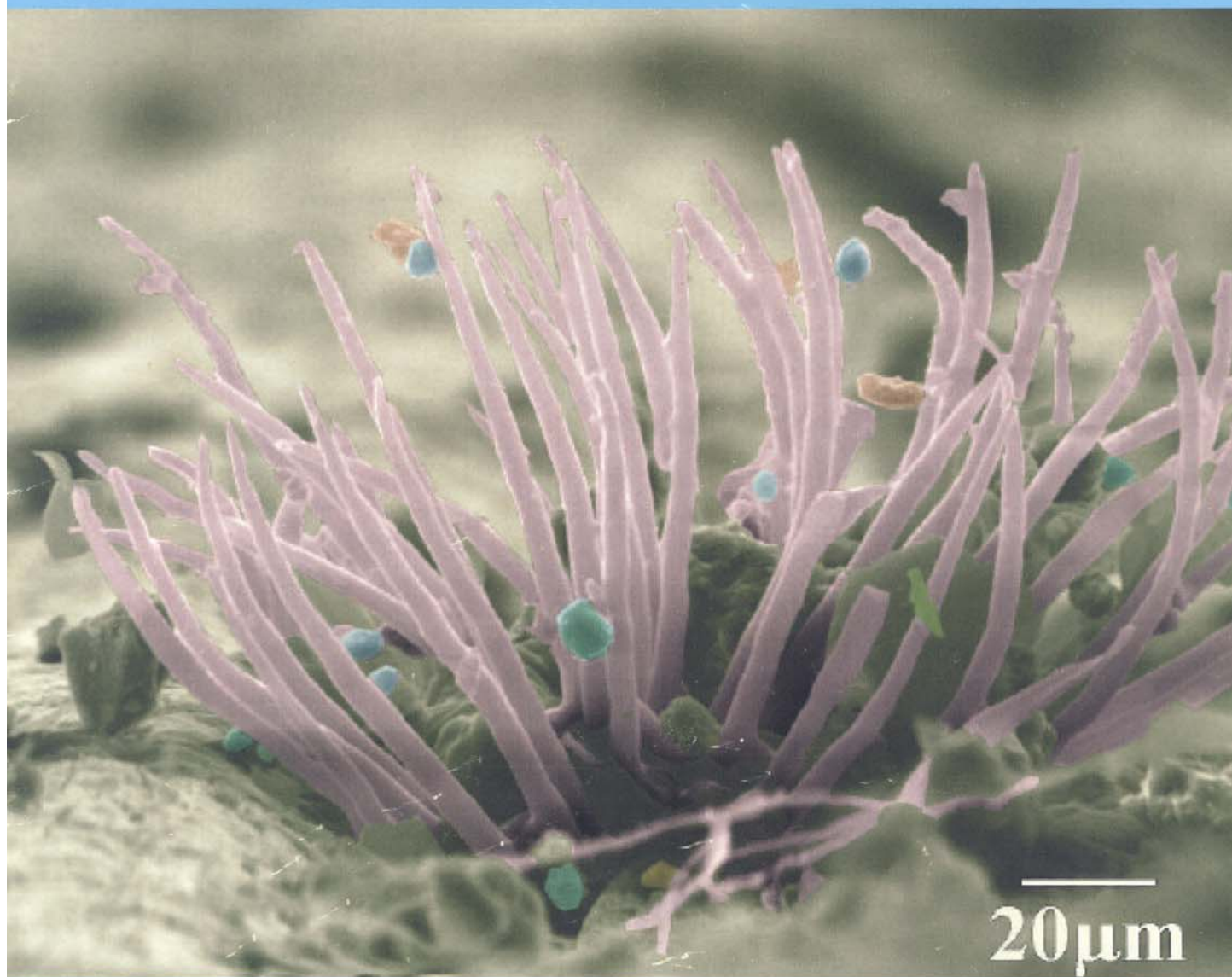




# *Texas Journal of Microscopy*



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**"TSM - Embracing all forms of microscopy"**  
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## PRESIDENT'S MESSAGE

When I look back over past years, it is evident that the Texas Society for Microscopy meeting activities succeeded in keeping pace with the fast moving developments in the world of microscopy. This is due in large part to the efforts made by previous executive councils as well as regular, corporate, and student members, most of whom are active members today. The field of microscopy is more diverse than ever and more integrated with other sciences and technologies, among those nanotechnology, medicine and materials science. The economic downturn and therefore the need to update our knowledge should increase the importance, visibility and work of our Society towards fulfilling its mission. Recent advances in microscopy instrumentation, automation and image analysis reveal both the new, finer atomistic details of material and cellular makeup and, at the same time, their organization on larger scales. There have been numerous variations on the types of microscopy in use nowadays, such as acoustic microscopy, x-ray microscopy, near-field optical microscopy (scanning near-field optical microscopy has recently shown its great potentials for fabricating various structures at the nanoscale), multiphoton imaging, and atomic force microscopy. One of the most amazing recent developments in microscopy involves the manipulation and rearrangement of individual atoms, opening up new directions in microscopy, where the microscope is both an instrument with which to observe and to interact with microscopic objects.

Recent advances in microscopy also have facilitated an effective convergence of biochemistry and cell biology to the point where we can visualize molecules and cells, and individual molecules within cells, in real time. New fluorescent probes and super-resolution light microscopy techniques, such as FRET and scanning probe microscopy, have greatly facilitated the study of dynamic processes in living cells. Atoms and bonds within individual molecules could be clearly resolved. New advances in intravital microscopy (IVM), such as two-photon microscopy, imaging chambers, and multicolor and fluorescent resonance energy transfer imaging, have recently been used to visualize the behavior of single metastasizing cells at subcellular resolution [J Cell Sci. 124(3): 299-310, 2011]. Microscopy using scanned neutral atom beams, technique called Atomic DeBroglie Microscopy, Neutral Beam Microscopy, and Scanning Helium Microscopy is a practical reality. Using thermal energy (under 70 meV) gas particles with neutral charge results in a probe beam that scatters from the first atomic layer of samples, with little chance of beam damage and no need of an aperture in close proximity to the sample. Molecular beam experiments show a wide range of surface properties, some that are difficult to see otherwise [Rev. Sci. Instrum. 82:10 (2011)].

What are future trends in microscopy? Most likely probe features within the atom? Whatever they are, they promise to be unbelievably exciting. Our Society needs to increase the breadth of its programs and activities to expose its members to a range of new techniques and tools that might benefit their current work, or that could be useful in their careers in the future.

This year's meeting program is taking shape nicely. I am grateful to members of the Executive Council for helping with meeting planning and other tasks. Ernest Couch has been a terrific Program Chair. He and Kevin Cronyn deserve much of the credit for the smooth planning process for our 2012 annual meeting at TCU in Fort Worth. Bob Droleskey deserves additional special thanks for taking on two extra jobs, the application for the re-instatement of the TSM non-profit organization status (pending) and work on the Society's finance records for a smooth transition to the new treasurer. His dedication and hard work for the Society are commendable. Many thanks to Nabarun Ghosh and Jennie Wojtaszek for initiating the Society's Facebook site (please follow us on Texas Society for Microscopy Facebook), to Ann Ellis and Michael Pendleton for formatting and publishing *Texas Journal of Microscopy* and to Becky Holdford for updating the Society's web page. The Society plans to upgrade its website and thus have a

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President's Message cont.

the website. The Society plans to upgrade its website and thus have a more efficient online presence to induce future growth. Balancing the use of electronic and social media along with the transition from regular mail to email brings us closer to maximizing use of mainstream information sharing technologies for effective communication.

