, modified lighting and HVAC to handle new laboratory conditions. On the third floor, one general purpose resource room (850 sq.ft.) and a microscopy laboratory (850 sq.ft.) were renovated by replacing water and steam lines, removing sinks and cabinets, revising electrical service, including connection to the building's emergency circuitry, installation of new internet wiring, installation of shelving, and painting. All of this work was completed on time.

As a result of the physical renovations, we were able to attract two new faculty members who have instituted research programs in genomics and biomechanics that have broadened our coverage of the field of Biology. We have also been able to install additional equipment that supports their research. Thus, it was possible to install a new glassware washer and ice machine have become essential components of our infrastructure, providing clean glassware, and sufficient ice for experiments for many faculty, students and postdoctoral fellows. We were also able to install a scanning electron microscope and spectrofluorimeter, which have been used by a number of graduate students, doctoral students, postdocs and faculty.

Equipment Purchases

We were able to acquire and install specific pieces of equipment that would be used as part of central facilities for the Department of Biology as well as other departments within the College of Science and Technology. We chose to install the following facilities:

1. A Spectrofluorimeter
2. An Ice Machine
3. A Glassware Washer
4. A Desk-Top Scanning Electron microscope.

All instruments have been delivered, and all have been installed. The outline below indicates the specific machines and the additional physical modifications that were necessary for their installation.

Ice Machine:
Scotsman Model number AFE325AS1B
SN 09051320012485
Remove old machine
Install new plumbing

Spectrofluorimeter
Horiba Fluoromax 4 Spectrofluorometer
no major infrastructure improvements needed

Glassware Washer
Lancer Model number 1300 LX
SN 1C033982
Disconnect and remove old machine
Install new 240 V supply
Install new hot water line
Install new cold water line
Install new demineralized water line

Scanning Electron Microscope- Installed, July, 2011
SEM - Agilent 8500 SEM
Removal of sink and cabinet
Removal of air, gas supplies, and pipes
Installation of Electric Receptacles, two connected to the emergency circuit
installation of desktop
painting walls

Uses of the Equipment

1. The glassware washer and ice machine have become essential resources for various components of the research of active members of the Biology department. Readily available ice is necessary for many of the biochemical experiments, to control enzyme activity during purifications and direct experiments. The glassware washer is necessary to maintain proper purity of the glassware that is used in the laboratories. The researchers who benefit from these facilities include:

Balciunas, Darius. Assistant Professor. Research Interests: zebrafish developmental genetics, genomic tools, regeneration
Chang, Frank, Professor. Research Interests: biochemistry, proteomics, cancer
Cordes, Erik Assistant Professor. Research Interests: ecology, genetics/genomics, deep-sea ecosystems
Feitelson, Mark. Professor. Research Interests: pathogenesis, hepatitis B/C, hepatocellular carcinoma.
Freestone, Amy. Assistant Professor. Research Interests: community ecology, macroecology, conservation biology.
Giordano, Antonio, Professor. Director of Sbarro Health Research Organization. Research Interests: cancer, diabetes, cardiovascular disease.

Gruberg, Edward, Professor. Research Interests: behavior and physiology of perception, neuroscience of vision.
Habas, Raymond, Professor. Research Interests: Wnt signaling pathway, embryogenesis, tumorigenesis.
*Hsieh, S. Tonia, Assistant Professor. Research Interests: biomechanics, evolutionary morphology
Klein, Michael, Carnell Professor of Science. Dean, College of Science & Technology, Research Interests: molecular dynamics theory, simulation, modeling.
*Kulathinal, Rob, Assistant Professor. Research Interests: evolutionary genetics, genomics, speciation theory.
Nicholson, Allen, Professor and Department Chair. Research Interests: ribonucleases, RNA processing, gene regulation, nanobiotechnology and biomarker detection.
Palter, Karen, Associate Professor. Research Interests: Drosophila development, sialylation, glycosylation pathway.
Sanders, Robert, Professor. Research Interests: ecology of marine/freshwater plankton, polar ecology, symbiosis.
Sheffield, Joel, Professor. Research Interests: retinal development, immunohistochemistry, imaging, microscopy.
Tanaka, Jacqueline, Associate Professor. Research Interests: structure-function of cyclic nucleotide-gated ion channels.
Waring, Richard, Associate Professor. Research Interests: RNA processing, ribozymes, RNA-protein complexes,
Yang, Weidong, Associate Professor. Research Interests: cell biology, biophysics, biochemistry, nanotechnology, imaging.
Castronovo, Matteo, Adjunct Assistant Professor. Research Interests: Nanobiotechnology, nanomedicine, biomarker detection.
Scoles, Giacinto, Distinguished Adjunct Distinguished Professor. Research Interests: nanomedicine, nanobiotechnology, biomarker detection.
Sewall, Brent J., Assistant Professor in Biology (Research). Research Interests: conservation biology, community ecology, behavioral ecology

*Drs. Hsieh and Kulathinal are the primary beneficiaries of the laboratory renovations on the second floor.

