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Oral Presentations (Presenters in bold, ** denotes participation in student competition)

Abstracts listed in alphabetical order by last name of presenter

1. Multi-species coexistence, habitat filtering, and speciation patterns in a radiation of Malagasy social spiders
   Ingi Agnarsson, Diego Agostini, Sahondra Hanitriniaina, and Matjaž Kuntner
2. High-throughput sequencing reveals co-expressed gene suites involved in silk synthesis in cob-web weaving spiders (Theridiidae)
   Nadia A. Ayoub, Jessica E. Garb, Alex K. Lancaster, Cheryl Y. Hayashi, Thomas Clarke
3. The role of trail following and volatile chemicals in *Schizocosa ocreata* sexual communication
   Ryan D. Bell
4. Evolutionary shifts in the material properties of dragline silk during the origin of orb webs
   Todd A. Blackledge
5. Alternative greenspace designs affect spider assemblages within an urban landscape
   ****Caitlin E. Burkman**, Mary M. Gardiner**
6. Comparative analysis of quantitative reproductive traits indicates precopulatory antagonism in the leiobunine harvestmen of eastern North America (Opiliones: Sclerosomatidae)
   ****Mercedes M. Burns**, Marshal Hedin, Jeffrey W. Shultz**
7. Cattle Grazing and Spider Populations: Effects Across Time and Comparisons Between Rotational vs. Conventional Grazing
   Alan B. Cady, Tim Bankroff
8. High Throughput Sequencing Derived Transcriptomes Demonstrate Tissue-Specific Gene Expression Evolution Among Cob-Web Weaving Spiders (Theridiidae)
   Thomas Clarke, Jessica Garb, Cheryl Hayashi, Nadia Ayoub
9. Location, Location, Location: Determination of the Range of Normal Spider Flora and The Effects of Habitat on Araneae Bacterial Populations
   Michelle Lynn Cohen, Norman Horner and Cathy R. Tugmon
10. A multifaceted account of microbial reproductive manipulation in *Mermessus fradeourm* (Linyphiidae)
    ****Meghan Curry**, Jenifer A White**
11. Phylogenetic analysis of the camel spider family Eremobatidae (Arachnida, Solifugae): towards a better understanding of the Spawn of Satan
    Paula E. Cushing, Jack O. Brookhart, Matthew R. Graham, Lorenzo Prendini & Patrick Casto
12. Genetic species delimitation, species tree inference, and morphological species descriptions in the *Sitalcina sura* species group (Opiliones: Laniatores)
    ****Angela DiDomenico**, Marshal Hedin**
13. Microhabitat distribution of *Drapetisca alteranda*, a tree trunk specialist sheetweb weaver (Araneae, Linyphiidae)
    Michael L. Draney, Jennifer A. Hegnet, Ashley L. Johnson, Brooke C. Porter, Clarissa K. Justmann, Patrick S. Forsythe
14. Evolution of *EF-1α*in pseudoscorpions
    Roberta Engel, Elizabeth Jockusch
15. Searching for genetic signals of dune adaptation in the *Aptostichus atomarius* species complex (CA): *de novo* transcriptome assembly and characterization
    ****Nicole Garrison**, Jason E. Bond**
16. Investigating the relationship between immune defense and secondary sexual characteristics in *Schizocosa ocreata* wolf spiders
    ****Rachel Gilbert**, Charity Combs, George W. Uetz**
17. An integrative approach to delimiting species using multiple DNA barcoding methods: a case study of the North American tarantula genus *Aphonopelma*
    ****Chris A. Hamilton**, Brent E. Hendrixson, Michael S. Brewer, Jason E. Bond**
18. The dominance of seismic signaling and selection for signal complexity in *Schizocosa* multimodal courtship displays
    Eileen A. Hebets, Cor J. Vink, Laura Sullivan-Beckers, Malcolm Rosenthal
19. A Precarious Existence: Conservation genetics of the federally endangered spruce-fir moss spider (*Microhexura montivaga* Crosby & Bishop)
    Marshal Hedin, Frederick A Coyle
20. Double vision? Visual subsystems in jumping spiders
    Elizabeth M. Jakob, Skye M. Long
21. A molecular phylogeny of the sub-family Argyrodinae (Theridiidae) – tentative biogeographical patterns of kleptoparasites relying on islands (webs) within islands (forests) on islands
    Heine Kiesbüy, Ingi Agnarsson
22. Spatial binding of multimodal courtship signals in a wolf spider, *Schizocosa ocreata*
    ****Elizabeth C. Kozak**, George W. Uetz**
23. First record of an opilionid from the suborder Eupnoi on New Caledonia: further evidence for Gondwanan relicts?

