School of Science and Mathematics

Student Research Poster Display

Caputo Hall Lobby

October 19—26, 2010

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Note: The names of Millersville University faculty advisors are designated by an asterisk (*) in the abstracts.

BIOLOGY

1. Chemical Analysis of Anti-herbivory Constituents Found in *Ailanthus altissima.*

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The anti-herbivory and insecticidal properties of *Ailanthus altissima* (Tree of Heaven) leaf tissue were investigated using bioassay-directed fractionation and correlated to known allelopathic activity. When applied topically, crude aqueous extracts of *A. altissima* were confirmed to be significantly toxic to several species of *Lepidoptera* larvae (*Spodotpera frugiperda, Trichoplusia ni, Malacosoma Americanum, Heliocoverpa zea, Heliothis verescens*), showing mortality rates as high as 84%. Crude extracts also demonstrated a clear reduction in leaf consumption in a variety of larval feeding experiments that were conducted. This crude extract was concentrated and size separated into 10 fractions via a Sephadex column, which were then bioassayed for both insecticidal and allelopathic activity. Only two fractions, which had retention times of 0-260 seconds and 261-520 seconds respectively, had activity. Fraction #2 provided 100% inhibition of *Raphanus sativus* (radish) seed germination as well as a 100% mortality rate of *Trichoplusa ni* (Cabbage looper), indicating a strong correlation between allelopathy and insectidal/anti-herbivory activity. Allelopathy in A. *altissima* has been attributed to a group of compounds known as quassinoids, and more specifically to a compound known as ailanthone. This group of compounds may also be responsible for the anti-herbivory and insecticidal activity found in *A. altissima*. Chemical analysis of this active fraction by additional refined fractionation such as TLC, GC-MS, and HPLC is in progress, and will result in the isolation of the active compound(s).

2. A Biosensor for the Detection of Ovarian Cancer

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According to the National Cancer Institute, ovarian cancer ranks fourth in cancer deaths among women and is the deadliest gynecological cancer. Most patients with ovarian cancer present in late clinical stages, where survival rate over five years is only 35%. However, patients presenting cases in stage-one ovarian cancer have a five year survival rate of 95%. Consequently, the development of early detection techniques is imperative to decrease the mortality rate of this disease. CA-125 is an ovarian cancer marker that is secreted by ovarian tumors and is an important clinical indicator that the cancer is present.

Aptamers, are single stranded RNA or DNA molecules that have the ability to bind biomedically important molecules with high specificity. We are attempting to use them in the development a biosensor probe to detect an ovarian cancer marker (CA-125). Toward this end we need to covalently link aptamers to a gold substrate. Here we describe the attachment of the 5' end of fluorescently labeled aptamers to a gold/ titanium substrate. To achieve this,

we explored various substrate preparation techniques and incubated gold/ titanium plated slides with chemically modified aptamers at various concentrations and time intervals. Degree of attachment was measured using quantitative fluorescence microscopy and by generating a standard curve (brightness units vs. aptamers concentration). Beyond this we plan to assess the functionality of attached aptamers by exposing the sensor to fluorescently labeled CA-125 and quantifying binding to the aptamers. The device that we are developing has the potential to replace current assays of most biomedically important molecules used clinical diagnosis with a single instrument that provides very rapid readouts and early diagnostic capacity.

3. A PCR Approach to Investigating Bone Formation in the Turtle Shell

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The turtle shell is an evolutionary novelty, and understanding how it is formed will help to explain a major puzzle in macroevolution. The goal of this research was to examine the cells that produce the plastron, or ventral shell, of the turtle, and to study their developmental origin. Previous studies have shown that the bones of the plastron form in a similar manner to facial and skull bones. The flat bones of the skull form through a unique process, in which loosely packed cells convert directly into osteoblasts (bone-forming cells), without first producing cartilage. In addition, the skull bones are produced by a unique population of migrating cells known as cranial neural crest cells. Our hypothesis is that the early plastron is constructed from neural crest cells, similar to those that produce the bones of the face and skull. To investigate this hypothesis, we are examining the pattern of gene expression in the plastron just as the bones are beginning to form by isolating RNA and performing PCR assays. We predict that the plastron cells will express genes involved in bone, but not cartilage, differentiation. Finally, we are also examining the expression of two genes expressed in cranial neural crest cells. If these two genes are expressed in the plastron, it would provide further support for the neural crest origin of the cells that form the bones of the plastron.