2. Spectrofluorimeter

a. Research Activities involving the Spectrofluorimeter

The Horiba Fluoromax-4 Spectrofluorimeter is housed in the Nanobiotechnology facility, Biology-Life Sciences building, Room 449. Since its installation it has been used to support a new research project involving the Department of Biology at Temple, and CRO-Aviano Cancer Research Center in Aviano, Italy. The Principal Investigators of the collaborative project "Molecular Nanoarrays for Life Science Applications" are Allen W. Nicholson (Department of Biology, Temple University) and Matteo Castronovo (CRO-Aviano). Dr. Giacinto Scoles is a secondary collaborator. This project seeks to develop novel nucleic acid (DNA, RNA) nanoarrays that can function as disease biomarker sensors at the single-cell level. An NIH RO3 application was written *("Detection, Properties and Reactivity of NanoArrayed dsRNA"*) and submitted for review. The research project is relevant to public health because accomplishment

of the aims would enable development of nanoarray-based devices that can detect single-cell levels of dsRNA as a biomarker of viral infection, or the presence of specific cancer cells. The project is relevant to the missions of the NIH and NIBIB because of the stated need for the development and application of new biomedical technologies, including the sensitive, accurate, and robust point-of-care devices for the detection and diagnosis of viral and neoplastic disease. A competitive score for the application was received, but was below the cut-off for funding.

In this project, the spectrofluorimeter is being used to measure the reactivity of RNA molecules towards specific ribonucleases that also are being used to probe dsRNA nanoarray structure-reactivity correlates. The RNA molecules carry specific fluorescent labels that can be used to detect the action of the ribonucleases. The action of the ribonuclease on the dsRNA can cause the appearance of a fluorescent signal, and verify the action of the enzyme. These results are guiding the interpretation of results of the ribonuclease on the dsRNA nanoarray, as probed by atomic force microscopy (AFM). A manuscript has been submitted for review: Redhu, S.K., Castronovo, M., and Nicholson, A. W. (2013) *"Digital imprinting of RNA recognition and processing on a self-assembled, nucleic acid-based matrix.*

b. Instructional Activities involving the Spectrofluorimeter

The spectrofluorimeter has been used in a new Professional Masters of Science in Biotechnology Program of the Department of Biology. The objective of the Biotechnology laboratory course (BIOL 5503) that used the device was to introduce students to laboratory skills with the goal of training them for independent research. Students began by learning basic lab skills and safety protocols, and advanced to an introduction of emergent techniques in ligand binding, molecular biology and invertebrate model systems. The course was taught through team-based learning. The Spectrofluorimeter was used in the study of Protein-ligand binding by fluorescence. This experiment is adapted from the published method of Moller and Denicola. Using the spectrofluorimeter, students were introduced to the concepts of fluorescence emission as a tool for studying ligand binding to protein. Specifically, the binding of the fluorescent probe, 1-Anilino-8-naphthalene sulfonate (ANS) to bovine serum albumin (BSA) resulted in a typical hyperbolic concentration response, which was used to determine binding constants (K_d). The derived Scatchard plot was used to determine the stoichiometry of complex formation. The use of the spectrofluorimeter enabled students to see first-hand the increased specificity and sensitivity of data as compared to previous enzyme-substrate experiments, wherein binding constants were obtained from data generated by a standard UV-Vis spectrophotometer. The direct application for this instrument to pharmacokinetics, drug discovery and lead optimization through the study of absorption, distribution, metabolism, elimination and toxicity (ADMET) of potential drug candidates, using the spectrofluorimeter, has made this an essential tool for the training of students. (Reference: Study of Protein-Ligand Binding by Fluorescence. Matias Moller and Ana Denicola (2002) Biochemistry and Molecular Biology Education vol. 30,No. 5, pp. 309-312.

3. Scanning Electron Microscope

There was a significant delay in the implementation of the scanning electron microscope, due to various problems in manufacture. These have been solved, and now the instrument is being used for several projects, from marine biology, to materials analysis. Most of these projects were well along by the time that the SEM was ready for use, and the images have not yet been incorporated

into the publications. However, sample images are included for each study to illustrate the potential of the instrument.

a. Marine Biology

i. Erik Cordes: Doughty CL*, Quattrini AM, **Cordes EE**. Insights into the population dynamics of the deep-sea coral genus *Paramuricea* in the Gulf of Mexico. In revision for Deep-Sea Res II.