TSM membership seems to have plateaued in recent years. The Executive Council continues searching for ways of increasing membership in all categories. To assist us in anticipating and understanding future needs of the Society, the Executive Council will send out a member survey. Please take the time to respond to this survey. What do we want our Society to be in the new era of technological advances in microscopy? Are we ready to go back to two meetings per year? I would like all TSM members to think about these issues and give their input to the Executive Council. This information will greatly aid us in developing strategies for increasing and better serving the membership. With proper planning and execution, the Executive Council expects to continue providing interesting and timely meetings that attract a diverse group of microscopists and increase meeting attendance and quality and number of presentations. Of a more urgent matter at this meeting, we should be able to put together a slate of nominees for next year's board members. All members are invited to run for office and I would encourage any of you to consider participating at whatever level feels comfortable. Our Society really is a great and supportive organization.

Financially, the Society has maintained a strong positive balance (over \$22,000 balance) for the past several years. In part, this is done through the generous support of our corporate sponsors. The corporate sponsors deserve special thanks for participation and stable financial support.

The Executive Board has made conscious efforts to minimize meeting registration costs and for this year's meeting grateful thanks go to Ernest Couch, Program Chair and to the TCU administration for the substantial donation towards covering the cost for renting the meeting rooms.

It has been a privilege serving as your President and I would like to thank all the membership for your continued support to the Society.

Sincerely,

Camelia Maier

TSM President 2011-2012



Dibyendu Dutta,, TWU, 1st place graduate student winner of the Howard J. Arnott Student Competition Award at the TSM 2011 meeting. Photo by N. Mills.



Howard J. Arnott Student Competition Award winners Jennie Wojtaszek (L,) (2nd place graduate TWU) and Yemisirach D. Gebeto (R.) (1st place undergraduate TWU) with TSM President Camelia Maier at the 2011 TSM meeting. Photo by N. Mills.



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# ABSTRACTS

## SPRING 2012

### BIOLOGICAL SCIENCES

#### PLATFORM PAPERS

##### STUDIES OF SEXUAL DIMORPHISM INVOLVING MINERAL DEPOSITS IN DIOECIOUS WHITE MULBERRY (*MORUS ALBA* L., MORACEAE)

J. CUFF and C. MAIER, Department of Biological Sciences, Texas Woman's University, Denton, TX 76204-5799

White mulberry (*Morus alba* L., Moraceae) is a dioecious plant species in which the sexes are separated in female and male trees. Several morphological, anatomical, and physiological traits were described to be different in male vs. female plants and are known as sexual dimorphism characteristics. Recent research showed that silkworms prefer male mulberry leaves, which contain less calcium mineral deposits and oxalate than female leaves. Silica mineral deposits also were found in white mulberry. In this study, silica mineral deposits were compared between sexes since silica is known to enhance resistance against herbivores in other plants. It was expected that mulberry female leaves contain more silica cystoliths than male leaves. Cystoliths were separated under a stereoscope from mulberry leaf ethanol extracts and observed in the Hitachi T1000 SEM. The elemental analysis was performed on 100 cystoliths for each male and female mulberry plant. No significant differences were found between the number of silica cystoliths between mulberry male and female plants. The majority of cystoliths in both male and female plants were composed of calcium and very few of silica. However, in both sexes significantly more neighboring cystoliths (much smaller, irregularly shaped, found in cells surrounding a much larger spherical cystolith) were found to contain silica than calcium. EDS analyses of cystoliths also revealed high abundance of certain elements in each sex. Aluminum was abundant in mulberry female cystoliths, while phosphorus was abundant in male cystoliths. In conclusion, white mulberry plants used in this study did not show sexual dimorphism involving silica depositions. It seems that both male and female mulberry plants metabolize and use silicon in a similar fashion.

##### LOVASTATIN DECREASES FOCAL COMPLEXES IN NEUROBLASTOMA GROWTH CONES

SHWETA DESHMUKH and DIANNA L. HYND, Department of Biology, Texas Woman's University, Denton, Texas, USA

Proper development of the nervous system requires efficient and precise axon growth and guidance, processes regulated in large part by guanine triphosphatases of the Rho family. Rho GTPases activation by binding GTP is facilitated by geranylgeranylation and membrane targeting as well as through bi-directional communication with  $\beta 1$  integrin containing focal complexes. Prior work demonstrated that decreasing geranylgeranylation decreases the neurite initiation and increases the loading of Rho A in the cytosol. Lovastatin, which is the inhibitor of HMG-CoA reductase, promotes neurite outgrowth in some system and inhibits it in others. Here we show that lovastatin affects the focal adhesion complexes in neuroblastoma growth cones. We use co-immunoprecipitation and immunocytochemistry to assess the focal adhesion of neuronal growth cones on different substrata and using treatments with lovastatin and lovastatin+geranylgeranylation. We find focal adhesions consisting of paxillin and vinculin complexes on glass showing little difference with lovastatin or lovastatin+geranylgeranyl. On laminin and collagen, we find decreased focal adhesion with lovastatin or lovastatin+geranylgeranyl. Lovastatin alters the morphology of cells, causing them to be rounded in shape with fewer more branched extensions, while geranylgeranyl rescues this alteration caused by lovastatin. Immunocytochemistry results qualitatively support the immunoprecipitation results. Data derived from these studies will be used to improve our understanding of axon growth, facilitate the construction of a predictive model of axon growth, and potentially provide novel targets for therapeutic interventions to promote axon regeneration.

##### MORPHOLOGICAL ANALYSIS OF ACOUSTIC STRUCTURES OF *TROPISTERMUS LATERALIS NIMBATUS* AND *TROPISTERMUS COLLARIS MEXICANUS*

DANIEL DONLEY, Eastfield College, Mesquite, TX 75150

The purpose of this study was to determine if the morphology of sound producing structures of *Tropisternus lateralis nimbatus* and *Tropisternus collaris mexicanus*, water scavenger beetles (Coleoptera:



Donley cont.

Hydrophilidae), results in species specific calling behavior. This research focuses specifically on morphology associated with sound production and acoustic behavior at varying temperatures. Past studies state that calling behavior is species specific. This specific acoustic behavior may produce barriers concerning inter-breeding. Size variations among the pars stridens (sound producing structure) of twenty one specimens from two different species were measured using a scanning electron microscope (SEM). Calls from the specimens were recorded and analyzed for differences in frequency, duration, and behavior. Based on the findings, size among the structures separates the two species and may result in species specific calling behavior. Further studies of these two species are needed to interpret acoustic behavior and the presence and function of inter-breeding barriers.

#### ALTERNATIVE STAINS FOR LIPIDS FOR TRANSMISSION ELECTRON MICROSCOPY

E. ANN ELLIS, Microscopy & Imaging Center, Texas A&M university, College Station, TX 77843-22576

Most studies for transmission electron microscopy (TEM) result in electron dense deposits at the site of staining and the specificity of many stains is not very well known. Osmium tetroxide (OsO<sub>4</sub>) binds to moieties other than lipids, does not stain some types of lipids very intensely and is not an exclusive indicator of lipids. Malachite green and *p*-phenylenediamine (PPD) have been used in the past to amplify the staining of lipid components after or in conjunction with osmium.

Malachite green is water soluble and is used in concentrations of 0.05-0.1% (wt/vol) in the primary fixative. This is an intense stain and has been used to preserve and identify the waxy layer of leaves, to trace the sites of production and translocation of waxes. Malachite green has also been employed in staining the cuticle in *Caenorhabditis elegans*, *Mycobacterium tuberculosis* and other microorganisms.

PPD has been used extensively in cardiovascular studies in atherosclerosis and amplifying carotene bodies from fruits and vegetables. PPD is used at 0.5% (wt/vol) either as a substitute for osmium or in the dehydration steps. When used in the dehydration steps for cell cultures, areas of cells are much easier to identify. PPD also improves antigenic preservation for colloidal gold based immunolabeling.