4. Spatial and Temporal Distribution of *Hydra (Chlorohydra) viridissima*

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Freshwater hydra are cosmopolitan in their distribution with multiple species found in Pennsylvania. Hydra have no medusa or free swimming larval form that might enhance their distribution, yet it seems that they are able to distribute and colonize their habitats with ease. This study focuses on the spatial and temporal distribution of *Hydra* (*Chlorohydra) viridissima* in a small pond in Lancaster County, PA. Although known methods of dispersal for green hydra include gas bubble flotation and the somersault method, it is unknown how far these organisms can travel and what environmental factors influence colonization. Monitoring stations containing vertically tiered settling plates (3 x 85 mm diameter Petri plates per depth) were placed at three locations in the Roddy research pond, a small

freshwater pond (0.2 ha) located at Millersville University. This research pond is teardrop shaped and is bordered by *Phragmites* and cattail vegetation. The narrow end of the pond, known as the apex, is shallow (~ 1 m) and contains extensive infralittoral vegetation made up of *Nitella* and *Phragmites* rhizomes. The mid-pond region is a deeper pelagic area (~ 4.5 m depth) with very little vegetation. Finally, the base of the teardrop has a moderate depth (~ 2 m) and lacks benthic vegetation. Sampling was conducted during two separate time periods (August - October 2009) and November 2009 - March 2010). Hydra colonized the shallow end of the pond in preference for other regions regardless of season (**apex:mid-pond:base**, winter 13:0:1, summer 8:0:1). We also found similar changes in population density during both sampling periods (~ 60 hydra/week). In the apex samples, hydra were present on the benthic plates in preference to those at surface (Wilcoxon's ranked sign test $p<0.02$). Colonization of hydra on the upper side as compared to underside of plates was the same in the winter, however there was a preference for the underside in the summer. This seems to show that green hydra will colonize in areas that provide optimal forging as opposed to optimal photosynthesis.

5. Examination of Receptor Cells on the Olfactory Lamellae of the Smooth Dogfish Shark, *Mustelus canis*

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This study utilized scanning electron microscopy to image the microvillous receptor cells found on the epithelium of olfactory lamellae in the smooth dogfish shark, *Mustelus canis*. Two specimens were collected from the wild, one male (63.5 cm TL) and one female (54 cm TL). Their olfactory lamellae were dissected and fixed in a 4% glutaraldehyde solution and then post-fixed in a 1% OsO_4 solution. Following dehydration via a graded series of acetone baths, the individual olfactory lamellae were critical-point dried and gold sputter coated. Micrographs were taken using an Amray 1820 scanning electron microscope and PC-based Orion imaging software. Previous studies indicate that the microvillous receptor cells found on the olfactory lamellae of various elasmobranch species represent a unique receptor cell type. Studies also indicate that this cell type differs in morphology and density across the olfactory epithelium. The current study confirms these findings. Microvillous cells were present on the epithelia of both specimens and were most dense on the areas of the lamellae not associated with traditional olfaction of odor molecules. The microvillous cells bear a strong morphological resemblance to previously published micrographs and descriptions of microvillous cells response for pheromone reception in other vertebrates.

6. Learning and Memory Retention in *Paramecium caudatum*

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Invertebrate model systems have yielded remarkable insight into the cellular mechanisms of learning and memory. The present study not only demonstrates a learned response in groups of singled celled organism, but also demonstrates retention of the learned response.

Paramecium caudatum is a cigar shaped, unicellular organism measuring about 250 μm in length that utilizes cilia for locomotion. Previous experimentation has shown that *P. caudatum* is attracted to a mild cathodal stimulus. This study couples the phenomenon of cathodal attraction with differential lighting to demonstrate learning in small groups of paramecia. Additionally, extinction trials were run to determine the retention time of the learned brightness discrimination.

A microscope concavity slide was fitted with stainless steel electrodes at opposite ends of the well. Approximately 20 paramecia were placed in the well in ~100 μl of Sonnenborn's culture medium. During a three minute training session one half of the well was illuminated (3,150 lux) and the other was shaded with black felt and an electrical stimulus was applied (6.5 V at 2 Hz). Next, the paramecia were exposed to a three minute resting session with uniform, reduced illumination (800 lux) and no electrical current. Finally, the paramecia were exposed to a three minute response session with the original differential illumination but without electrical current.

Chi Square analysis (df=1) showed that during the training session paramecia were attracted to the cathode without regard for the type of illumination ($p<0.05$). During the resting session, no significant difference in distribution was observed ($p>0.10$). In the response period, paramecia moved to the light or dark area that was originally associated with the cathode during training ($p<0.05$). A reversal experiment showed that paramecia responded to the lighting regime rather than the physical positioning of the overhead light ($p<0.05$). Thus, these groups of paramecia not only demonstrated a learned response after a single training trial, they demonstrated the ability to retain the learned response.

To determine memory extinction, additional resting and response periods were run following the initial three minute response session. Chi square analysis showed that the learned response lasted twelve minutes.