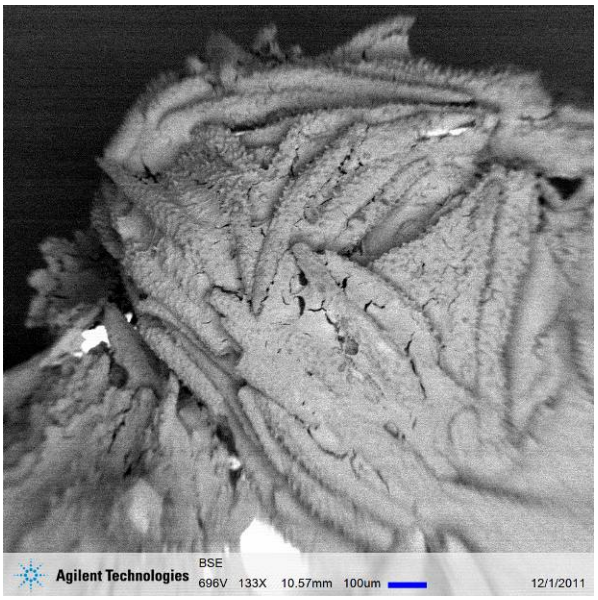
Species in the genus *Paramuricea* are among the most common corals in deep waters (>200 m) of the Gulf of Mexico. *Paramuricea* spp. increase habitat heterogeneity and provide substrate for numerous faunal associates, including ophiuroids that occur on the majority of coral colonies. In light of the Deepwater Horizon oil spill in the Gulf of Mexico, it has become apparent that there is a critical need for data on population dynamics within this genus. To date, at least one species of *Paramuricea* (*P. biscaya*) is known to be negatively impacted by the spill. Using remotely operated vehicles from 2009-2011, we documented the abundances and size frequency distributions of *Paramuricea* across 21 sites at depths of 250-2500 m. Molecular barcoding (*mtCOI+igr+MutS)* was used to delineate species boundaries among the specimens collected. Results suggested that seven haplotypes are present in the Gulf, and appear to be partially segregated by depth [(type H: < 260 m) (type E: 278-445 m) (type A: 443-541 m) (type B1-B3: 800-2600m)]. Abundance of *Paramuricea* spp. determined by ROV surveys was mapped onto high-resolution bathymetric data, which confirmed the corals' preference for topographic highs composed of hard substrata. At any one site, abundances ranged from a single colony (GC246, 894 m) to 523 colonies (AT357, 1050m). Mortality and recruitment rates were estimated from size-frequency data combined with estimated growth rates. Mortality rate decreased with size, from approximately 20% and 70% in new recruits of *Paramuricea* B3 and *P. biscaya* respectively, to less than 10% in colonies over 40 cm in both species. Recruitment rates were estimated from less than one to over 30 individuals per year per site, but patterns in size-frequency histograms suggest that this process is highly variable at the different sites and at different times. These data illustrate that populations of the slow-growing *Paramuricea* species are sparsely distributed and exhibit low recruitment rates, making them highly susceptible to anthropogenic threats.

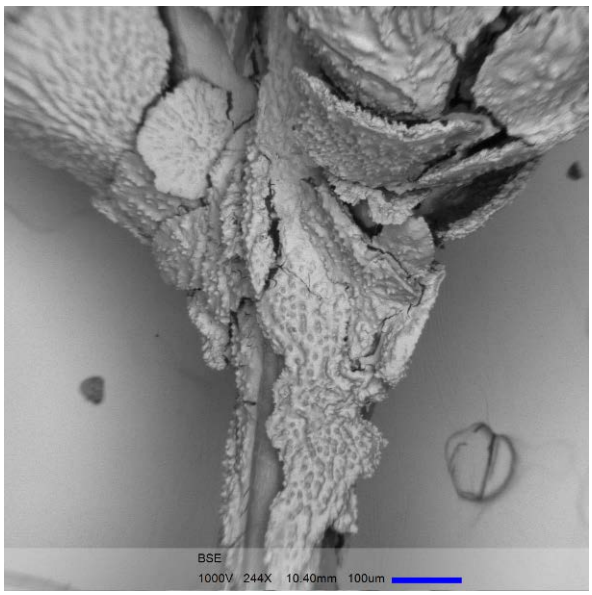
ii. Quattrini AM, Georgian SE, Byrnes L*, Stevens A*, Falco R*, **Cordes EE**. Niche divergence by deep-sea octocorals in the genus *Callogorgia* across the continental slope of the Gulf of Mexico. In revision for Mol Ecol.

Environmental variables that are correlated with depth have been suggested to be among the major forces underlying speciation in the deep sea. This study incorporated phylogenetics and ecological niche models (ENM) to examine whether congeneric species of *Callogorgia* (Octocorallia: Primnoidae) occupy different ecological niches across the continental slope of the Gulf of Mexico (GoM), and whether this niche divergence could be important in the evolution of these closely related species. *Callogorgia americana americana, C. americana delta,* and *C. gracilis* were documented at 13 sites in the GoM (250-1000 m) from specimen collections and extensive video observations. On a first order, these species were separated by depth, with *C. gracilis* occurring at the shallowest sites, *C. a. americana* at mid-depths, and *C. a. delta* at the deepest sites. *Callogorgia a. delta* was associated with areas of increased seep activity whereas *C. gracilis* and *C. a. americana* were associated with narrow, yet warmer, temperature ranges

and did not occur near cold seeps. ENM background and identity tests revealed little to no overlap in ecological niches between species. Temporal calibration of the phylogeny revealed the formation of the Isthmus of Panama was a vicariance event that may explain some of the patterns of speciation within this genus. These results elucidate the potential mechanisms for speciation in the deep sea, emphasizing both bathymetric speciation and vicariance events in the evolution of a genus across multiple regions.

These two papers will include scanning electron micrographs illustrating the organisms, similar to these low magnification images with extraordinary detail of the plates that make up the surfaces of these corals.