Dallas Krentzel, James Cokendolpher

24. **On the issue of taxonomic status of *Latrodectus hasseltii* Thorell, 1870 (Araneae: Theridiidae) from India**
****Siddharth Kulkarni**, Cor Vink
25. **Exotic brown widows versus native black widows in urban southern California**
****Marty A. Lewis**, Dessie L.A. Underwood
26. **Life history, web ontology and cribellum use in *Tengella perfuga* Dahl 1901**
Rachael Mallis
27. **Morphological differences in the legs of three South Texas scorpions**
C. Neal McReynolds, Oliver K Long
28. **Phylogeny of Chilean *Porteria*, with comments on the families Desidae and Stiphidiidae**
Elizabeth Morrill, Charles Griswold, Anthea Carmichael
29. **Ontogenetic development of defensive behaviors in the western widow spider (*Latrodectus hesperus*)**
****David R. Nelsen**, William K. Hayes
30. **South Dakota Spider Survey: Preliminary findings of field sampling and literature review**
L. Brian Patrick, Kevin Pfeiffer
31. **Consanguineous cannibals: Does relatedness reduce sexual cannibalism and mating frequency in the wolf spider *Tigrosa helluo*?**
Matthew Persons, Gina Palazzi, Colin Sweet
32. ***The Tom Sawyer effect*: the effect of keystone individuals on collective behavior starts when the keystone individual ceases participation**
Jonathan N. Pruitt, Virginia Settepani, Lena Grinsted, Carl N. Keiser
33. **Multilocus genomic analysis of the harvestmen superfamily Ischyropsalidoidea**
****Casey H. Richart**, Cheryl Hayashi, Marshal Hedin
34. **Test for Temporal Stability of an Aggressive Syndrome in the Desert Spider, *Agelenopsis aperta*: Gregarious Phase Manipulations**
Susan Riechert, Jennifer Bosco, Jonathan Pruitt
35. **The effect of thermal environment and activity on anaerobic respiration in a wolf spider**
J. Andrew Roberts, Michael Wilhelm, Ian Ackers
36. **The effect of food resource heterogeneity on male mating success in the wolf spider *Schizocosa stridulans***
****Malcolm F. Rosenthal**, Eileen A. Hebets
37. **Temporal and spatial positioning for foraging optimization in subsoical *Anelosimus studiosus* (Araneae: Theridiidae)**
****Chelsea R. Ross**, J. Colton Watts, Edith Seier, Michele Joyner, Thomas C. Jones
38. **Info-disruption and the function of courtship in the wolf spider, *Pardosa milvina* (Araneae, Lycosidae)**
Ann L. Rypstra, Alex W. Webb
39. **Mechanical performance of spider orb webs is tuned for high speed prey**
Andrew T. Sensenig, Sean P. Kelly, Kimberly A. Lorentz, Brittany Lesher, and Todd A. Blackledge
40. **Temperature affects an orb-weaving spider's viscous droplet responses to humidity**
****Sarah D. Stellwagen**, Brent D. Opell, Kelly G. Short
41. **Visual imprinting for a secondary sexual characteristic in *Schizocosa ocreata* (Hentz)**
****Brent Stoffer**, George W. Uetz
42. **Insights into the communication of the "purring" wolf spider, *Gladicosa gulosa***
****Alexander L. Sweger**, George W. Uetz
43. **Color use by tiny predators: jumping spiders exhibit biases and color learning during foraging**
Lisa A. Taylor, Emily B. Maier, Zarreen Amin, Kevin J. Byrne, and Nathan I. Morehouse
44. **Consistencies and Differences in Araneae/Bacterial associations detected from 2006-2012**
Cathy R. Tugmon , Norman Horner
45. **Multimodal signals increase active space of communication by wolf spiders in a complex litter environment**
George W. Uetz, J. Andrew Roberts, David L. Clark, Jeremy S. Gibson, Shira D. Gordon, A. Kluckman1
46. **Arachnophobic entomologists: when two more legs makes a big difference**
Richard S. Vetter
47. **Chronoethology of *Cyclosa turbinata* (Araneae: Araneidae)**
****J. Colton Watts**, Ashley Herrig, William D. Allen, Darrell J. Moore, Thomas C. Jones
48. **Effects of season and prey consumption on *Schizocosa* cold tolerance**
****Thomas D. Whitney**, Benjamin N. Philip, & James D. Harwood
49. **Female receptivity of *Schizocosa ocreata* wolf spiders (Araneae: Lycosidae) towards potential mates of varying quality in sequential male encounters**
****Maggie Williams**, Brent Stoffer, George W. Uetz
50. **Limb-Loss, Regeneration, and Orange Bitey Things (*Pterinochilus murinus*): The impact of autotomy on metabolism**
****Brenan Wilson**, Cara Shillington
51. **The role of different eye types in mate recognition and prey detection by female *Schizocosa ocreata* (Hentz) wolf spiders (Araneae: Lycosidae)**
****Rebecca Wilson**, Tess Piening and George W. Uetz
52. **The phylogenetic scale of venom variation in Haplogyne spiders**
Pamela A. Zobel-Thropp, Emily Thomas, Jennifer Mullins, Miles Dale, Andrew Wood, **Greta J. Binford**