7. Do Cuttlefish Sleep? A Behavioral Investigation

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Typical humans spend up to one-third of their lifetimes sleeping. Why? What is sleep, who does it, and what is its function? Sleep is a physical and mental resting state in which an organism becomes relatively inactive and has a higher threshold for responding to most external stimuli. There are many hypotheses for why humans sleep, but one important hypothesis is that sleep is essential to memory consolidation. To better understand the biological significance of sleep, a comparative approach has been taken; sleep has been documented in reptiles, fish, birds, and mammals. A sleep-like state has also been documented in the nematode *Caenorhabditis elegans* and the fruit fly, *Drosophila melanogaster*. The common cuttlefish, *Sepia officinalis*, could provide an ideal new species for comparative studies: it has a large, complex brain and sophisticated learning capabilities, its neurobiology and behavior have been well studied, and it are easy to maintain in the laboratory setting. A set of three experiments were performed to determine if the common cuttlefish sleeps. Four questions were addressed: (1) do cuttlefish rest? (2) do cuttlefish have a distinct activity cycle in the laboratory? (3) do cuttlefish show an expanded period of

rest (rebound) after they are deprived from resting? and, (4) does stage in the cuttlefish life cycle influence resting habits and duration? In Experiment 1, no distinct activity cycles or evidence for rebound was found in senescent adults; however, a quiescent state and a REM-like sleep state were observed. In Experiment 2, nocturnal activity cycles were documented in juveniles. In Experiment 3, a distinct nocturnal activity cycle and a significant rebound period were found in sub-adults. These results indicate that cuttlefish probably do sleep. Future investigations addressing whether resting cuttlefish show a higher threshold for responding to external stimuli are needed to confirm these promising findings.

8. Diaziridines: Progress Towards the Synthesis of Some 2-Sulfobenzoyl-, and 1,2-Dibenzenesulfonyldiaziridines

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Diaziridines are a class of three-membered ring heterocycles that contain one carbon and two nitrogen atoms. They are useful intermediates in the synthesis of more complex heterocyclic compounds, some of which have found applications in the pharmaceutical industry. Although several studies on the synthesis and chemistry of 1-aroyl-, and 1,2-diaroyldiaziridines have appeared in the chemical literature, there are no reports on the synthesis and chemistry of their ***dibenzenesulfonyl***, or their ***2-sulfobenzoyl*** analogues. Thus, we are attempting to synthesize a variety of these diaziridine derivatives in order to study their chemical reactivity. It is anticipated that this project will shed some light on how different electron withdrawing groups influence the reactivity of the three-membered diaziridine ring. The results of the progress of this investigation are the subject of this poster.

9. Coupling Receptor Targeting Ligands on Biodegradable Polymer Micro- and Nanoparticles

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Biodegradable polymer micro- and nanoparticles have found application as carriers of drugs for controlled release. If suitably functionalized, the micro- and nanoparticles may also be used to deliver encapsulated drugs to specific tissue in order to minimize toxicity and improve efficacy. However, commonly used polymers such as polylactic acid (PLA) and polyglycolic acid (PLGA) lack the high density reactive surface groups necessary for ligand conjugation. Layer by layer self assembly was used to introduce $-NH_2$ groups on the surface of PLA micro- and nanoparticles. Folic acid, a small ligand molecule that has a high binding affinity for the folate receptor was then covalently attached on the spherical particles. The ligand functionalized micro- and nano particles may be useful in delivery of drugs to specific tissue based on folate-folate receptor binding.

10. Geopod: A Virtual Platform for Geoscience Data Visualization

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Geopod is an intuitive, interactive Java module that allows users to navigate and probe an immersive 3-D world. Geopod features authentic geophysical data which is based on actual physics, thus exhibiting technical accuracy and scientific soundness. Geopod leverages the Unidata Program Center's open source Java-based visualization software, the Integrated Data Viewer (IDV), to import and render meteorological data.
Geopod provides the user with the unique experience of flying through the data volume, as opposed to the third-person perspective that IDV offers. This intuitive, engaging interaction style is designed to appeal to students and motivate them to explore the data volume and gain a comprehensive understanding of meteorological concepts. Geopod provides a number of useful devices and features to facilitate and enhance this exploration process, while maintaining a clean, easy-to-use interface which is accessible to novices and experts alike. Navigation features include keyboard and mouse controls, a compass, an autopilot system capable of traversing isosurfaces, and forward and reverse geocoding. User-selected meteorological parameters are continuously updated on customizable display panels, with a particle imaging device for viewing ice crystal formations, a dropsonde device for vertical profiling, and a grid point displayer for viewing the underlying dataset. A noted-locations system allows users to annotate (and later view, edit, or save) parameter values at points of interest.
In addition, a mission subsystem provides instructors with a powerful yet flexible teaching tool. This feature allows educators to create and distribute missions which can include background material and any number of specific objectives, each of which has an assessment or 'quiz' feature which becomes available to students after they have completed a specific set of requirements.