#### INVESTIGATION OF CELL WALL STRUCTURE OF $\Delta$ CLPX *BACILLUS ANTHRACIS*

C. EVANS, E. COUCH and S. MCGILLIVRAY, Dept. of Biology, Texas Christian University, Fort Worth, TX 76129

ClpXP is an intracellular protease that regulates the life span of multiple bacterial proteins such as transcriptional regulators, rate-limiting enzymes and damaged proteins. ClpXP consists of two proteins, ClpP, the proteolytic core, and ClpX, a regulatory ATPase. ClpXP is conserved across many bacterial species and is often associated with cellular stresses such as heat shock, nutrient deprivation and oxidative stress. ClpX/P has also been implicated in the virulence of several pathogens. We recently demonstrated that ClpX is critical for the pathogenesis of *Bacillus anthracis*, a Gram-positive bacterium that is the causative agent of anthrax, and that the loss of ClpX leads to susceptibility to multiple cell wall-acting antimicrobials. We hypothesize that the misregulation of cell wall genes due to the loss of ClpX leads to a change in cell wall structure, making it susceptible to cell-wall targeting antimicrobial agents. In order to gain a better understanding of how the loss of ClpX is affecting *B. anthracis* cell wall, we are investigating gross differences in  $\Delta$ ClpX *B. anthracis* using scanning and transmission electron microscopy.

#### DISCOVERY OF NOVEL VIRULENCE FACTORS IN *BACILLUS ANTHRACIS*

S. ELIZABETH FRANKS<sup>1</sup>, ERNEST COUCH<sup>1</sup> and SHAUNA M. MCGILLIVRAY<sup>1,2</sup>, <sup>1</sup>Department of Biology, Texas Christian University, Fort Worth TX and <sup>2</sup>University of California, San Diego, La Jolla CA

*Bacillus anthracis*, the causative agent of anthrax, must avoid an array of antibacterial defenses by the host during the course of infection. Although anthrax toxin and capsule, located on two large plasmids, play important roles in the pathogenesis of this disease, evidence indicates that chromosomal genes also contribute. We have generated a random chromosomal mutant library of *B. anthracis* Sterne using a mariner-based transposon mutagenesis system in order to identify novel chromosomal virulence factors. Bacterial mutants were screened for loss of virulence in the invertebrate animal model *Caenorhabditis elegans*. *C. elegans* were fed *B. anthracis* Sterne under conditions that enabled up to 80% of worms to become infected. Bacterial mutants unable to infect *C. elegans* were selected for and the site of transposon insertion was identified. *In vitro* assays assessing susceptibility to specific antimicrobial defenses were used to identify potential mechanisms of action of attenuated mutants. Approximately 0.25% of the mutants screened were attenuated in their ability to infect *C. elegans*. The most highly attenuated mutant was found to have a disruption in an operon containing multiple tellurium resistance genes. Tellurium resistance genes are conserved throughout bacteria and aid in the reduction of tellurium to a less toxic compound resulting in the formation of black tellurium deposits in the cell



wall. We find the mutant bacteria are more susceptible to tellurium toxicity. We will investigate differences of the WT and mutant bacteria to form these deposits using transmission electron microscopy. In addition to increased susceptibility to potassium tellurite, we found this mutant is more susceptible to reactive oxygen species, such as H<sub>2</sub>O<sub>2</sub>. The tellurium resistance operon of *B. anthracis* may have an additional novel role in resistance to reactive oxygen species, a critical host defense.

#### MUTATING ARP3 RESIDUES DISRUPTS WAVE1 AND ARP2/3 COMPLEX INTERACTIONS AND DECREASES NEURITE OUTGROWTH

S. HALDAR, A. S. MAHADIK, B. W. BECK and D. L. HYND, Department of Biology. Texas Woman's University, Denton, TX 76204-5799

Axon extension results from branched actin nucleation and polymerization leading to lamellipodial expansion, a process regulated by interaction of actin binding proteins Arp3, of the Arp2/3 complex, and WAVE. However, the amino acid residues in Arp3 that contribute to this association are not yet identified. To accomplish this, we predicted arginine 161 (R161) to have the most contribution to Arp2:Arp3 interactions. We have expressed wild-type and R161A mutant clones of Arp3 fused at the N-terminal with green fluorescent protein (GFP) in B35 neuroblastoma cells and determined how expressing the mutant affects Arp2/3 complexing with WAVE, actin filament content and neurite outgrowth using immunocytochemistry, co-immunoprecipitation and image analysis. Expression of R161A decreased neurite outgrowth and increased actin filament content. We interpret these data to indicate that R161 is important for assembling the Arp2/3 complex, perhaps indicating a potential site for therapeutics to promote axon regeneration following traumatic and neurodegenerative lesions.

[Supported by the TWU Department of Biology and a TWU Multidisciplinary Grant]

#### CORRELATION OF SPORES BETWEEN *RUSSULA OCHRICOMPACTA* AND *RUSSULA EARLEI* (ANCIENT) VS. *RUSSULA MUTABILIS* AND *LACTARIUS LANUGINOSUS* (MODERN) USING THE SCANNING ELECTRON MICROSCOPE

STEPHANIE ROSE, Eastfield College, Mesquite, TX 75150

A study was conducted on the length and width of spores for four genera of fungi. Two ancient *Russula ochricompacta* and *Russula earlei* were compared with two modern mushrooms, *Russula mutabilis* and

*Lactarius lanuginosus*. The two ancient *Russulas* were more similar in size than the modern mushrooms. Spore sizes of the four genera that were collected and measured did not show the same dimensions. The sizes of the modern spores were similar in size to the four specimens in the out group (*Russula compacta*, *Russula eccentrica*, *Russula vesicatoria*, and *Lactarius arcuatus*). In conclusion, comparing ancient to modern species displayed few size similarities and the data obtained did not support the original hypothesis.

#### SEM OBSERVATIONS OF *ACAULOSPORA SPOROCARPIA*, SEM OF THE RECENTLY-ERECTED FUNGAL CLASS GLOMEROMYCETES WHICH ARE KNOWN AS ARBUSCULAR MYCORRHIZAL FUNG

BILLY STONE, Fort Worth, Texas

Arbuscular Mycorrhizal Fungi (AMF) are described as fungi which form specific types of mycelial connections with vascular plants. These associations are generally thought to be mutually beneficial, with the fungal hyphae providing to the plant an increased potential for absorption of water and uptake of phosphorus and other nutrients in exchange for carbohydrates produced through photosynthesis. Only two verified collections of the AM fungus *Acaulospora sporocarpia* have ever been made. The "type specimens" were collected in 1955, about 40 miles SW of Winslow, AZ, though they were not described and added to the scientific literature until 1985. The second collection, examples pictured, was made near Ribera, NM, in September 2006, more than 50 years after the first. To date, these remain the only known encounters with this particular fungus in the wild.

The infrequency of these finds can be partly explained by observing the environment these "balls of dirt" inhabit. The habitat is high desert/grassland with an average annual rainfall in the single digits, which is not generally considered by mushroom enthusiasts to be particularly fertile hunting ground for fungi. Consequently, very little extensive fieldwork has been done in these areas and therefore, if one happens to be in the right place at the right time, the potential to make rare or unusual discoveries remains fairly high. *A. sporocarpia* is rare even among its kind in that it produces a "sporocarp" (spore-bearing structure) containing hundreds or thousands of spores, unlike most of its relatives which generally produce spores only singly or in pairs. Scanning electron microscopy provides a revealing look at this truly unique organism.

#### REGULATION OF Bcl2 FAMILY OF GENES IN TESTES BY TESTOSTERONE

ARPITA TALAPATRA, DIBYENDU DUTTA, IN PARK, HIWOT GUILILAT, SAMUEL SANG and



NATHANIEL MILLS. Department Of Biology, Texas Woman's University, Denton, TX 76204.