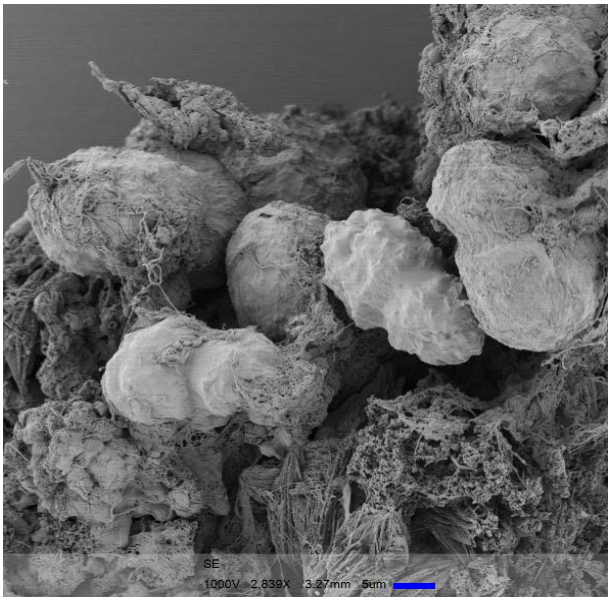


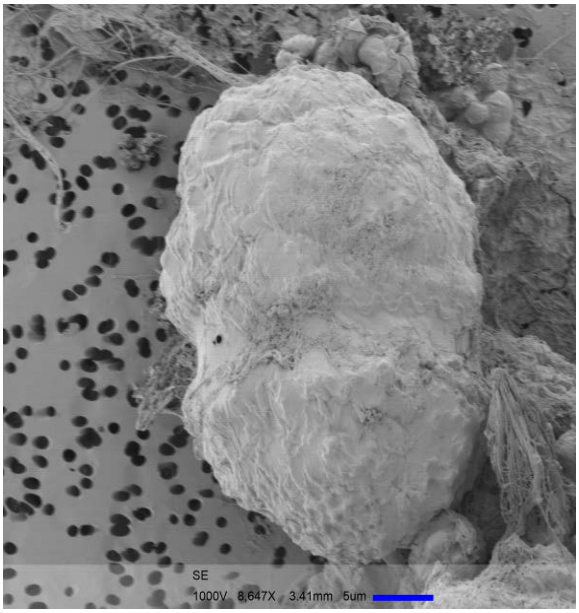
iii. Sellers, C.G., Gast, R.J. and Sanders, R.W. in preparation. Selective feeding and foreign plastid retention in an Antarctic dinoflagellate.

A dinoflagellate strain (RSD) isolated from the Ross Sea, Antarctica was previously shown to feed on and sequester plastids from the haptophyte alga *Phaeocystis antarctica.* We followed populations of RSD in long-term co-culture with *P. antarctica* in separate batch culture experiments with either an Antarctic haptophyte *Chrysochromulina* sp., or with the cryptomonad *Geminigera cryophila* (the plastid source for an Antarctic *Mesodinium rubrum*). Over a period of 12 days, RSD selectively fed on *P. antarctica*, substantially reducing its abundance, but no ingestion of *Chrysochromulina* sp. or *G. cryophila* by RSD was detected. Additionally, a population of RSD cells isolated from *P. antarctica* using flow cytometry grew slowly during nine months with a marked reduction in mean numbers of chloroplasts / cell and an increased frequency of cells lacking chloroplasts. Together, these results suggest a feeding specificity and a longevity of the association that are consistent with the proposal that RSD represents a model for an early stage of plastid evolution.

iv. McKie-Krisberg, Z.M. and Sanders, R.W. in preparation. Mixotrophy in the Arctic picoeukaryotic alga *Micromonas*.

Mixotrophic phytoflagellates have been identified as major phagotrophs with significant impact on bacterial and small algal communities in numerous freshwater and marine environments. Though there are very few published studies of mixotrophs in polar oceans, data suggest that they may be common and sometimes dominant bacterivores in those systems. A recent study suggested that photosynthetic picoeukaryotes are important bacterivores in temperate oceanic systems, and we have identified this group as dominant grazers of bacteria in the Arctic Ocean. The prasinophyte *Micromonas* sp., perhaps the most common photosynthetic picoeukaryote in Arctic waters and one present throughout the year, was tentatively identified as the most abundant of these picoeukaryotic mixotrophs in an autumn cruise to the Beaufort Sea. Picoeukaryotic mixotrophs were estimated to be responsible for removing up to 25% of the bacterial standing stock per day. We have confirmed uptake of bacteria-sized particles in laboratory cultures of *Micromonas* sp. isolated from the Arctic Ocean (Baffin Bay) at rates comparable to those we determined in situ in the Beaufort Sea.