Multi-species coexistence, habitat filtering, and speciation patterns in a radiation of Malagasy social spiders

Ingi Agnarsson1, Diego Agostini2, Sahondra Hanitriniaina3, and Matjaž Kuntner4

1University of Vermont
2University of Puerto Rico
3University of Antananarivo
4Slovenian Academy of Arts and Sciences

Classical ecological and evolutionary theories predict that through allopatric speciation and competition among similar species, multiple closely related and ecologically similar species rarely coexist. When such species coexist, they are expected to evolve differences through processes like habitat filtering and morphological differentiation. *Anelosimus* species are found worldwide and range in behavior from solitary to social, where behavior dictates ecological factors such as web type and size. Multiple, socially comparable, *Anelosimus* species rarely overlap in distribution. In Madagascar, however, we have shown previously that a forest fragment may contain several similar and apparently cohabiting subsocial species. To explore their diversity, and patterns of speciation and coexistence, we sampled 350 *Anelosimus* colonies along a fine-scale transect (2 km) spanning open and closed forest and during different seasons, identified colonies using DNA barcoding, and analyzed phylogenetic patterns at local to global scales. These data reveal a major monophyletic radiation of subsocial *Anelosimus* species within Madagascar in the Pliocene (<6 mya). Speciation probably primarily occurred allopatrically, with subsequent sympatry resulting in the cohabitation of multiple, close relatives. Some recently diverged monophyletic single-forest lineages suggest possible cases of sympatric speciation. The species can be segregated based on their habitat use, size, web size, and phenology, a pattern that might be referred to as habitat filtering. In sum, we reveal a remarkable pattern of diversification and coexistence of *Anelosimus* species in Madagascar. Why *Anelosimus* species elsewhere do not show similar patterns and why no fully social species occur on Madagascar are open questions.

High-throughput sequencing reveals co-expressed gene suites involved in silk synthesis in cob-web weaving spiders (Theridiidae)

Nadia A. Ayoub1, Jessica E. Garb2, Alex K. Lancaster3, Cheryl Y. Hayashi4, Thomas Clarke1

1Department of Biology, Washington and Lee University, Lexington, VA
2Department of Biological Sciences, University of Massachusetts, Lowell, MA
3Whitehead Institute, Boston, MA
4Department of Biology, University of California, Riverside, CA

Most spiders have multiple silk gland types with each type synthesizing a high-performance, task specific fiber or glue. For example, dragline silk, which originates in the major ampullate glands, has a tensile strength comparable to steel, and prey wrapping silk, which originates in the aciniform glands, is incredibly tough. Each fiber type is composed of one or more unique structural proteins called spidroins, which belong to a single gene family. Thus, the spider silk system represents a spectacular example of functional diversification via gene duplication followed by sequence and expression divergence. Presumably, gland-specific silk synthesis must require non-spidroin genes, but these have yet to be characterized. Using high-throughput sequencing of mRNAs isolated from seven functionally differentiated silk gland types we *de novo* assembled >50,000 transcripts, identified their putative functions, and profiled expression patterns in three cob-web weaving spiders (Theridiidae): Western black widows, *Latrodectus hesperus*, brown widows, *L. geometricus,* and false black widows *Steatoda grossa*. As expected, we found a unique spidroin paralog was the most highly expressed gene in each gland type for all three species with the exception of tubuliform glands in *L. geometricus* and the aggregate glands. However, each spidroin paralog is also expressed at varying levels in most silk glands. Furthermore, expression levels of each spidroin paralog are tightly correlated ($R > 0.99$) with expression levels of 40-100 other genes. These suites of co-expressed genes are strong candidates for direct involvement in differentiating functionally distinct silk glands.