Germ cell apoptosis is an important regulatory process during spermatogenesis. While testosterone depletion results in germ cell apoptosis, the underlying molecular events are unknown. To determine the levels of pro- and anti-apoptotic genes of the Bcl2 family in testes, male rats were injected with ethane dimethane sulfonate (EDS) (75mg/kg body weight) to selectively eliminate mature Leydig cells thereby ablating testosterone. Tissues were collected after 7-days of treatment for TUNEL assay, gene expression and hormone analysis. To substantiate testosterone's role, separate groups received exogenous testosterone for either supplementation or replacement of testosterone following EDS. Significant germ cell apoptosis in EDS-treated rats was demonstrated by TUNEL assay and testosterone replacement prevented the germ cell apoptosis. The Bcl2 gene family, pro-apoptotic BAK1, and anti-apoptotic BclW, Bcl2 and Mcl1 genes increased in testes of EDS-treated rats as detected by RT-PCR. We suggest that Bcl2 family of genes is involved in germ cell apoptosis without testosterone.

#### PHAGOSOMAL ESCAPE BY *RICKETTSIA PROWAZEKII*

TED WHITWORTH<sup>1</sup>, VSEVOLOD L. POPOV<sup>2</sup>, XUEJIE YU<sup>2</sup>, DAVID H. WALKER<sup>2</sup>, and DONALD H. BOUYER<sup>2</sup>, <sup>1</sup>Analytical Cytology Core Facility, Dept. of Biological Sciences, The University of Texas at El Paso, El Paso, TX 79968, and <sup>2</sup>Faculty of The Dept. of Pathology, The University of Texas Medical Branch at Galveston, Galveston, TX 77555

Members of the genus *Rickettsia* possess the ability to invade host cells and promptly escape from phagosomal vacuoles into the host cell cytosol, thereby avoiding destruction within the endosomal pathway. Previously, the mechanisms underlying rickettsial phagosomal escape were unknown. This study was undertaken to determine which of several selected genes from the annotated *Rickettsia prowazekii* genomic sequence mediate the escape process. Reverse transcriptase PCR analyses determined that two genes, *tlyC* and *pld*, were transcribed during the period of active phagosomal escape by rickettsiae. The functionality of both *tlyC* and *pld* was determined by complementation studies in *Salmonella*, which replicates within endosomes. Complementation of *Salmonella* organisms with either *tlyC* or *pld* resulted in the escape of transformants from endosomal vacuoles into the host cell cytosol as demonstrated by quantitative ultrastructural analyses. Immunogold electron microscopy experiments confirmed the presence of Pld

and TlyA within the disrupted phagosomal membranes during the period of escape. These data offer more evidence for a role of TlyC, Pld and TlyA in the process of phagosomal escape by *Rickettsia*.

[Supported by National Institute of Health Grant RO1 AI21242]

#### ESSENTIAL OILS AND REPRODUCTION IN WHITE MAGNOLIA, *MAGNOLIA GRANDIFLORA* (MAGNOLIACEAE)

MORGAN WILSON and CAMELIA MAIER, Department of Biological Sciences, Texas Woman's University, Denton, TX 76204-5799

It is known that floral scents play an important role in plant reproduction success. Floral scents are derived from essential oils, a heterogeneous group of plant secondary compounds. The essential oils are believed to play an important role in the attraction of pollinators. In white magnolia, *Magnolia grandiflora* (Magnoliaceae), flowers and seeds are aromatic due to essential oils glands found in the petals, androphore, and seed coat. The goal of this study was to localize the essential oil glands in Magnolia flower organs and identify the main oils involved in this species pollination, seed dispersal, and germination.

Microscopy techniques were employed for localization of the essential oil glands in seeds. The Magnolia seed has two coats originated from the two ovular integuments, which form three layers. The outer coat has a red fleshy layer outside and a hard, bony layer inside. The inner coat is thin and is the closest layer to the endosperm. Essential oil cells were found in the outermost layer of the seed coats, specifically in the red fleshy outside layer. They are large cells dispersed more or less uniformly in the fleshy tissue and containing dark material.

Magnolia flowers were collected in the morning before they open and insects found inside the flower were preserved in ethanol and observed under the microscope. Pollinator insects were the beetles *Trichiotinus piger*, *Conotelus obscurus*, and *Strangalina luteicornis* (Coleoptera). Dispersal of seeds on the TWU campus was mainly performed by squirrels, which are attracted by and eat the red fleshy outmost seed layer containing the essential oils. The outer seed coat is impermeable to water and gases and thus it is thought to prevent premature germination. Future work will focus on determining the function of essential oils in seed dormancy. When completed, this study will enhance our understanding of reproduction, dispersal and seed germination in *Magnolia grandiflora*.



## MATERIALS SCIENCE

### PLATFORM PAPERS

#### UNDERSTANDING THE RELATIONSHIP BETWEEN CRYSTALLOGRAPHY AND MATERIAL PROPERTIES WITH ELECTRON BACKSCATTER DIFFRACTION

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Electron Backscatter Diffraction (EBSD) is one of the most powerful microstructure characterization techniques that allows users to investigate the crystallography of the material. EBSD provides detailed microstructural information such as grain size, preferred orientation distributions, grain boundary character, strain distribution, and phase distribution. Applications of EBSD in a variety of industries, especially with alloy development, quality control, and failure analysis will be reviewed. The problems involved in working with volatile and beam sensitive materials will also be discussed.

#### INVESTIGATION OF METAL OXIDE ELECTROLYTE VIA ELECTRON MICROSCOPY

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Metal oxide electrolytes have been used extensively in fuel cell and gas sensing applications due to their ability to permit fast ion conductance as a solid electrolyte at slightly elevated temperatures[1]. Yttrium oxide-stabilized zirconium oxide (ZrO<sub>2</sub>) (YSZ) is among the most widely used materials for these applications.

In our research, the syntheses of nanostructured solid electrolytes were accomplished via two routes: (1) selected sol-gel reactions and (2) preformed suspended nanoparticles, each in conjunction with electrospinning techniques. The characterization of these materials was done primarily by Scanning Electron Microscopy (SEM), Transmission Electron Microscopy (TEM), and energy-dispersive X-ray (EDX) spectroscopy with a JEOL JSM-6100 SEM and JEOL JEM-2100 TEM. The data obtained from this investigation was used to develop an understanding of the relationship between nanowire structure and ion transport.

#### References

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#### CHARACTERIZATION OF COATINGS ON RFID P-CHIPS WHICH PROVIDE PLASMONIC ENHANCEMENT OF BIOASSAYS

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Mirotransponders (RFID p-Chips) derivatized with a silver island film (SIF) have been employed as a powerful multiplexing platform for the quantification of low-abundance biomolecules in nucleic acid-based assays and immunoassays. In this study, we further characterized the morphology of the SIF applied to p-Chips through scanning electron microscopy (SEM). The SIF and the entire p-Chip was enveloped by a silane-based polymer matrix, and, through a series of SEM and confocal fluorescence microscopy experiments, we found that the depth of the polymer matrix was 1-2  $\mu\text{m}$ . The polymer was conjugated to a fluorophore (Alexa Fluor 555), and the radiative effects of the SIF/polymer layer were assessed by fluorescence lifetime imaging (FLIM). FLIM images showed that the addition of the SIF/polymer layer was accompanied by an 8.5-fold increase in fluorescence intensity and a decreased fluorescence lifetime, the latter of which is associated with improved photostability. This fluorescence enhancement through plasmonic resonance was found to extend uniformly across the p-Chip and, interestingly, to a depth of about 1.2  $\mu\text{m}$ . The substantial depth of enhancement suggests that the SIF/polymer layer constitutes a three dimensional matrix which is accessible to solvents and small molecules, such as fluorescent dyes. Finally, we confirmed that no surface enhanced Raman scattering (SERS) is seen from the SIF/polymer combination. The analysis provides a possible mechanism by which the SIF/polymer coated p-Chips allow a highly sensitive immunoassay and, as a result, lead to an improved, multiplex, bioassay platform.

