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## DINOSAURS OF CIMARRON COUNTY, OKLAHOMA

**Kent Smith[1,2], Richard Cifelli[2], and Nicholas Czaplewski[2]. [1]Oklahoma State University Center for Health Sciences, Department of Anatomy and Cell Biology, Tulsa, OK; [2]Sam Noble Oklahoma Museum of Natural History, Norman, OK**

In 1931, a large fossil was discovered east of the town of Kenton, Oklahoma, by a road worker. Geologists J.W. Stovall and L.I. Price, from the University of Oklahoma, visited the fossil site in 1932 and collected what proved to be a large rib and two caudal vertebrae of an *Apatosaurus*. J.W. Stovall received federal funding from the Works Progress Administration, which supported the "Kenton Project" from 1935 to 1942. During the tenure of this project, Stovall and crew discovered seven significant fossiliferous localities in the Morrison Formation near the town of Kenton. Over the course of the project, numerous remains from both saurischian and ornithischian dinosaurs were discovered, together with fossils belonging to other groups (Osteichthyes, Testudines, Crocodilia).

After a seventy year hiatus in dinosaur collecting at Black Mesa, a new quarry was discovered in April 2012. To date, this new quarry has already yielded several taxa of dinosaurs as well as turtle, fish, and crocodilian remains. Thus, the collecting of dinosaurs and other vertebrates from the Morrison Formation near the town of Kenton, Oklahoma, has been rejuvenated for another generation of scientists and students. These new fossils, together with those previously collected (many of which have never been studied), promise to provide new insights into the vertebrate life of western Oklahoma 150 million years ago.

## COMPARISON OF LEAF AREA INDEX (LAI) OF TOMATO PLANTS GROWN UNDER CONVENTIONAL OR PLASTICULTURE TECHNIQUES

**Alicia M. Fisher[a], Ashton S. Fisher[a], William Phillips[b], Laura Gruntmeir, and William Baker, Redlands Community College, El Reno, OK 73036, Casey Sharber, OSU-Extension Service, El Reno, OK 73036, Micah Anderson, Oklahoma Department of Agriculture, Oklahoma City, OK 73015**

Vegetable producers can use plasticulture techniques to conserve water and control weeds. The objective of this research was to compare tomato plant canopy growth under conventional (CONV) and plasticulture (PC) techniques. Eight rows of tomatoes (4 pairs of rows with treatment randomly assigned within each pair) were established on Canadian soils at the Darlington Applied Agriculture Research Center (Lat. 35.58 N Long. 98.00 W). Rows were 24.1 ± 0.18 m (78.4 ± 0.57 ft) in length. Plants were planted 1.5 m (4.5 ft) apart. More than one variety was used, but all varieties were represented in each row. Canopy size was estimated using a ceptometer (AccuPar model LP-80; Decagon Devices, Pullman, WA) to measure photosynthetically active radiation (PAR) during the first week of July, 2012, when plants were mature. A total of four above and four below canopy PAR readings were made on each plant. To adjust for the lack of canopy symmetry, above and below canopy readings were taken parallel, perpendicular, and diagonally (n =2) in relation to the row. Data was analyzed using the paired T-test procedure. Number of live plants per row, ratio of above and below canopy PAR readings, and estimated LAI were not different ($P > 0.40$) between CONV and PC treatments. In this experiment, a ceptometer was used to estimate

canopy size. Plasticulture techniques did not increase canopy size, but additional research is needed to determine the impact of plasticulture on productivity and cost of production.
[a] Current address Animal Science Department, Oklahoma State University, Stillwater, OK 74074
[b] Corresponding author

## *GET GREEN FOR BLUE* AN ENVIRONMENTALLY BASED SUMMER ACADEMY PROGRAM

**K. McDowell, M. Parrott, and P. Christol. Northeastern State University at Broken Arrow, 3100 East New Orleans Street, Broken Arrow, OK 74014.**