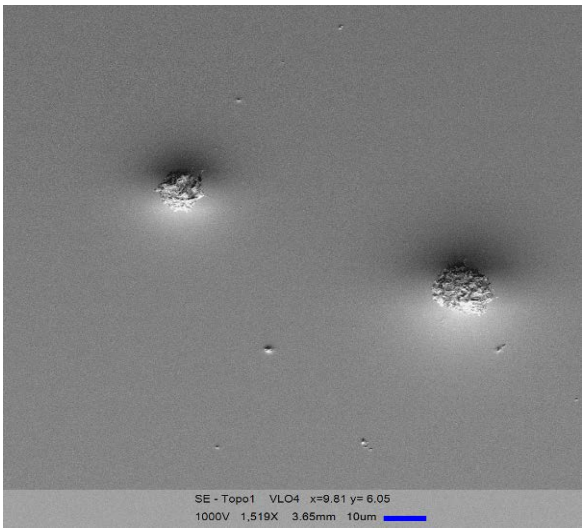


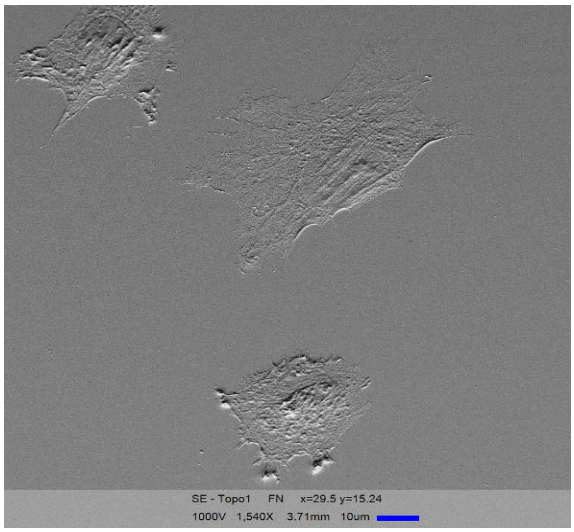
b.Cell and Developmental Biology

i. Piotr Jakubowski a, Juan J. Calvete b, Johannes A. Eble c, Philip Lazarovici d, Cezary Marcinkiewicz, Toxicology and Applied Pharmacology 269 (2013) 34–42

Identification of inhibitors of α2β1 integrin, members of C-lectin type proteins, in Echis sochureki venom Snake venom antagonists of α2β1 integrin have been identified as members of a C-lectin type family of proteins (CLP). In the present study, we characterized three new CLPs isolated from Echis sochureki venom, which interact with this integrin. These proteins were purified using a combination of gel filtration, ion exchange chromatography and reverse phase HPLC. Sochicetin-A and sochicetin-B potently inhibited

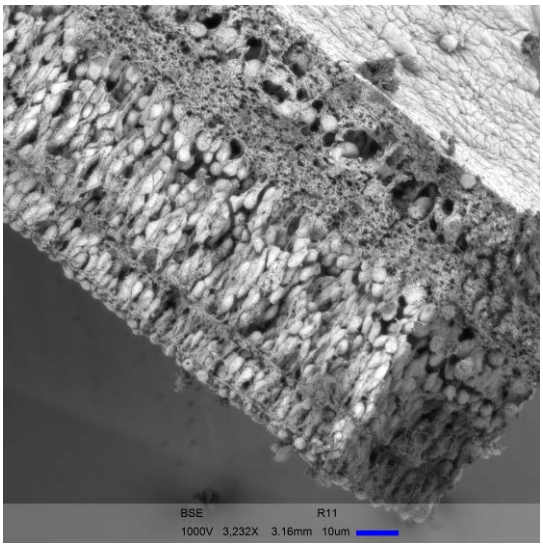
adhesion of cells expressing α2β1 integrin and binding of isolated α2β1 ectodomain to collagen I, as well as bound to recombinant GST-α2A domain in ELISA, whereas activity of sochicetin-C in these assays was approximately two orders of magnitude lower. Structurally, sochicetin-B and sochicetin-C are typical heterodimeric αβ CLPs, whereas sochicetin-A exhibits a trimer of its subunits (αβ)3 in the quaternary structure. Immobilized sochicetins supported adhesion of glioma cell lines, LN18 and LBC3, whereas in a soluble form they partially inhibited adhesion of these cells to collagen I. Glioma cells spread very poorly on sochicetin-A, showing no cytoskeleton rearrangement typical for adhesion to collagen I or fibronectin. Adhesion on CLP does not involve focal adhesion elements, such as vinculin. Sochicetin-A also inhibited collagen-induced platelet aggregation, similar to other CLPs' action on the blood coagulation system.