## SCIENCE EDUCATION

### PLATFORM PAPERS

A PROGRESS REPORT: "ENHANCING VOCABULARY ACQUISITION OF SCIENCE LANGUAGE IN A MIDDLE SCHOOL SCIENCE CLASSROOM USING INQUIRY-BASED SCIENCE METHODS, A WORD WALL, AND THE SCANNING ELECTRON MICROSCOPE"



C. SIEBER AND S. WESTMORELAND, Department of Biology, Texas Woman's University, Denton, TX, 76204

This report will show the progress of research that is currently being conducted to determine if three specific methods will enhance the vocabulary acquisition skills of English Language Learners in the sixth grade science classroom. Pictures of the classroom Word Wall and a bulletin board of micrographs taken from the scanning electron microscope will be presented. The purpose of this study in its entirety is to determine if vocabulary and language acquisition skills of English language learners can be accelerated by using three specific methods in a rural sixth grade science classroom in North Texas. This study will incorporate hands-on science learning through the use of Inquiry-Based Science Methods as well as a Word Wall and a Discovery Wall for micrographs taken with a scanning electron microscope. In this nine month, action research study, a mixed-method research design will be used to collect and analyze data from criterion referenced pre-tests and post-tests as well as qualitative sources of data. Descriptive statistics will be used to analyze data from the 2010-2011 school year baseline test and the 2011-2012 school year pre-test and post-test. A T-test will be used to analyze the data from the baseline test and the post-test of the study group. Additionally, qualitative data will be collected through a journal, photographs of the word wall, and photographs of the specimens that students chose to have micrographs taken under the scanning electron microscope. The specific research question that I am attempting to answer is, "Can vocabulary acquisition increase over a nine month study period by using a science Word Wall, a discovery wall with micrographs from a scanning electron microscope, and the process of Scientific Inquiry"? I predict that the incorporation of these specific techniques into the sixth grade science curriculum the scores of the English language learners will increase on the end of year vocabulary assessment by 10 % or higher as compared to the 2010-2011 baseline test results.

#### **INTEGRATING ELECTRON MICROSCOPY INTO THE GENERAL MICROBIOLOGY LABORATORY**

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A laboratory exercise to immerse undergraduate general microbiology students into hands-on investigations using scanning and transmission electron microscopes was developed. Students first learned the components, care and maintenance of brightfield

microscopes and viewed specimens such as plankton, yeasts and bacteria using low-power, high-dry, and oil immersion lenses. During the next lab period, the class met in the electron microscopy center where they learned about preparing specimens for viewing and how to operate both scanning and transmission electron microscopes. The students then worked in teams to view and interpret electron micrographs of specimens of bacteria and fungi. Results of pre/post testing revealed that this approach helped the class to appreciate the array of microscopic tools available and to understand the applications of each instrument.

## **BIOLOGICAL SCIENCES**

### **POSTERS**

#### **MICROSCOPIC EVALUATION ON AEROALLERGEN AND REDUCTION ON CANINE ALLERGY INDEX ON USING AIR PURIFIERS WITH ADVANCED HYDRATED PHOTO-CATALYTIC OXIDATION**

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Aeroallergen sufferers are not excluded to only humans. Dogs suffer from inhalant allergies like humans, but the difference is in how they show their symptoms. Canine atopic dermatitis is the most common sign of allergies in dogs and can generate many other secondary ailments. According to the SPCA about 20 % of pets suffer from allergies. Treatments of allergies range from oral medication, topical treatments, hypo-sensitization or avoidance. We have assessed the efficiency of air purifiers in reducing the indoor aeroallergens to control the allergy related ailments. The wall mount Nano Air purifier was used to assess the concentration of indoor aeroallergens at Coulter Animal Hospital in Amarillo, TX. Air Oasis air purifiers utilize advanced hydrated photo-catalytic oxidation (AHPCO) technology that produces very low doses of negative ions that remove the aeroallergens, dust particles, animal dander etc. from the air electrostatically. We carried out experiments with Nano with airborne mold, bacteria and VOCs, at the BSA Hospital with MRSA and at the Coulter Animal Hospital, Amarillo, TX with animal dander, VOCs and airborne mold. Slides with double sticky tape were exposed to room air for 24, 48, 72 and 120 hours before and after running the Nano air purifiers. The exposed slides were stained with 2% safranin and observed using a BX-40 Olympus microscope, DP-70 digital camera. The images were



analyzed with Image Pro 6.0 software. The data were correlated with the aeroallergen index and the frequency of inhalant allergy cases in dogs. GC-MS Spectra analysis data showed a gradual reduction of the VOCs in the indoor air in the clinics after running the Nano air purifiers at different intervals. Analysis of slides and the petri plates exposed at different intervals of 24 hr, 48 hr, 72 hr and 120 hr showed reduction in aeroallergen after running the air purifiers. The high indoor VOC concentrations reduced on running the Nano Air Purification units. Indoor aeroallergens such as, mold spores, airborne bacteria and animal dander reduced significantly. Experiments utilizing the Nano air purifiers established that the use of a negative ion purification system is an effective means of eradicating aeroallergens such as mold and microbes in room air. Allergy indices reduced considerably on reduction of VOCs and aeroallergens in the room air after running the air purifiers.

#### **IDENTIFICATION AND EFFECTS OF NECROTROPHIC FUNGAL PATHOGENS ON INDIANGRASS (*SORGHASTRUM NUTANS*)**

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Indiangrass (*Sorghastrum nutans*) is a perennial warm-season bunch grass susceptible to a number of fungal pathogens. The purpose of this investigation was to compare symptomatic and asymptomatic cultivars of indiangrass obtained from the East Texas Plant Materials Center, Nacogdoches, TX using a three-pronged approach: 1) evaluate the physiological effects of fungal infection through studies of photosynthesis, light saturation, and chlorophyll quantity, 2) examine the host/pathogen relationship through various forms of microscopy, and 3) determine fungal presence using a polymerase chain reaction (PCR) assay. Symptomatic and asymptomatic leaf samples were tested for photosynthetic response under decreasing light intensities and separately under established light saturation using a LI-COR 6400XT Portable Photosynthesis System. Symptomatic leaves consistently exhibited decreased physiological activity. Through microscopic analysis acervuli of the fungal pathogen *Colletotrichum* were identified on 16.7% of symptomatic leaves. Epifluorescence light microscopy and transmission electron microscopy revealed limited

growth of intercellular/intracellular hyphae and extensive disintegration of plant cells, consistent with the necrotrophic phase of *Colletotrichum* infection. Three distinct fungal phenotypes were cultured from infected tissues. Molecular DNA studies verified the presence of fungal DNA in both symptomatic and asymptomatic samples using a primer pair specific for ascomycetes and basidiomycetes.

#### **COMPARATIVE ANALYSIS OF SELECTED MOSS CAPSULES AND PERISTOME MICROMORPHOLOGY UTILIZING SCANNING ELECTRON MICROSCOPY**

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Patterns of capsule surfaces and morphometric variability in ten species of moss were investigated. Length of peristome teeth, length and width of capsule, and diameter of spore openings were measured. Capsule surfaces were examined for elevations, depressions, and cell length. Morphological distinctions between capsules and peristomes were expected to vary in all ten species. The examination of peristome was indeterminable due to broken or absent teeth. Capsule measurements showed a diverse range of sizes among the species observed. Utilizing a scanning electron microscope, measurements were taken and recorded. The capsule and diameter of spore opening as well as the surface details can be used to identify and classify species.