Although we live on a planet consisting of about 71 percent water, most of the earth's small percentage of freshwater is not available to us. This irreplaceable chemical, critical for all life, is one of our most poorly managed resources. This Summer Academy program emphasized the importance of both water quality and conservation and the critical role it plays in the environment as a whole. This one-week program was open to students from 8th through 10th grades for the past three summers. The students collected qualitative and quantitative data on three water bodies at two different locations and also analyzed the biodiversity around the water as a further indicator of environmental health. Dressed in thigh-high waders and wielding D-nets and collection tanks, students waded into the ponds to collect macroinvertebrates for analysis of water quality. Data was collected and analyzed concerning the waters' alkalinity, transparency, nitrate levels, dissolved oxygen levels, level of biodiversity and temperature. Comparisons were made between the three water bodies and their relative health, and the findings given as group presentations. A service learning component was embedded into the experiences as connections were made among environmental science concepts and mathematics. The sense of reciprocity between the community partner, Rogers County Conservation District Education Reserve, and the Summer Academy students constituted a step beyond simply volunteering. Eastern Red Cedars were removed after connections were made to invasive species and their impacts on the environment. *The Get Green for Blue* Summer Academies were developed to provide students with relevant hands-on, inquiry-based activities that integrated environmental science concepts and issues while students honed their teamwork skills, learned about science STEM careers, and gave families and students knowledge concerning future attendance at an institute of higher education. The academies were sponsored by the Oklahoma State Regents for Higher Education.

## NOVEL ROLE OF MCM10 IN LEADING STRAND DNA REPLICATION

**Brandy Fultz, Chance Hendrix, and Sapna Das-Bradoo, Department of Natural Science Northeastern State University 3100 E New Orleans Broken Arrow, OK 74014**

Evolutionarily conserved minichromosome maintenance protein 10 (Mcm10) plays a key role in replication initiation, fork assembly, and elongation. In eukaryotic cells, Mcm10 has been shown to interact with replication fork proteins Mcm2-7, origin recognition complex (ORC), Cdc45, and proliferation cell nuclear antigen (PCNA). In addition, Mcm10 has been shown to have a crucial interaction with polymerase alpha primase complex (Pol α). While Pol α is essential in initiating DNA replication, polymerase delta (Pol α) and polymerase epsilon (Pol α) carry out the bulk of DNA replication. Recent studies indicate that, during DNA replication, Pol α synthesizes lagging strand DNA while pol α synthesizes leading strand DNA. Here, we set out to determine if Mcm10 interacts with Pol α and Pol α in budding yeast, *Saccharomyces cerevisiae*. Both Pol α and Pol α are composed of several subunits in *S. cerevisiae*. Pol α is a heterotrimer that contains the subunits pol 3, pol31, and

pol32. Pol α is a heterotetrameric complex that comprises three small subunits (Dpb2, Dpb3, and Dpb4) and one large catalytic subunit (pol2). Our studies show a direct interaction between Mcm10 and polymerase epsilon. Furthermore, we show that Mcm10 interacts with the catalytic subunit of polymerase epsilon, pol2, by yeast two-hybrid assay. Interestingly, no interaction between polymerase delta and Mcm10 was observed. The expression of the reporter gene, LacZ, was monitored qualitatively by the ability of the yeast to break down 5-bromo-4-chloro-3-indolyl-β-D-galactopyranoside (X-Gal). A quantitative analysis of β-galactosidase activity was measured in liquid culture with ortho-nitrophenyl-β-D-galactosidase (ONPG) as a substrate. Our findings lead us to believe that Mcm10 is part of a protein complex involved with replication of the leading strand DNA.

**MCM10 AND MRC1 INTERACTION IS IMPORTANT FOR MAINTAINING GENOME STABILITY IN *SACCHROMYCES CEREVISIAE***

**Chance Hendrix and Sapna Das-Bradoo, Department of Natural Science, Northeastern State University 3100 E. New Orleans St. Broken Arrow, OK 74014**

Genomic instability is a hallmark of cancer cells, so there is a continuous search for proteins and pathways that are critical for maintenance of genome stability. A previous study has identified proteins that play an important role in maintaining genome stability. Two of these proteins are minichromosomal maintenance protein 10 (Mcm10) and mediator of replication complex 1 (Mrc1). Evolutionarily conserved Mcm10 is an essential part of the replication fork and plays a vital role in fork stability through interactions with proliferating cell nuclear antigen (PCNA), DNA polymerase alpha, and Mcm2-7. Mrc1 is involved in the activation of S phase checkpoint and has also been shown to interact with DNA polymerase epsilon on the replication fork. Our laboratory's goal is to further examine the roles of these two proteins as protectors of genomic stability. Since Mcm10 and Mrc1 are highly conserved through eukaryotes, we chose budding yeast, *Saccharomyces cerevisiae*, as a model organism. We have observed that Mcm10 interacts strongly with Mrc1. Yeast two-hybrid technique was used to measure this interaction both qualitatively and quantitatively. This initial study suggests that Mcm10 may have an integral role in S phase checkpoint because of its interaction with Mrc1 at the replication fork. In order to better understand the role of this interaction, we have mapped the interaction domains on both these proteins. Truncations of Mcm10 and Mrc1 were constructed in yeast two-hybrid vectors. Both the proteins were systematically truncated to preserve their conserved domains. Moreover, we have confirmed the expression of these truncations by Western blot analysis. Our results indicate that Mcm10 interacts through its N-terminus while Mrc1 interacts through its conserved C-terminus. We are now investigating the role of this interaction in yeast cells using fluorescence microscopy.