The role of trail following and volatile chemicals in *Schizocosa ocreata* sexual communication

Ryan D. Bell

The Ohio State University

For male spiders in complex habitats, the ability to chemically detect a female isolated with regard to other signaling modalities has strong potential implications on survival and fitness. After detecting a female, a male can engage in courtship behaviors, reducing the likelihood of being cannibalized, and increasing her receptivity to mating. This is the case with the forest dwelling wolf spider *Schizocosa ocreata*, where the introduction of a male to a substrate laden with female cues is sufficient to induce courtship displays. Two unresolved questions arise; can males follow a chemical trail to a hidden female, and is this trail volatile or substrate bound? The first objective of this study was to assess the ability of focal male spiders to follow the trails of females, subadults or other males using a Y-maze where a stimulus individual was induced to deposit a trail down one randomly selected path. Males were able to follow female trails, but showed no preference on subadult or male cues. In addition to trails associated with passing females, males in the wild may also encounter volatile cues emanating from females upwind or within the leaf litter. Therefore our second objective was to assess the ability of males to detect and respond to any volatile cues present in 1) an airstream passed over a female and 2) within an enclosed arena above a sequestered female. Male response in both experiments indicated that volatile cues likely do not play a large role in *S. ocreata* sexual communication.

Evolutionary shifts in the material properties of dragline silk during the origin of orb webs

Todd A. Blackledge

Department of Biology and Integrated Bioscience Program, The University of Akron

In contrast to ancestral sheet webs, orb webs capture fast flying insects, placing a premium on how orb webs dissipate the kinetic energy of flight. Thus, the origin of aerial orb webs placed unique selective pressures on the evolution of dragline silks.Using a phylogenetic approach, I show that several features of orb spider major ampullate (MA) silk – including its extraordinary toughness and highly non-linear stress-strain behavior – are unique to Orbiculariae and therefore plausibly adaptive. In contrast, at least one key component – high damping capacity – evolved long before the orb web.

Alternative greenspace designs affect spider assemblages within an urban landscape

****Caitlin E. Burkman**, Mary M. Gardiner

Ohio State University, Department of Entomology

Many Midwestern cities in the United States have experienced high levels of economic decline and home foreclosure, which has led to thousands of acres of vacant land. The remaining community has conceptualized a variety of reutilization strategies for this land, including urban agriculture and the creation of small parks of native plantings. In this two-year study, we used spiders as a model system for examining the impacts of vacant land conversion on arthropod generalist predators within residential vacant lots, community gardens, and planted prairies within the peri-urban park system. At the time of the study, planted prairies on previously vacant land were not common, so those within the park system may be informative for future plantings. We found that maintained vacant lots contained the most active spider assemblage, but was dominated by disturbance-tolerant habitat generalist species. Community gardens contained the fewest spiders with a somewhat more diverse assemblage than vacant lots. Similarly low numbers were also found in planted prairies, but the assemblage was comparatively most diverse. We also compared these results at the family and genus level in order to assess the taxonomic sufficiency relevant for future studies, and found that genus level

Comparative analysis of quantitative reproductive traits indicates precopulatory antagonism in the leiobunine harvestmen of eastern North America (Opiliones: Sclerosomatidae)