#### **EFFECTS OF STORAGE CONDITIONS ON THE MORPHOLOGY AND TITER OF LENTIVIRAL VECTORS**

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Lentiviral vectors are commonly used in laboratory experiments to stably integrate transgenes into host genomes. These lentiviral vectors are constructed in a tissue culture setting by simultaneously transfecting three expression constructs into Human Embryonic Kidney (HEK) 293T cells. The transfected cells shed viral particles into the media which is then collected. It has long been observed that freshly collected virus-containing media has the highest titer and that storage of the media leads to a decreased titer, but the mechanisms driving this decrease have yet to be identified. To that end we have generated and collected lentiviral vector stocks and stored them in several different conditions. Specifically, aliquots from an initial viral collection were



stored at: (1) room temperature for less than one hour, (2) -80°C for 24 hours, (3) 4°C for three days and (4) 4°C for 7 days. These stocks were subsequently evaluated with regard to their transducing ability and morphology. To assess transducing ability, each viral stock was used to transduce fresh HEK 293T cells. Cells were visualized using fluorescence microscopy to estimate the percentage of transgenic cells which express Green Fluorescent Protein (GFP) as a part of the inserted transgene. The virus stored at room temperature for less than one hour exhibited the highest titer and was able to infect ~90% cells. In contrast, virus stored at -80°C for 24 hours had a lower titer only infecting ~60% cells. The virus that was stored at 4°C for three and seven days infected ~50% and ~30% - 40% of the cells respectively. In addition, we will measure GFP production quantitatively using the Typhoon Trio Plus biomolecular imager (GE LifeSciences) and ImageQuant software system (GE LifeSciences). Viral morphology observed with transmission electron microscopy (TEM) was characteristic of that of family Retroviridae, with virions spherical to pleomorphic in shape and 80-100 nm in diameter. Both storage conditions reduced the number of intact virions identifiable under TEM.

#### **EFFECTS OF LANTHIONIZATION ON NATURAL AFRICAN AMERICAN HAIR**

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Hydroxide relaxers are used by African American women to straighten hair to create a smoother and more manageable texture of hair. Hydroxide relaxers rely on the cleavage of disulphide bonds to allow the hair to relax and be pulled straight. The alkaline relaxer penetrates the cortex and breaks the structural disulphide bonds of the hair (Harrison and Sinclair, 2003). This process is called *lanthionization*. When a hydroxide relaxer breaks a disulfide bond the bond is permanently broken and can never be reformed.

For this experiment samples of unprocessed, virgin hair (control) underwent the lanthionization process. A total of 4 samples were used, each representing a 15 minute time difference (A:15 minutes, B:30 minutes, C:45 minutes, and D:60 minutes) to compare and contrast the effect of how the lanthionization process affects the qualitative structure of hair over time. Micrographs of "relaxed" hair from each time interval were compared to the structure of the untreated hair (control) to view the similarities and differences from the lanthionization process using the Hitachi TM 1000 scanning electron microscope. We hypothesized that

after using a regular strength no-lye relaxer on untreated hair, the overall appearance of the hair would appear more damaged as the time intervals increased. The hypothesis was confirmed as a result of micrographs showing a noticeable difference in the amount of weathering found on hair strands from the control sample and sample D which showed the largest amount of weathering when compared to the remaining samples.

#### **References**

Harrison, S. and Sinclair, R. 2003. *J. Cosmetic Dermatology* 2(3):180.

#### **TESTERONE REGULATES BLOOD-TESTIS BARRIER PERMEABILITY FOR MACROMOLECULES AND IONS VIA REGULATION OF TRICELLULIN AND CLAUDINS 5 AND 11**

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Testosterone (T) regulates genes in males needed for fertility and virility. Testosterone withdrawal causes up or down regulation of target genes. Using ethane dimethane sulfonate (EDS) we selectively destroyed Leydig cells in adult rat testis and then examined genes under testosterone control that may be involved in blood-testis-barrier formation. In mammalian testis spermatogenesis takes place in a unique microenvironment behind the blood-testis barrier (BTB), which is created between the adjacent Sertoli cells near the basement membrane of the seminiferous epithelium. Among other functions, BTB acts to regulate the paracellular diffusion of water, electrolytes, nutrients, and biomolecules from the systemic circulation in the interstitium to the developing germ cells.

Tricellulin and claudins are tight junction proteins that are thought to mediate cellular junctions via protein:protein interaction as part of the signaling pathway for regulation of the paracellular barrier permeability necessary for maintenance of the BTB. Here we show that withdrawal of testosterone following EDS treatment results in down regulation of tricellulin and claudin.

[Support: Research Enhancement Program 2010- TWU]

#### **LEYDIG CELL MEMBRANE RECEPTORS LOSS IN EDS TREATED ADULT RATS AT EARLY TIME POINTS DUE TO LEYDIG CELL APOPTOSIS**

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Testosterone levels in serum following treatment of



male rats with ethylene dimethane sulfonate (EDS) to induce apoptosis of Leydig cells has traditionally been the marker used to establish the effectiveness of the drug treatment. Using RT and qPCR, we assayed LH receptor mRNA expression (*Lhr*) and insulin-like 3 (*Insl-3*) expression and have found these genes to be primarily expressed in Leydig cells of rat testes since *Lhr*, *Prl-R* and *Insl-3* are all very low in 5 days and 7 days post-EDS treated rats. Therefore, we assayed these genes with the expectation that loss of these mRNAs would be a more rapid and accurate indicator of Leydig cell depletion than using testosterone decline in serum. Using this assay we observed a different time profile for *Lhr* loss and *Insl3* loss although these markers are in the same cells. In addition we have used the TUNEL assay that detects DNA fragmentation at later time points of cellular apoptosis to evaluate when Leydig cell loss was well under way. With TUNEL, we detected Leydig cell apoptosis at early time points of 6 hours (hr), 15 hr and 24 hr after EDS. To further evaluate the loss of genes in apoptotic Leydig cells, we next evaluated the prolactin receptor mRNA (*Prl-R*) to compare with the time of loss profiles of LH receptor mRNA expression (*Lhr*) and insulin-like 3 (*Insl-3*) expression with Leydig cell ablation. The TUNEL assay confirmed that the Leydig cell apoptosis is time dependent and it increased with time post-EDS. *PrlR* declined with cell loss and testosterone supplement also suppressed its expression. We found that *Insl-3* had reduced mRNA expression with T supplementation. *LhR* decreased with time but not at the same rate as *Insl-3*, and exogenous testosterone does not seem to be responsible for suppression of the *LhR* expression. We further find that with EDS treatment there is loss of *PrlR*, *LhR* and *Insl-3* and all 3 appear to decline at different rates as Leydig cell are depleted.

[Texas Woman's University- Research Enhancement Program & Department of Biology.]

#### GERMINATION TECHNIQUES FOR SELECTED BIRD'S NEST FUNGI IN GENUS *CYATHUS*

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The bird's nest fungi (genus *Cyathus* Haller) have a characteristic basidiocarp morphology, consisting of a cup resembling a tiny bird's nest filled with peridioles (spore-bearing structures) that look similar to eggs. Since the mid-1800s, mycologists have debated the germination requirements for this group. This study tested five techniques previously reported to stimulate germination using dried peridioles of *C. annulatus* and *C. striatus*. The methods investigated were: (1) immersion in water, (2) direct plating onto water agar, (3) presoaking in water prior to direct plating, (4)

rupturing of peridioles prior to direct plating, and (5) scarification in acid-pepsin combinations prior to direct plating. The latter treatment simulates passage through the gastrointestinal tract of an animal, as bird's nest basidiocarps are frequently found on animal dung. *C. annulatus* germinated more readily than *C. striatus* under all conditions, exhibiting near 100% germination rates with methods 1-4. *C. striatus* peridioles germinated at rates of 50% or less regardless of the treatment. The scarification treatments generally had a detrimental effect on germination rate and subsequent mycelium growth in both species. Light and scanning electron microscopy were used to examine germinated specimens and revealed the presence of intercalary chlamydospores and fruitbody primordia.