***VISCOELASTIC PROPERTIES OF GLUTEN PROTEINS FROM HARD RED WINTER WHEAT EVALUATED BY COMPRESSION-RECOVERY AND CORRELATIONS WITH FUNCTIONALITY**

**P. Chompoorat, Food & Agricultural Products Center, Oklahoma State University, Stillwater, OK, 405-744-4401; Hernandez, Z., Centro de Investigación y de Estudios Avanzados del Instituto Politecnico Nacional, Unidad, Querétaro, Quéretaro, Mexico; Oklahoma State University, Stillwater, OK, 405-744-1549; Rayas-Duarte, P., Biochemistry & Molecular Biology, Food & Agricultural Products Center, Oklahoma State University, Stillwater, OK 74078, 405-744-6468**

Compressional stress and elastic recovery were used for evaluating the viscoelastic properties of gluten proteins extracted from 22 hard red winter wheat samples. A rapid

large deformation (8 N of force compression holding for 5 s) was applied to gluten and its recovery response (55 s) was analyzed with a three-element model consisting in a Hookean spring in series with one Kelvin element (one spring and dashpot in parallel) ($r^2 \approx 0.974$; $P<0.0001$). Relationships of physical and chemical properties were analyzed including resistance to mixing and extension, gluten index, ratio of polymeric to monomeric proteins, baking performance, and viscoelastic properties of gluten. Relationships of the parameters were illustrated in principal component analysis (PCA) and Pearson correlation. Total explained variance of this sample set was 52.3% (PC1 = 31.7 and PC2 = 20.6%) and 72.2% when excluding the parameters that have low contribution to the variance. Strength and extensibility were the highest contributors to the variation (first principal component) while dough mixing properties were the next highest contributors to variation (second principal component). Chemical properties of these flours showed low variation. In general, the sample set of gluten proteins had an initial Strain of 0.10, Strain in the viscoelastic response of 0.41, and retardation time (retarded elastic response) of 2.99 s. A rapid deformation of gluten can be used to evaluate strength (strain in the viscoelastic response) and extensibility (retarded elastic response).

**ROLE OF RND TRANSPORT SYSTEMS IN CALCIUM-INDUCED ANTIBIOTIC RESISTANCE IN *PSEUDOMONAS AERUGINOSA***

**Sharmily S Khanam*, Dirk L. Lenaburg, Ryan C. Kubat, and Marianna A. Patrauchan, Oklahoma State University, Stillwater, OK.**

*Pseudomonas aeruginosa* is a facultative pathogen infecting lung airways of patients with cystic fibrosis and causing infective endocarditis and severe device-related infections. It is a highly adaptable organism demonstrating resistance to practically all antimicrobials available for clinical treatments, and therefore represents a great challenge in medicine, as well as clinical and fundamental sciences. Calcium ($Ca^{2+}$) is a well-established signaling molecule that regulates essential processes in eukaryotes including innate immune responses. Earlier we showed that $Ca^{2+}$ triggers biofilm formation and production of virulence factors in *P. aeruginosa.* Here we study the effect of $Ca^{2+}$ on antibiotic resistance in *P. aeruginosa* strain PAO1. We performed proteomic analyses of *P. aeruginosa* membrane and extracellular proteins using 2D PAGE-MS/MS and LC-MS/MS-based spectrum counting. Four multidrug efflux pumps MexAB-OprM, MexGHI-OpmD, TriABC-OpmD and MuxABC-OpmB were affected by $Ca^{2+}$. We used both traditional assays and Etest strips to assess the effect of $Ca^{2+}$ on minimal inhibitory concentrations (MICs) of several structurally different antibiotics including ceftazidime (cephalosporin), ciprofloxacin (quinolone), doripenem (carbapenems), tobramycin (aminoglycoside), and polymixin B (polycationic) and determined that $Ca^{2+}$ increases the MICs of tobramycin and polymixin B at least tenfold. To study the role of resistance-nodulation-cell division (RND) superfamily of efflux pumps in $Ca^{2+}$ - induced antibiotic resistance, we used transposon mutants lacking functional genes encoding 11 RND systems identified in the PAO1 genome. We characterized the mutants for the effect of $Ca^{2+}$ on their growth rate, MICs of tobramycin and polymixin B, and virulence. The results showed that the mutants lacking MexB (PA0426), MuxC(PA2526), MexJ(PA3677), CzcB(PA2521), MexP(PA3522) and MexE(PA2493) are significantly affected by $Ca^{2+}$. This suggests possible involvement of these systems in $Ca^{2+}$-wired adaptation mechanisms, which provide *P. aeruginosa* with the ultimate protection against available treatments.