11. Hardware Accelerated Ray Tracing with OpenCL

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Interactive computer graphics has traditionally been rendered using rasterization algorithms which transform geometric primitives from 3D space to screen space using matrix multiplication. Other techniques such as ray tracing are capable of producing more realistic images but at a significantly higher computational cost. In this project, we are exploring how the massively parallel nature of current graphics cards can be used to accelerate ray tracing algorithms, with a goal of achieving real-time animation. To achieve this, we are using a framework called OpenCL that allows code written in a C-like language to be run on highly parallel devices such as graphics cards. To date, we have implemented several limited ray tracing algorithms that are capable of operating at smooth frame rates for simple scenes.

Implementing the complex data structures required to render more complex scenes, however, has been more problematic due to OpenCLs restrictive programming model and is the current focus of this research.

12. A Comparison of Different Green Roof Constructions on Runoff Nitrate Fluxes

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The Green Roof Project is of environmental significance because it helps to improve the overall quality of our surface water. Green roofs reduce nitrate (NO_3^-) concentrations in waters, thereby counteracting eutrophication. In addition, by using green roof media on rooftops across the Susquehanna Valley, we can neutralize the pH and reduce the volume of rainwater runoff to streams. To measure the benefits of green roof media, we established three plots; the first reflecting standard roof construction, the second containing only green roof media, and the third containing the green roof media as well as a lining of recycled fabrics and a drainage conduit. Using a standard EPA method for measuring nitrates in runoff from each of the three plots for a 308 day period from August 2009 to June 2010, we find that nitrate levels were reduced from 423 µg/day in plot 1 to 24 µg/day in plot 2 and 29 µg/day in plot 3. Other findings include a more neutral pH of 6.79 in plot 2 and a pH of 6.78 in plot 3 as opposed to the average pH of precipitation in plot 1 of 5.43. The green roof media in plots 2 and 3 also reduced the total rainwater runoff from 0.99 L/day to 0.69 L/day in plot 2 and 0.70 L/day in plot 3. These results verify the many benefits of having green roofs. The nitrate flux decline shows that eutrophication in surface waters may be reduced by the use of green roofs. The findings of a precipitation pH of 5.43 in the Susquehanna Valley may be harmful to the region's wildlife and waterways if not neutralized by an effort such as green roofs. The reduction in runoff volume also leads us to believe that flooding in urban areas can be minimized by the use of green roofs.

13. Stream Carbon Dioxide Degassing in Watershed 2 at Coweeta Hydrologic Laboratory, Western North Carolina

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We studied the degassing of carbon dioxide (CO_2) at Coweeta Hydrologic Laboratory located in Otto, North Carolina. Chemical weathering of silicate bedrock removes CO_2 from the atmosphere and requisitions it in ocean bedrock. In order to better understand the process, we studied the stream located in Watershed 2 (W2). Calculations were completed in order to understand the difference in dissolved inorganic carbon (DIC) at the beginning of the stream and at the weir/outlet. DIC produced by the chemical weathering of bedrock in W2 yield flux values that are 250 mol ha^{-1} yr^{-1} higher than that measured at the weir. There are three proposed explanations for the loss of DIC between chemical weathering and the weir/outlet. The first is that carbon may be consumed by biomass. A second possibility is that CO_2 degassing of the stream may be occurring. Lastly, carbon may be utilized for in-stream metabolism; however, previous work by Worrall et al. (2005) found that in-stream

metabolism in W2 was negligible. There is evidence that degassing may occur because the pCO_2 of the W2 stream is $10^{-3.06}$ atm, above that of the atmospheric value of $10^{-3.5}$ atm. In order to determine if the DIC loss is the result of biomass uptake or stream degassing pH measurements were collected at several locations in the stream during baseflow. If the pH measurements increase from source to outlet (weir) then it can be concluded that degassing is occurring. However, if pH measurements decrease from source to outlet, it could be the result of biomass uptake and not from stream being over pressurized with respect to CO_2. The first data collection event showed a decrease in pH from the source to the weir, indicating that the biomass may be playing a large role in the removal of the CO_2. However, given the overpressurization of the stream with respect to CO_2, the degassing cannot be ruled out. In order to make a more definite conclusion additional pH data is needed when the vegetation is dormant.

14. Measuring the Force of Light

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A light source is directed onto a mirror that is magnetically levitated so that it will recoil in response to the force of the photons. By measuring the rotational response of the mirror assembly, the force is calculated. In order to minimize thermal effects due to energy dissipation in the form of heat, this experiment is performed under a high vacuum. The highest vacuum achieved was 1.4x10-6 torr with the force of a 19.1mW laser measured to be (1.81±0.59)x10-11 Newtons. The expected value of the force of the laser is 5.88x10-11 N with eighty percent of the light transmitted through the glass of the vacuum chamber, forty-five percent reflected by the mirror, and twenty percent transmitted through the mirror.

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