#### THE RELATIONSHIP OF STRUCTURE AND FUNCTION OF *CORALLUS CANIUS* SCALES

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*Corallus canius* commonly referred to as the emerald tree boa, is a reptile that thrives mainly in the Amazon Basin of South America. Even though emerald tree boas can grow up to 9 ft they tend to habitat only in trees. These creatures have some of the largest teeth in the snake family. However, they are non-venomous and feed instead by suffocating their prey to death. Keeping these factors and functions in mind I propose that samples taken from different locations of the emerald tree boa will show structural differences that reflect their function. Using the Hitachi III Tabletop scanning electron microscope, I studied the skin shedding of the emerald tree boa at different locations of the body. Using a dissecting microscope, samples were taken from the jaw, mid-back, and mid-belly; these samples were then examined and imaged using the Hitachi 1000 Tabletop scanning electron microscope. Scales from the belly of the snake, which is exposed to mainly to tree, are much different than that of the scales examined of the back of the snake. Taking ten measurement of each area I have found these differences to be present: the back of emerald tree boa measured at 800X has an average scale size of  $9.68 \pm 0.75 \mu\text{m}$  while the belly taken at the same magnification has an average size much bigger averaging  $13.22 \pm 1.23 \mu\text{m}$ . Pictures indicate that the scales near the jaw and mouth are much smaller and compact than those on their back or belly with an average scales measurement to be  $6.87 \pm 0.97 \mu\text{m}$  under 800X magnification. These preliminary results indicate that samples taken from areas on the body that are more mobile (the jaw) have much smaller scales than areas of the body that are less mobile such as the back and the belly of the snake. The morphologies of the different locations coincide with the measurements taken. The



mid-belly of the snake has the roughest texture as well as the largest scales; the mid-back has the second largest scale size as well as the second roughness in texture, while the jaw has the softest and smallest scales. These findings support my hypothesis in that the scales of a snake are like chain-mail, the smaller and softer the scales the more movement allowed, such as when the jaws open to feed. Emerald tree boas spend approximately 99 % of their time in one position, the exception being during feasting and mating, this coincides with the largest and roughest scales being on the belly of the snake.

## MATERIALS SCIENCES

### POSTERS

#### AN X-RAY ENERGY DISPERSIVE ANALYSIS OF METALS FOUND IN JEWELRY FROM CHINA

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When lead was outlawed in products imported into the United States in 2008, China began to use cadmium metal for manufacturing jewelry. Cadmium is known to cause cancer, kidney and liver failure, kidney and liver disease, brain defects and other organ damage as well according to the EPA [U.S. Environmental Protection Agency]. In 2010 the CPSC (Consumer Product Safety Commission) alerted consumers of the dangers caused by this toxic metal. California, Connecticut, Illinois, and Minnesota banned the sale of children's jewelry with more than 3% cadmium by weight in 2010. Canada called for a voluntary ban of children's jewelry sold with cadmium in it.

In the current study jewelry samples from China were re-tested using the TM 1000 Hitachi Tabletop Microscope and SwiftED-TM Energy Dispersive X-ray spectrometer. It was hypothesized that cadmium metal would be found in jewelry manufactured in China. Sixteen jewelry samples manufactured in China and purchased in the United States (11 samples in December 2011, 3 samples in 2008 and 2 samples in 2005) have been tested for cadmium. Elements that have been found in these samples are tin, nickel, aluminum, copper, iron, bromine, silver, zinc, and technetium. No cadmium was found in any of the jewelry samples tested to date.

#### References

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#### TRACING PIGMENT PATTERNS OF IRON-BASED PAINT ON ANCESTRAL PUEBLOAN POTTERY BY SEM-EDS WITHOUT THE USE OF CARBON COATING

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A previous publication by the authors [1] has described a method of detecting mineral-based paint pigments (containing iron) using Scanning Electron Microscopy and Energy Dispersive Spectroscopy (SEM-EDS) on an Ancestral Puebloan black-on-white painted pottery sherd. An evaporative carbon coating was applied to this pottery sherd to allow the mineral-based paint pigment to be observed with fewer charging effects while using backscatter imaging and SEM-EDS iron elemental mapping techniques. The application of a carbon coating for the reduction of charging effects has also been used by other researchers such as Stewart and Adams [2] for backscatter imaging of mineral-based pigments. A piece of the same pottery that was used in our previous publication [1] was selected for the present study but instead of carbon this pottery piece was coated with ruthenium vapor as described in [3]. The advantage held by ruthenium vapor coating compared to carbon coating was that the ruthenium coating reduced charging effects but did not obscure the pottery surface with a darkened carbon coating. Along with the ruthenium coating, aluminum foil was applied to the surface of the sherd (except for the section to be observed) to reduce the effects of charging during SEM-EDS image and x-ray elemental map production.

A JEOL JSM-6400 SEM (15 KeV, 15 mm working distance) was used to produce secondary and backscatter images and an EDS x-ray map of the sherd painted with iron-based pigment. A SEM-EDS x-ray map of iron was produced using a PGT (Bruker) detector and PGT (Bruker) Spirit software. The x-ray map was produced despite the presence of the ruthenium metal coating which can suppress the detection of SEM-EDS map X-ray signals.

This technique is not meant to make paint pigments which are easily observed more visible using SEM images and SEM-EDS maps, but to serve as a method of detecting pigments which are so abraded or weathered



that the pigment patterns can no longer be easily observed.

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[2] Stewart, J. D. and K. R. Adams, *American Antiquity* 64 (1999) 675.

[3] Ellis, E. A. and M. W. Pendleton, *Microscopy Today* 15 (2007) 44.

## POSTERS

BERNABE RODRIGUEZ and SANDRA L. WESTMORELAND, Department of Biology, Texas Woman's University, Denton, TX 76201

The purpose of this project was to illustrate the basic concepts of an inquiry based science activity to primary grade level students and their teachers who may be limited in their experiences with this teaching method. Participants received a copy of the story entitled "*The Elves and the Shoemaker*" by Horace Scudder. The printed copy had five words highlighted. The highlighted words represented a physical object of which a scanning electron micrograph was provided. Participants were to match the micrographs to the highlighted words in the story. After matching photos to the highlighted words, participants were to discuss and share their reasons for making those decisions. To assess this project's effectiveness in promoting inquiry, observers not participating in the project rated the quality of the discussions shared by the participants.

Annie Means (R) explains to Ann Ellis (L) the details of her poster during the 2011 TSM meeting. Photo by H. Arnott.



Guest speaker Dr. Paul Kotula, Sandia National Laboratories, Albuquerque, NM, presents a talk at the 2011 TSM meeting about the forensics aspects of the anthrax attacks that killed five people in the fall of 2001. Photo by N. Mills.



## TEXAS SOCIETY OF MICROSCOPY PRESIDENTS

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1982	Bruce Mackay	2006	Joanne T. Ellzey
1983	Charles Mims	2007	Ernest Couch
1984	W. Allen Shannon, Jr./Charles Mims	2008	Sandra Westmorelane
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Please type or print legibly. Fill out completely. Though we will mail to your home address, we prefer to mail to your work address. Please note that membership is for the months of January to December of each year.

Check one:    ☐ I am applying for new membership in the Texas Society for Microscopy.  
                 ☐ I am a member and wish to change my mailing address.  
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Date: \_\_\_\_\_ Are you a member of the Microscopy Society of America?   ☐ Yes    ☐ No

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If this is a student membership, what is your degree level? \_\_\_\_\_ What is your major? \_\_\_\_\_

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What broad field of interest do you utilize in microscopy: (circle one)    **Zoology**    **Botany**    **Microbiology**

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Applicants for membership should include a check or money order (no credit cards can be processed by TSM) for one year's dues with application (Regular: \$30.00; Student: \$10.00; Corporate: \$300.00). Application for new membership or for upgrading a membership from Student to Regular category will be presented to the Executive Council at their next meeting for their approval (by majority vote). The applicants will then be presented by the council to the membership at the next general business meeting for their approval (by majority vote). Applicants will be added to the membership rolls at that time.

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Your membership in the Texas Society for Microscopy demonstrates your commitment to the advancement of your career in microscopy. A membership in the parent society of TSM, the Microscopy Society of America (MSA), will also achieve this end but at a national rather than a local level. For information concerning the benefits of MSA membership, go to [www.microscopy.org](http://www.microscopy.org).