## MCM10 AND MRC1 INTERACT TO COORDINATE DNA REPLICATION AND THE S-PHASE CHECKPOINT PATHWAYS

**Ian Schalo and Sapna Das-Bradoo, Department of Natural Sciences, Northeastern State University, 3100 East New Orleans Street, Broken Arrow, OK 74014**

Minichromosome maintenance protein 10 (Mcm10) is an essential replication factor necessary for replication initiation and elongation, and plays a central role in replication fork assembly. Due to the essential nature of Mcm10 in coordinating replication, investigation into the role of Mcm10 in the S-phase checkpoint pathway was undertaken. To investigate this function, interaction between Mcm10 and the Mediator of replication checkpoint 1 (Mrc1) was studied. Mrc1 is known to play a key role in activation of the S-phase checkpoint. *Saccharomyces cerevisiae* was used as the model organism to study this interaction. We utilized yeast two-hybrid assay where the desired proteins (Mcm10 and Mrc1) were transformed into yeast and the interaction between the two proteins was determined both qualitatively and quantitatively. For qualitative analysis, the interaction was confirmed by monitoring LacZ gene expression using X-Gal (5-bromo-4-chloro-3-indolyl-β-D-galactopyranoside) as a substrate. Quantitative analysis involved preparing whole cell extracts and then measuring β-galactosidase activity in the extracts using ONPG (*ortho*-Nitrophenyl-β-galactoside) as a substrate. We also employed a non-phosphorylatable mutant of Mrc1, $Mrc1_{AQ}$ to understand the functional significance of this interaction. Our results indicate that Mcm10 and Mrc1 interact strongly. This is a robust interaction as compared to the positive control, which is a previously known interaction between proliferating cell nuclear antigen (PCNA) and polymerase δ (Pol32). Furthermore our results indicate that $Mrc1_{AQ}$ demonstrates a strong interaction with Mcm10 similar to that as observed with the wild type Mrc1. Appropriate negative controls were used throughout the assay. We conclude that Mcm10 and Mrc1 interact by yeast two-hybrid assay and this novel interaction may play a role in either DNA replication or in activating S-phase checkpoint. Further experiments are being carried out to understand the functional significance of this interaction.

## A STUDY OF VIRUS TYPE AND BIOMASS CORRELATION IN *PANICUM VIRGATUM*.

**JulieAnna Rohde and Ulrich Melcher**

In 2005 to 2008 Oklahoma EPSCoR supported a project entitled Plant Virus Biodiversity and Ecology (Wren 2006) coordinated by Ulrich Melcher, and Marilyn Roossinck of the SR Noble Foundation. As part of that project, plants of the Tallgrass Prairie Preserve (TPP) of northeastern Oklahoma were surveyed for the presence of viruses (Melcher 2008; Muthukumar 2008)V., Melcher, U., Pierce, M.L., Wiley, G.B., Roe, B.A., Palmer, M.W., Thapa, V., Ali, A., and Ding, T.

Switchgrass, *Panicum virgatum*, was targeted for frequent sampling. Those studies revealed that some specimens of this species hosted viruses. Plants from which these viruses were identified were not carefully monitored to detect any but the strongest symptoms of disease. This study follows up on this result to ask whether virus presence or absence affects plant traits specifically to test for a correlation between the presence of certain viruses and increased plant mass in *P. virgatum*. Identification of a positive or negative correlation can assist development of new cultivars with increased biomass production potential.