****Mercedes M. Burns1**, Marshal Hedin2, Jeffrey W. Shultz1

1 University of Maryland, College Park, MD, USA
2 San Diego State University, San Diego, CA, USA

Sexual selection mechanisms, such as female choice and sexual conflict, may produce reproductive syndromes, suites of reproductive traits that coevolve in males and females. Detecting relationships between traits and transitions in syndromes requires the reconstruction of the direction, frequency and relative timing of phenotypic evolution of male and female structures in species-rich clades. For example, our previous work using discrete characters showed an association between the loss of

nuptial gift-bearing penile sacs and gain of female pregenital barriers, a pattern that appears to reflect multiple shifts from female choice to intersexual antagonism. Here we examine the evolution of quantitative reproductive traits in 29 eastern North American harvestmen species using features hypothesized to covary with the intensity of precopulatory antagonism. Size residuals of male and female relative genital muscle forces were found to have significant positive correlations in the tip states. A Bayesian application of the threshold model used to assess correlation through time also yielded positive male and female reproductive trait correlations. These results indicate that precopulatory antagonism has increased during the evolution of the leiobunine harvestmen of eastern North America and that quantitative muscular traits may provide a better indicator of precopulatory antagonism than categorical reproductive traits.

Cattle Grazing and Spider Populations: Effects Across Time and Comparisons Between Rotational vs. Conventional Grazing

Alan B. Cady, Tim Bankroff

Department of Zoology, Miami University, Oxford OH

Understanding arthropod community structures and the dynamics associated with different grazers and grazing schedules may provide a metric allowing identification of sustainable grazing schemes. Sampling the arthropod & spider communities inhabiting pastures under a rotational and conventional grazing provided an opportunity to start characterizing the ecological conditions which could signify 'healthy' and sustainable pastures. Arthropods in pastures at Polyface Farms (Swope, VA) under high-intensity, low-frequency rotational cattle (and chicken) grazing, and three immediately adjacent continuously grazed pastures were collected from 0.25-m2 areas employing an intense two-stage suction-sampling technique of the vegetation and substrate. Although arthropod abundances decreased after initial rotational grazing events, their numbers, diversity, and evenness were still greater than those found in conventionally grazed pastures. The overall proportional representations of major taxa were maintained pre-and post-grazing under rotational grazing, indicating better resiliency to tis disturbance. Arthropod and spider abundances continued to increase after rotational grazing, especially where deferment was longest, with spider numbers doubling between 42-64 days post-grazing. In contrast, spider numbers decreased during this same time period in conventionally grazed pastures. Increased pasture size in the early season decreased the strength of the correlation between deferment period and spider abundance that was found later in the season. The Linyphiidae, Lycosidae, and Tetragnathidae showed prominent and steady increases as grazing deferment progressed, while the Thomisidae, Araneidae, and Salticidae did not. With further sampling and analyses to identify 'signatures', pasture health may be assessed in the future by determining spider community structure, allowing remediation plans to be formed.

High Throughput Sequencing Derived Transcriptomes Demonstrate Tissue-Specific Gene Expression Evolution Among Cob-Web Weaving Spiders (Theridiidae)

Thomas Clarke1, Jessica Garb2, Cheryl Hayashi3, Nadia Ayoub1

1Department of Biology, Washington and Lee University, Lexington, VA USA
2Department of Biological Sciences, University of Massachusetts, Lowell, MA USA
3Department of Biology, University of California, Riverside, CA USA

Spiders possess a diverse molecular toolkit for synthesizing high-performance materials, such as silks and venoms. However, this toolkit is poorly characterized due to the lack of genomic resources for spiders. High throughput sequencing (HTS) technologies facilitate genomic characterization of non-model organisms such as spiders. We generated transcriptomes (all expressed genes) from hundreds of millions of sequenced mRNA fragments expressed in multiple tissues of three cob-web weaving species, *Latrodectus hesperus, L. geometricus*, and *Steatoda grossa* (Theridiidae). For each tissue type, including seven functionally distinct silk gland types, venom glands, ovaries, and cephalothoraxes, we *de novo* assembled transcripts. The assemblies from the individual glands were combined to construct species-specific transcriptomes. Our transcriptomes resulted in 40-50 thousand unique transcripts per species that could be functionally annotated by homology to published protein-coding genes. We also identified 30-50 thousand transcripts with significant expression levels in at least one tissue, as assessed by number of raw sequence reads that align the transcript. Of the significantly expressed genes, ~20 thousand have known homology. Using this set of high-quality transcripts, we document several comparative genomic applications, including gene family gains and losses across the three spider species and the functional annotation of both tissue-specific and widely expressed genes. We finally demonstrate that of the top 25 most highly expressed genes in a particular tissue in one species between 40 and 90% possess homologous genes that are also highly expressed in the same tissue of another species. Our results suggest varying rates of gene expression evolution among tissue types.