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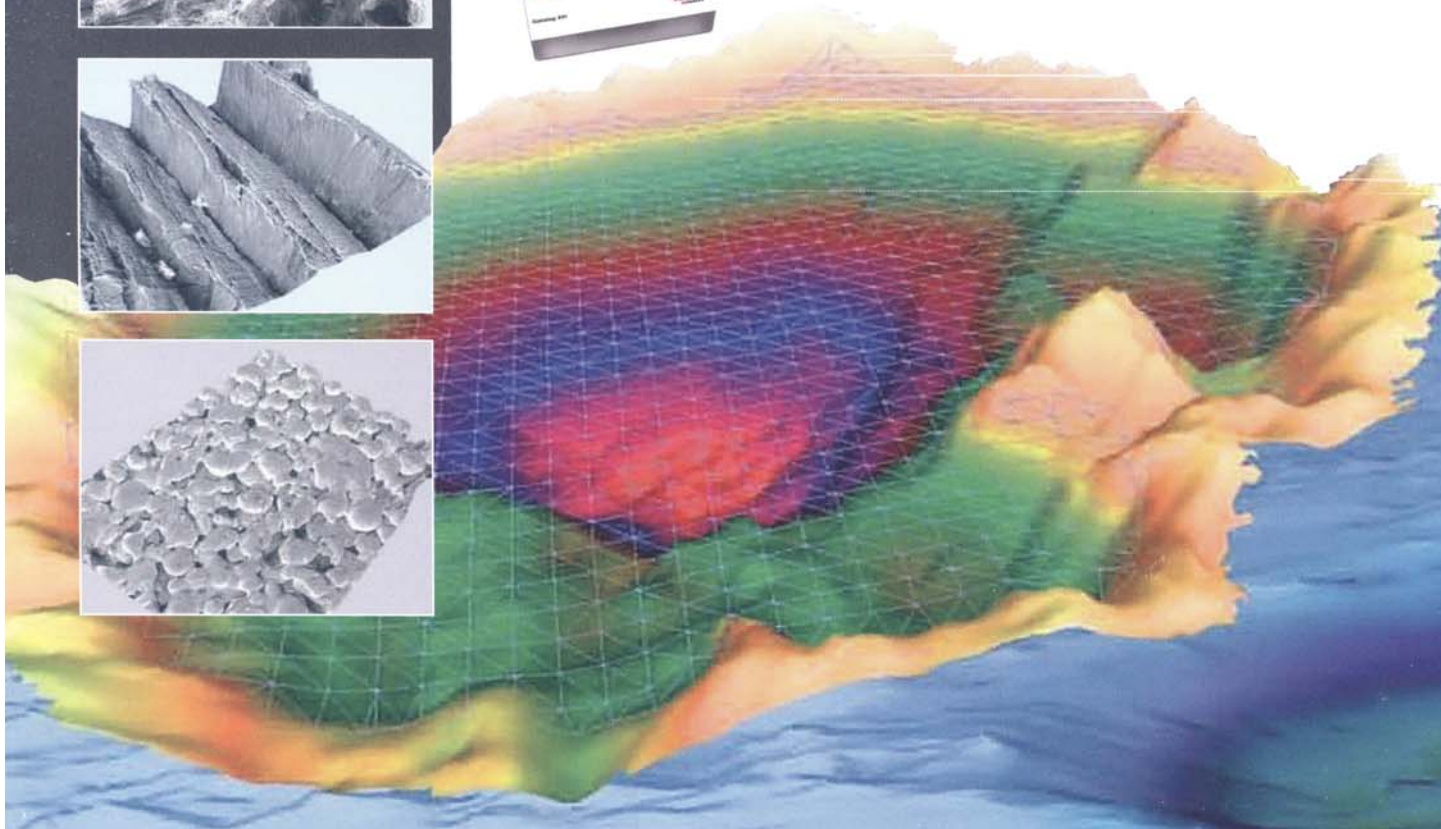
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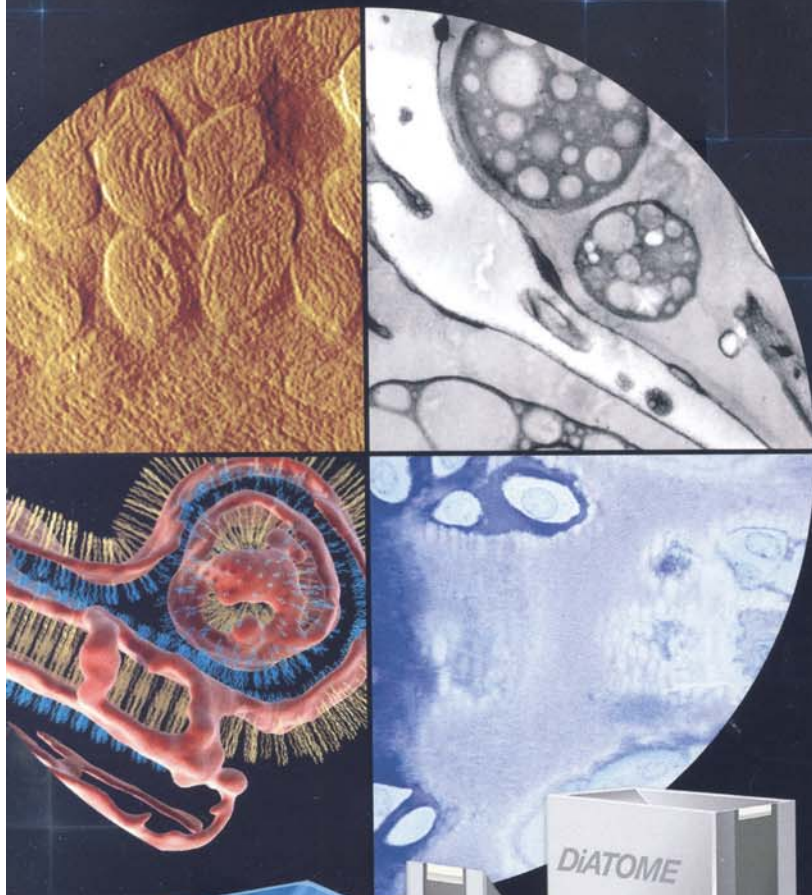
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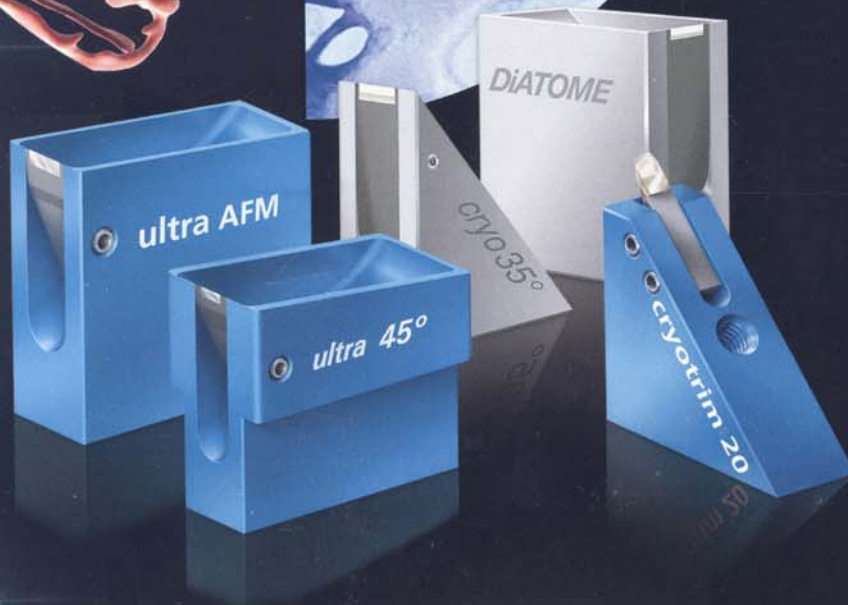
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