The study sampled multiple plants that exhibited different levels of robustness (plants producing more mass vs. plants of less mass) within the same plot or stand. Samples from Oklahoma State University (OSU), Southeast Kansas Agricultural Research Center (SEKARC) and the TPP were analyzed for the presence of five viruses: Wheat Streak Mosaic Virus, Triticum Mosaic Virus, High Plains Virus, Barley and Cereal Yellow Dwarf Virus, and Asclepius Asymptomatic Virus. The only samples that tested positive for any of the

five viruses were from the SEKARC plots and were positive for Wheat Streak Mosaic Virus (WSMV) only. When the WSMV positive varieties from SEKARC were compared to the WSMV negative samples of the same varieties from OSU, the SEKARC samples did show a lower biomass. Soil samples revealed that none of the locations were deficient in any of the nutrients tested. Both locations were experiencing a drought. This could indicate a possible negative correlation between the presence of WSMV and biomass production. However, more data would be needed to verify this since this experiment was not designed to account for all of the variables between locations.

**NO-COST SOFTWARE FOR STUDENT ANALYSIS OF NUCLEIC ACIDS AND PROTEINS**

**E.L. Blewett, Department of Biochemistry and Microbiology, Oklahoma State University – Center for Health Sciences, Tulsa,** 74107

There are many excellent commercial programs for manipulating and comparing nucleic acids and proteins. However the cost of these programs is prohibitive for students and many faculty members. A number of free, public domain or educationally licensed programs are used in our graduate program. The students are able to install the software on their own computers and retain the programs when the course is complete. This presentation will examine: Staden package for DNA analysis and trace alignment, pDraw for basic analysis and presentation, Clustal for nucleic acid or protein alignment, BLAST for nucleic acid or protein database searches and MEGA for inference of phylogeny. These programs run on Windows XPthrough– Windows 7 OS's and to some extent on Mac OS's. The programs and further information can be obtained at the following locations: Staden (staden.sourceforge.net), pDraw (www.acaclone.com), Clustal (www.clustal.org), BLAST (blast.ncbi.nlm.nih.gov/Blast.cgi) and MEGA (www.megasoftware.net). Uses of these programs in the classroom setting, uses in the laboratory and potential problems will be discussed.

**LIGHT RHYTHM INFLUENCE ON THE GROWTH AND PERITHECIA SYNTHESIS OF *CHAETOMIUM GLOBOSUM*, A COMMON INDOOR MOLD**

**Poudyal, Shubhra, Thapa, Ankita, Biles, C., and Cluck, T., Biology Department, East Central University, Ada, OK 74820.**

*Chaetomium globosum* is a fungus commonly found in water-damaged buildings and was one of the most prevalent fungi associated with damage resulting from the Katrina hurricane. The ascospores and hyphae produced by *C. globosum* can be highly allergenic to immuno-compromised people and has been reported to cause more severe respiratory health problems. Light plays a major role in growth and reproduction in several organisms and is a major determinate in circadian rhythms of mammals. *Chaetomium globosum* 5 mm hyphal plugs were transferred to potato dextrose (PD) agar media plates (90 mm diameter). Isolates of *C. globosum* were exposed to light rhythms; continuous dark, continuous light, 12 h light/12 h dark, 6 h light/18 h dark, and 3 h light/21 h dark. Diameter of growth was measured every 7 days. The number of ascospores and perithecia was measured after 21 days. Results indicated that growth was not significantly influenced by different light rhythms, but ascospore and perithecia synthesis was greater in the dark when compared to light treatments. Ring patterns of fungal perithecia growth was evident on the 12 h light/12 h dark, 6 h light/18 h dark, and 3 h light/21 h dark, suggesting that light/dark cycles stimulate a circadian-like rhythm. Proteins were extracted from *C. globosum* grown on PD broth cultures exposed to the light rhythms previously described. The continuous light treatment stimulated a unique protein that was approximately 25 kD. All treatments that included a dark sequence showed unique bands at 15 kD. Future research will involve isolating and

identifying proteins and genes stimulated or inhibited by light. This research will provide further information on physical factors that elicit ascospore formation and possibly lead to methods to control growth of indoor molds.