Location, Location, Location: Determination of the Range of Normal Spider Flora and The Effects of Habitat on Araneae Bacterial Populations.

Michelle Lynn Cohen1, Norman Horner2 and Cathy R. Tugmon3

1 Medical College of Georgia, Georgia Regents University, Augusta, GA
2 Director of Natural Laboratories, Midwestern State University, Wichita Falls, TX
3 Department of Biology, Georgia Regents University, Augusta, GA

Exploration of spider bacterial flora to date has been limited. The purpose of this research is to build a database that will assist in determining a "normal" spider bacterial flora and assess the effects of habitat on Araneae bacterial populations including their associated antibiotic resistance. For the 2012 part of this data base 251spiders from 20 families were captured aseptically from 14 sources in 13 wards of isolation across eight counties. Approximately 705 bacteria were isolated using selective media streaking and crystal violet staining was performed to determine cell morphology. Subsequently, MicroScan® autoSCAN-4 system was implemented to identify 268 bacteria as well as test each bacterium for antibiotic susceptibility. The results of the data tabulation and analysis will be discussed.

A multifaceted account of microbial reproductive manipulation in *Mermessus fradeourm* (Linyphiidae)

****Meghan Curry**, Jenifer A White

University of Kentucky, Department of Entomology

Spiders, like most arthropods, are host to an ample variety of heritable endosymbiotic bacteria. Endosymbionts ensure their own transmission by conveying a facultative benefit to the host or by manipulating reproduction to eliminate males which serve as evolutionary dead-ends for maternally inherited bacteria. Common modes of reproductive manipulation include parthenogenesis, male killing, feminization, and cytoplasmic incompatibility. Recent broad-taxa screening studies indicate that endosymbionts are particularly common among spiders; however, little is known about how these bacteria affect their spider hosts. To investigate this potentially unique symbiont-spider relationship *Mermessus fradeourm* (Linyphiidae) were collected in the field, reared in a laboratory, and screened for common endosymbionts. Two distinct infections were detected: a double infection of both *Rickettsia* and *Wolbachia* and a single *Wolbachia* infection. Mating assays revealed that *Rickettsia-Wolbachia* infected mothers produced extremely female-biased offspring. Antibiotic treatment of *Rickettsia-Wolbachia* infected females successfully eliminated both endosymbionts and restored the sex ratio to the expected 1:1 in subsequent generations. Conversely, mothers infected with only *Wolbachia* produced offspring with an even sex ratio and antibiotic elimination of *Wolbachia* revealed no differences in fecundity, size, or survivorship between infected and unifected offspring. However, in a two-way factorial mating assay of cured and *Wolbachia* infected males and females cytoplasmic incompatibility was detected such that mating between cured females and infected males produced 70% fewer offspring than all other pairings. A simple probe into the nature of endosymbiont infection in *M. fradeourm* has revealed rich and complex layers of reproductive manipulation likely driving host evolution and ecology

Phylogenetic analysis of the camel spider family Eremobatidae (Arachnida, Solifugae): towards a better understanding of the Spawn of Satan

Paula E. Cushing1, Jack O. Brookhart1, Matthew R. Graham2, Lorenzo Prendini3 & Patrick Casto1

1Denver Museum of Nature & Science
2Eastern Connecticut State University
3American Museum of Natural History

In this study, we present the first phylogenetic analysis of any family in the arachnid order Solifugae. Currently the taxonomy of the family Eremobatidae is based entirely on shared morphological similarities between species. The family, which includes 187 described species, is divided into the subfamilies Therobatinae and Eremobatinae. The former includes the genera *Chanbria, Eremochelis,* and *Hemerotrecha*. The Eremobatinae includes the genera *Eremobates, Eremocosta, Eremorhax, Eremothera,* and *Horribates*. The genera *Eremobates, Eremochelis,* and *Hemerotrecha* are further divided into various species groups. For the molecular and morphological phylogenetic analyses, we used 82 exemplar taxa representing all genera and all but four species groups. We used a multilocus approach for the molecular phylogeny based upon two mitochondrial gene fragments (cytochrome oxidase subunit 1 and the 3' end of the 16S rRNA gene) and two nuclear gene fragments (the large subunit 28S rDNA gene and a variable fragment of the histone H3 protein coding gene). We assessed phylogenetic patterns using Bayesian inference (BI) and Maximum Likelihood (ML). We found weak support for the subfamily classification. Several of the genera were strongly monophyletic: *Chanbria, Eremocosta, Eremorhax,* and *Eremothera*. The genera *Hemerotrecha, Eremochelis,* and *Eremobates* were polyphyletic. Three of the species groups were monophyletic: *Eremobates scaber* group, *Eremboates pallipes* group, and the *Hemerotrecha banksi* group. This phylogenetic analysis provides the first objective assessment of the taxonomy of any family of Solifugae.