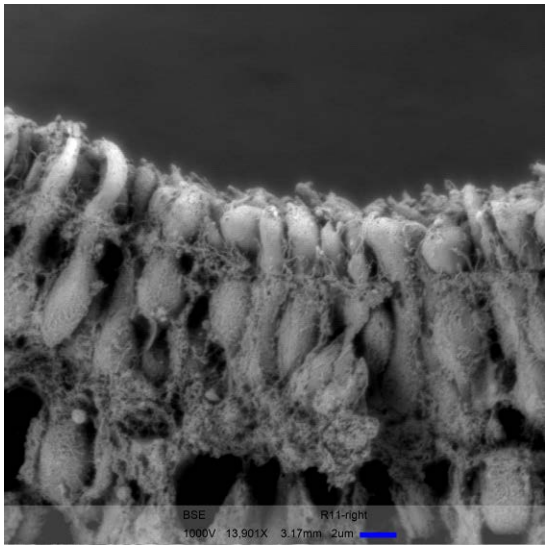




A comparison of cell growth on two different media

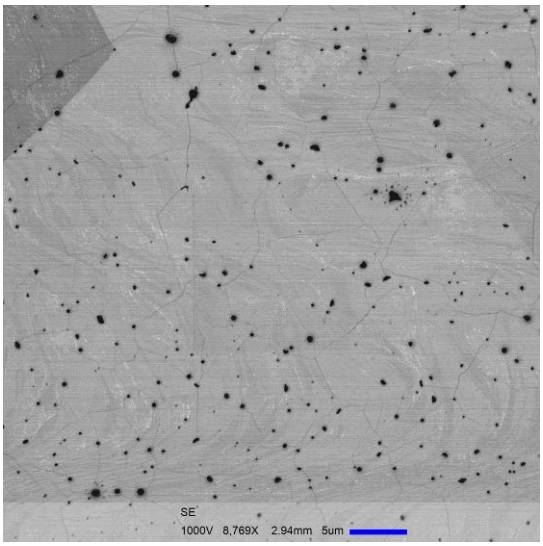
ii. Sheffield. Studies of the embryonic chick retina and development of the vasculature. (in development). Bird retinas are unique in that they have no internal vasculature, and derive their nutrient supply primarily from anaerobic processes. However, they do contain a vascular structure, the *pectin oculi*, which extends from the entry of the optic nerve into the vitreous space. Thus, nascent angiogenic cells migrate into the eye and enter the primordium of the pectin, without entering the neural portion of the retina. This provides an opportunity to analyze the factors that lead to a choice of one angiogenic pathway over another. A portion of this study is to acquire scanning electron microscope images of the relevant portions of the tissue for comparative analysis. These images represent different views of the neural retina at the time that the *pectin oculi* is developing.

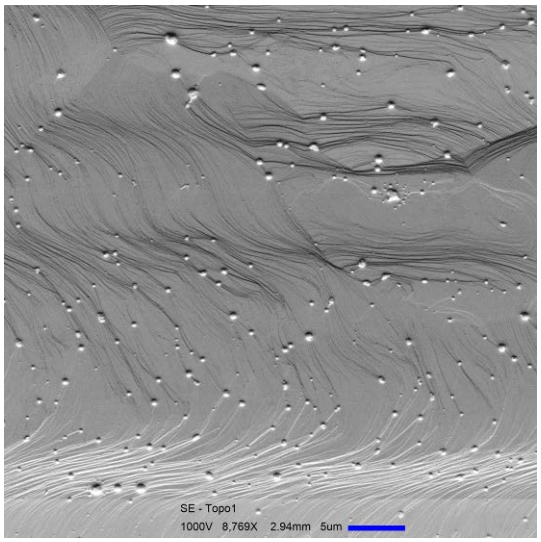




c.Materials Science

i. Ying Feng, Ke Chen, Dry Transfer of CVD-grown Graphene to Fabricate Mg/MgO/Graphene Tunnel Junctions (Thesis project) No abstract available yet.





Two images of the same graphene structure, visualized with different SEM techniques

ii.Blends of Methyl Cellulose and an Amorphous Dendritic Polyethylene Glycol, POSS-PEG8
Chinnam, Parameswara; Jimenez, Jayvic; Zhang, Hanjun; Kulkarni, Sunil; Sheffield, Joel; Wunder, Stephanie (submitted, in revision)

18. Extent of Clinical Activities Initiated and Completed. Items 18(A) and 18(B) should be completed for all research projects. If the project was restricted to secondary analysis of clinical data or data analysis of clinical research, then responses to 18(A) and 18(B) should be "No."

18(A) Did you initiate a study that involved the testing of treatment, prevention or diagnostic procedures on human subjects?

_____Yes
__x___No

18(B) Did you complete a study that involved the testing of treatment, prevention or diagnostic procedures on human subjects?

_____Yes
__x___No

If "Yes" to either 18(A) or 18(B), items 18(C) – (F) must also be completed. (Do NOT complete 18(C-F) if 18(A) and 18(B) are both "No.")

18(C) How many hospital and health care professionals were involved in the research project?

_____Number of hospital and health care professionals involved in the research project

18(D) How many subjects were included in the study compared to targeted goals?

_____Number of subjects originally targeted to be included in the study
_____Number of subjects enrolled in the study

Note: Studies that fall dramatically short on recruitment are encouraged to provide the details of their recruitment efforts in Item 17, Progress in Achieving Research Goals, Objectives and Aims. For example, the number of eligible subjects approached, the number that refused to participate and the reasons for refusal. Without this information it is difficult to discern whether eligibility criteria were too restrictive or the study simply did not appeal to subjects.

18(E) How many subjects were enrolled in the study by gender, ethnicity and race?

Gender:
_____Males
_____Females
_____Unknown

Ethnicity:
_____Latinos or Hispanics
_____Not Latinos or Hispanics
_____Unknown

Race:
_____American Indian or Alaska Native
_____Asian
_____Blacks or African American
_____Native Hawaiian or Other Pacific Islander
_____White
_____Other, specify:_____
_____Unknown

18(F) Where was the research study conducted? (List the county where the research study was conducted. If the treatment, prevention and diagnostic tests were offered in more than one county, list all of the counties where the research study was conducted.)

19. Human Embryonic Stem Cell Research. Item 19(A) should be completed for all research projects. If the research project involved human embryonic stem cells, items 19(B) and 19(C) must also be completed.