**INFLUENCE OF HISTORIC COFFEE CULTIVATION ON TERRESTRIAL SNAIL COMMUNITIES IN THE LUQUILLO EXPERIMENTAL FOREST, PUERTO RICO**

**Craig R. Zimmermann, Renee Morse-Heenan and Nadia Kyrylov, Department of Biology, Rogers State University, Claremore, OK**

Regular censusing of terrestrial snails in the Luquillo Experimental Forest, Puerto Rico, has been ongoing since 1989. These data have routinely shown significant spatiotemporal variation in the distribution of species in this area. The influence of habitat factors, including land use history, underlying this variation has not been fully explored. This study investigated the influence of historic coffee cultivation on extant snail communities in secondary forest. Snail diversity was surveyed in an area near the El Verde Research Station known to support coffee cultivation until 1928. Snails were sampled during the summer for three consecutive years (2009-2011) along three linear transects running perpendicular to known plantation boundaries. Each transect consisted of 10 plots (3 m radius by 3 m height) with 5 plots in the historical plantation and 5 plots in adjacent undisturbed forest. Overall snail abundance was found to be significantly higher in the secondary forest on the abandoned coffee plantation. *Caracolus caracolla* and *Nenia tridens* were the most common snails in both land uses. These species, however, were 2-5x more abundant on plots in old coffee. *C. caracolla* populations also supported a greater proportion of juveniles. *Platysuccinea portoricensis*, a common forest floor species, was also much more abundant on old coffee plots. No difference in species richness or diversity was found between land use classes. Differences in habitat, arising from land-use history, may underlie these differences. Previous surveys of these same areas found distinct differences in tree species composition and soil properties between land uses. Notably, calcium, nitrogen, and pH were elevated in old coffee soils. Limestone, applied to coffee fields to raise pH, is still present is high concentration. Calcium is needed by snails for shell growth, so limestone soil amendments would benefit resident snails. Higher soil pH would accelerate litter decomposition and increase available food for detritivorous snails. Elevated soil nitrogen likely arose from leguminous *Inga vera* trees planted to shade coffee plants. The resulting increase in soil fertility would promote faster forest growth and greater litterfall production. These increased detrital inputs coupled with faster decomposition would act to enhance snail habitat.

**USING PHYTOLITH ASSEMBLAGES TO RECONSTRUCT THE HISTORY OF A DEGRADED DESERT GRASSLAND IN BIG BEND NATIONAL PARK**

**Craig R. Zimmermann[1], Larry Green[1], John C. Zak[2], Bryan M. Noblitt[1] and Richard Hart[1;] [1]Department of Biology, Rogers State University, Claremore, OK; [2]Department of Biological Sciences, Texas Tech University, Lubbock, TX**

Written historical accounts indicate the northern region of Big Bend National Park once supported extensive arid and semi-arid grasslands. Decades of overgrazing together with periodic drought, however, has transformed this landscape into an advanced desertified state. As part of an effort by the national park to restore native grasslands to this region, evidence of the pre-disturbance plant assemblage within the affected area was sought through phytolithic reconstruction of paleovegetation. The study site is located at the foot of the Santiago Mountains in the northeastern corner of the park near Dog Canyon. The plant community at this location is currently characterized by a banded vegetation system commonly associated with arid environments. Here, dense parallel bands of grasses and

shrubs are distributed within a matrix of bare soil and scattered shrubs. Soil cores (1.5 m depth) were taken from three scattered points within areas of bare soil. Core samples were extracted at 15 cm intervals to create a record of historical change with time. Phytoliths were extracted from each sample using a standard ashing method, mounted to microscopes slides, then counted and classified to morphotype. Raw counts within each sample were transformed to proportions and averaged for each depth. Seven different morphotypes were found. These include six morphotypes commonly associated with grass (festucoid, saddle, rondel, pyramidal, panicoid opaque, bulliform) and one morphotype typically associated with shrubs (spherical). Substantial variation in morphotype representation was found with increasing soil depth indicative of a plant assemblage in considerable flux over time. A clear pattern of directional change in community composition is absent. Notably, however, several pairs of morphotypes show a strong negative correlation in abundance with changes in soil depth. That is, an increasing abundance of one type is coupled with decreasing abundance of a second. Moreover, these associations appear to show an oscillating trend over the full soil profile analyzed. This is indicative of periodic or regular changes in plant community composition over time. Such findings could be a product of the extant plant community. Evidence suggests that the bands in some banded vegetation systems do migrate slowly across the landscape. Our evidence of oscillating changes in plant composition might be a historical footprint of such movement in our system. Alternatively, they could reflect a larger-scale pattern of change in these systems. Further research is needed to identify morphotypes with known species and to establish dates for the time scales analyzed.

**LIGHT CYCLES PLAY A ROLE IN REGULATING SPORULATION IN THE FUNGUS CHAETOMIUM GLOBOSUM.**

**Shubhra Poudyal, Ankita Thapa, and Charles Biles**

Poster presentation, Section H, Microbiology; Contact information: Dr. Charles Biles, Biology Department, East Central University, Ada, OK 74820, cbiles@ecok.edu, 580-559-5498