Genetic species delimitation, species tree inference, and morphological species descriptions in the *Sitalcina sura* species group (Opiliones: Laniatores)

****Angela DiDomenico**, Marshal Hedin

San Diego State University

California is home to numerous endemic harvestmen (Arachnida, Opiliones), including members of the genus *Sitalcina*. The *Sitalcina sura* group is ideal for testing species limits because these dispersal-limited arachnids show minimal gene flow between populations, which typically results in many genetically distinct endemic populations. Prior to this study, very limited genetic data was available for *Sitalcina* species and previous studies focused only on broad relationships of taxa within the genus, not closely related species. Here, molecular phylogenetic and species delimitation analyses will be conducted including all species in the *S. sura* group. In addition to the eight currently described species, we have discovered four possible new species. For each species in the *S. sura* group, genetic species delimitation should provide the framework needed for determining species limits as well as the operational taxonomic units. Phylogenetic analyses were used to examine evolutionary relationships and biogeographic structure. Species delimitation was accomplished using the program BP&P, which uses reverse-jump MCMC (rjMCMC). Species trees were reconstructed using multispecies coalescent methods implemented in *BEAST. Preliminary genetic analyses using DNA sequence data from 5 genes delimit 13 species in the *S. sura* group. Molecular phylogenetic evidence reveals distinctive genetic groupings between species residing in desert versus those in costal habitats. The study and classification of these rare arachnids is an important project to undertake for evolutionary, biogeographic, and conservation purposes. Conservation may be an important issue for several species within the *S. sura* group given their restricted endemic distributions, and defining operational taxonomic units will help aid conservation efforts.

Microhabitat distribution of *Drapetisca alteranda*, a tree trunk specialist sheetweb weaver (Araneae, Linyphiidae)

Michael L. Draney, Jennifer A. Hegnet, Ashley L. Johnson, Brooke C. Porter, Clarissa K. Justmann, Patrick S. Forsythe

Department of Natural and Applied Sciences, University of Wisconsin – Green Bay, Green Bay, WI

ABSTRACT: In an undergraduate course on ecological methods and analysis, teams of students conducted structured sampling to examine the microhabitat distribution of *Drapetisca alteranda*; adults are found almost exclusively on tree trunks. Sampling was conducted in a 1 ha plot in which all trees over 10 cm dbh had previously been identified, mapped, and measured. The lower 2 m of tree trunks were sampled for *Drapetisca* by brushing spiders into beating sheets. We sampled equal trunk surface areas of the largest of the four most abundant tree species on the plot: *Quercus alba, Fraxinus pennsylvanica*, *Tilia americana*, and *Carya ovata*. We measured average tree bark furrowing depth at 15 locations around each tree. We analyzed the data with a General Linear Mixed Model assuming Poisson distribution. Tree species and furrowing depth, but not tree size, were significant predictors of total number of *Drapetisca* collected. 84% of spiders were collected on *T. americana,* and the relationship between spider abundance and furrowing depth was negative. To test for distribution above ground surface, we divided the lower portion of selected tree trunks into six 0.5 m sections, sampling each separately. Regardless of tree species, tree trunk height was a significant predictor of female (but not male) *D. alteranda* occurrence, with 52% of the females found 0.5- 1.0 m above the forest floor. We also measured the carapace width (an index of body size) for all 30 sampled males and an equal number of females. Interestingly, male carapace width is significantly greater in this species.