19(A) Did this project involve, in any capacity, human embryonic stem cells?
_____Yes
___x__ No

19(B) Were these stem cell lines NIH-approved lines that were derived outside of Pennsylvania?
_____Yes
_____ No

19(C) Please describe how this project involved human embryonic stem cells:

20. Articles Submitted to Peer-Reviewed Publications.

20(A) Identify all publications that resulted from the research performed during the funding period and that have been submitted to peer-reviewed publications. Do not list journal abstracts or presentations at professional meetings; abstract and meeting presentations should be listed at the end of item 17. **Include only those publications that acknowledge the Pennsylvania Department of Health as a funding source** (as required in the grant agreement). List the title of the journal article, the authors, the name of the peer-reviewed publication, the month and year when it was submitted, and the status of publication (submitted for publication, accepted for publication or published.). Submit an electronic copy of each publication or paper submitted for publication, listed in the table, in a PDF version 5.0.5 (or greater) format, 1,200 dpi. Filenames for each publication should include the number of the research project, the last name of the PI, the number of the publication and an abbreviated research project title. For example, if you submit two publications for PI Smith for the "Cognition and MRI in Older Adults" research project (Project 1), and two

publications for PI Zhang for the "Lung Cancer" research project (Project 3), the filenames should be:

Project 1 – Smith – Publication 1 – Cognition and MRI
Project 1 – Smith – Publication 2 – Cognition and MRI
Project 3 – Zhang – Publication 1 – Lung Cancer
Project 3 – Zhang – Publication 2 – Lung Cancer

If the publication is not available electronically, provide 5 paper copies of the publication.

Note: The grant agreement requires that recipients acknowledge the Pennsylvania Department of Health funding in all publications. Please ensure that all publications listed acknowledge the Department of Health funding. If a publication does not acknowledge the funding from the Commonwealth, do not list the publication.

Title of Journal Article:	Authors:	Name of Peer-reviewed Publication:	Month and Year Submitted:	Publication Status (check appropriate box below):
1. None				☐Submitted ☐Accepted ☐Published

20(B) Based on this project, are you planning to submit articles to peer-reviewed publications in the future?

Yes_____ No_____

If yes, please describe your plans:

21. Changes in Outcome, Impact and Effectiveness Attributable to the Research Project. Describe the outcome, impact, and effectiveness of the research project by summarizing its impact on the incidence of disease, death from disease, stage of disease at time of diagnosis, or other relevant measures of outcome, impact or effectiveness of the research project. If there were no changes, insert "None"; do not use "Not applicable." Responses must be single-spaced below, and no smaller than 12-point type. DO NOT DELETE THESE INSTRUCTIONS. There is no limit to the length of your response.

None

22. Major Discoveries, New Drugs, and New Approaches for Prevention Diagnosis and Treatment. Describe major discoveries, new drugs, and new approaches for prevention, diagnosis and treatment that are attributable to the completed research project. If there were no major discoveries, drugs or approaches, insert "None"; do not use "Not applicable." Responses must be single-spaced below, and no smaller than 12-point type. DO NOT DELETE THESE INSTRUCTIONS. There is no limit to the length of your response.

None

23. Inventions, Patents and Commercial Development Opportunities.

23(A) Were any inventions, which may be patentable or otherwise protectable under Title 35 of the United States Code, conceived or first actually reduced to practice in the performance of work under this health research grant? Yes_____ No___x_____

If "Yes" to 23(A), complete items a – g below for each invention. (Do NOT complete items a - g if 23(A) is "No.")

a. Title of Invention:

b. Name of Inventor(s):

c. Technical Description of Invention (describe nature, purpose, operation and physical, chemical, biological or electrical characteristics of the invention):

d. Was a patent filed for the invention conceived or first actually reduced to practice in the performance of work under this health research grant?
Yes_____ No____

If yes, indicate date patent was filed:

e. Was a patent issued for the invention conceived or first actually reduced to practice in the performance of work under this health research grant?
Yes_____ No____
If yes, indicate number of patent, title and date issued:
Patent number:
Title of patent:
Date issued:

f. Were any licenses granted for the patent obtained as a result of work performed under this health research grant? Yes_____ No____

If yes, how many licenses were granted?_____

g. Were any commercial development activities taken to develop the invention into a commercial product or service for manufacture or sale? Yes___ No____

If yes, describe the commercial development activities:

23(B) Based on the results of this project, are you planning to file for any licenses or patents, or undertake any commercial development opportunities in the future?

Yes_____ No____x_____

If yes, please describe your plans:

24. Key Investigator Qualifications. Briefly describe the education, research interests and experience and professional commitments of the Principal Investigator and all other key investigators. In place of narrative you may insert the NIH biosketch form here; however, please limit each biosketch to 1-2 pages. *For Nonformula grants only – include information for only those key investigators whose biosketches were not included in the original grant application.*

Joel B. Sheffield

UNIVERSITY ADDRESS: Department of Biology (215) 204-8839
College of Science and Technology jbs@temple.edu

Professional Preparation

Univ. of Chicago, Chicago, Ill.	Develop. Biology	1969	Ph.D.
Brandeis Univ., Waltham, Mass.	Biology	1963	A.B.

Thesis: An electron microscope study of the development of the chick neural retina and aggregates of its cells. 1969: A. A. Moscona

Postdoctoral Trainee Netherlands Cancer Inst. 1970-71

Appointments

Department Chairman Temple University	1990-02	
Professor	Temple University	1989-pr.
Associate Professor	Temple University	1982-89
Permanent Consultant-Membranes	CIDAC	1977-87
Assistant Professor	Temple University	1977-82
Associate Member of Faculty	Rutgers University	1974-77
Assistant Member	Inst. for Med. Res (Coriell Inst.).	1971-77
Guest Investigator and Fellow	Rockefeller Institute	1965
Guest Investigator and Fellow	Rockefeller Institute	1963-64

Publications

Selected Publications

Sheffield, J.B. Sorting behavior among cells from the 14 day embryonic chick neural retina. Devel. Biol. 89:41-47, 1981.

Sheffield, J.B. and H.-P. Li. Interactions among cells of the developing neural retina in vitro. Amer. Zool. 27:145-159, 1987.

Hilfer, S.R., Marrero, L., and Sheffield, J.B. Patterns of Cell Movement in Early Organ Primordia of the Chick Embryo. Anat. Rec. 227:508-517, 1990.

Buono, R.J. and J.B. Sheffield. Changes in expression and distribution of lactate dehydrogenase isoenzymes in the developing chick retina. Exp. Eye Res. 53:199-204, 1991.

Ruggiero, F. and J.B. Sheffield. The Distribution of Mitochondrial Carboxylases in the Developing Chick Neural Retina: Avidin as a Probe. J. Histochem. Cytochem, 1998, 462:177-18

Zhou, J, V. Rothman, I Sargiannidou, S. Dimitrov, C. Qiu, E. Smith, J. Sheffield, M. Sharma and G.P. Tuszynski. Cloning and Characterization of Angiocidin, a Tumor Cell Binding Protein for Thrombospondin-1. J. Cell. Biochem. 92:125-46. 2004

Claudio PP, Cui J, Ghafouri M, Mariano C, White MK, Safak M, Sheffield JB, Giordano A, Khalili K, Amini S, Sawaya BE Cdk9 phosphorylates p53 on serine 392 independently of CKII. . J Cell. Phys. 2006.

*Selected Publications with Undergraduate Authors. (Authors are indicated with a *)*

Sheffield, J.B., D. Pressman* and M. Lynch. Cell isolated from the embryonic neural retinal differ in behavior in vitro and membrane structure. Science. 209:1043-1045, 1980.

Sheffield, J.B., Z. Stauber*, B.J. Hayden and G.J. Chader. Morphological description of Y79 Retinoblastoma cells by a simple image analysis system. In vitro 26:325-329, 1990.

Woodruff-Pak, D.S., J.F. Cronholm* and J.B. Sheffield. Purkinje cell number related to rate of classical conditioning. Neuro Report 1:165-168, 1990.

Sheffield, J.B. and M. Moyer*. The emergence of flat cells in stationary cultures of embryonic neural retinal cells. In Vitro 26:1073-1078, 1990.

Sheffield, J.B., V. Krasnopolsky* and E. Dehlinger*. Inhibition of Retinal Growth Cone Activity by Specific Metalloproteinase Inhibitors *in vitro*. Development Dynamics 200:79-88, 1994.

Synergistic Activities.

1. Awards: Lindback Award for teaching, Temple University, 1990; Honorary Member of Golden Key International Student Honor Society, 2001; Temple University Great Teacher Award, 2003.
2. Co-Chair of the "Smart Classrooms" committee, responsible for developing designs for effective classroom teaching.
3. Introduced the use of mobile wireless laptop laboratories into classrooms for computer access.

Collaborators and Other Affiliations.

a. Collaborators and Co Editors

Michael Davidson, National High Energy Magnet Laboratory, Tallahassee, FL
George Tuszynski, Department of Biology, Temple University
Gregory Smutzer, University of Pennsylvania

b. Graduate Advisor

Doctoral Advisor: Aron Moscona, University of Chicago, (retired)
Postdoctoral Advisor: Pieter Emmelot, Netherlands Cancer Institute (retired)

c. Ph.D advisees (out of 7. 5 additional M.A, advisees)

Hongzhuang Peng , 1/97 (Current Address: Wistar Institute)
"A Study of the Expression of Proteases in Embryonic Chick Retinal Cells"
Jennee Loi, 6/99 (Current Address: Avitar Technologies)
"A Study of Programmed Cell Death (Apoptosis) in the Developing Chick Neural Retina and the Effects of Dexamethasone on its Expression"
Bashar Hanna, 12/99 (Current Address: Dean for Academic Affairs, DeVry University)
"Characterization of Chicken Metallo-proteinase Inhibitor-3 (ChiMP3) During Development of the Chicken Neural Retina"

Former Undergrad Honors Students (Partial List)

Adedunle Akera-Alle, Elizabeth Pia Beasley, Lev Belder, David Brown, Florencia Bullrich, Russell Buono*, Jignesh Dholoria, Maria DiCarlantonio, Dalfoni Frazer, Sonela Hajno, Bashar Hanna* , Ed Hart, Michael He, Migdalia Hernandez, Cynthia Jurkovich, Vladimir Krasnopolsky, David Lerner, Asma Mahedavi, Kimberly Mattson, Delana Miles, Elana Miles, Jawaad Mohiuddin, MaryLou Moyer, Ryan O'Donnell, Nupa Patel, David Rutter, Sajaad Sabir, Inna Shukher, Rewandee Soontharothai, Brian Wong

*Bashar Hanna and Russell Buono were undergraduate students before they joined the graduate program at Temple.