Evolution of *EF-1α* in pseudoscorpions

Roberta Engel1, Elizabeth Jockusch2

University of Notre Dame, University of Connecticut

Gene duplication events are important evolutionary events; gene copies are a source of variation for selection to act on. While many duplicates are lost to purifying selection, some may be the source of functional innovations or evolutionary novelties. Elongation factor 1- subunit alpha (*EF-1a*) has a central role in protein synthesis across all domains of life. Early gene characterization studies revealed multiple functional copies of *EF-1a* in *Drosophila* and *Artemia*. Gene copies are thought to be differentially expressed depending on life stage, sex, and possibly tissue type. More recent work suggests that many other arthropods harbor multiple copies of *EF-1a*, including hexapods (e.g.,bees), crustaceans (e.g., copepods), and chelicerates (e.g., spiders). Fragments of *EF-1a* were amplified from pseudoscorpions, an understudied group of arachnids. In this study, we targeted the genus *Synsphyronus* (Pseudoscorpiones). We present evidence that suggests *Synsphyronus* harbors four functional loci. The *EF-1a* fragments sort to three distinct exon/intron patterns: intronless, one intron, and two introns. *EF-1a* fragments were characterized by exon-intron pattern and homology across arthropods. Intron position is shared across multiple arachnids but differs from that inferred in insects. A phylogenetic framework was used to examine the evolutionary history of *EF-1α* in pseudoscorpions and more broadly across chelicerates. Exemplars of the remaining eleven extant orders of chelicerates were retrieved from GenBank. Three duplication events were inferred from phylogenetic analyses. The deepest duplication may have occurred along the stem lineage of pseudoscorpions.

Searching for genetic signals of dune adaptation in the *Aptostichus atomarius* species complex (CA): *de novo* transcriptome assembly and characterization

Nicole Garrison, Jason E. Bond

Auburn University

Recently, NGS technologies and advances in bioinformatic tools have come together to allow *de-novo* assembly of expressed genes in non-model organisms. This research was designed to explore genetic variation within the *Aptostichus atomarius* species complex in central California; a group of trapdoor spiders which displays habitat specific pigmentation patterns. Species endemic to coastal dunes (*A. stephencolberti* and *A. miwok*) are lightly colored relative to their inland sister species (*A. angelinajolieae* and *A. stanfordianus*). To help understand the evolution of this variation, mRNA from each species was extracted, sequenced on the Illumina platform, assembled using the Trinity pipeline, and analyzed via custom scripts and freely available programs. Assembly yielded transcriptomes with an average of 18213 contigs. After filtering, mean transcriptome size was 8130 sequences. The filtering process indicated the presence of potential symbionts, generated four nearly complete mitochondrial genomes, and confirmed a lack of human contamination. A BLAST against the nr database revealed that 70% of the pooled sequences were novel genes. About 750 genes containing ORFs were shared by all four species. This new transcriptomic resource has provided a catalog of genes for primer design and a list of candidate genes that may be associated with the observed pigment variation.

Investigating the relationship between immune defense and secondary sexual characteristics in *Schizocosa ocreata* wolf spiders

Rachel Gilbert, Charity Combs, George W. Uetz

Department of Biological Sciences, University of Cincinnati, Cincinnati, OH

Ground-active wolf spiders must combat constant exposure to soil-dwelling parasites and pathogens, as well as the potential for exposure from food and water sources. As a consequence, these spiders have developed an effective innate immune system. The encapsulation response has been shown to be a good measure of immune function in spiders and other invertebrates, since the melanization which forms around a parasite or pathogen can be quantified. In this study, we evaluate the viability of this method in examining the impact of bacterial infection on immune responses of the wolf spider *Schizocosa ocreata* (Hentz). The multimodal courtship of *S. ocreata* has been shown previously to be energetically costly, and we also examined whether active courtship affects immune function. We found that males who had been previously exposed to the bacterial pathogen *Pseudomonas aeruginosa* in the laboratory as juveniles had significantly higher adult immune function (melanization density) than those not previously exposed. In addition, the size of adult foreleg tufts (a condition-dependent secondary sexual character) was significantly positively correlated with adult immune function in males not previously exposed. Males that had courted for 5 minutes showed lower adult immune function (melanization density) than other males (either exposed as juveniles or unexposed). These results support assumptions of sexual selection theory regarding trade-offs between courtship and immune function, i.e., (a) previous infection increases adult immune function; (b) secondary sex characters and energetically costly courtship behaviors enforce signal honesty by being good indicators of immune function.

An integrative approach to delimiting species using multiple DNA barcoding methods: a case study of the North American tarantula genus *Aphonopelma*

Chris A. Hamilton1, Brent E. Hendrixson2, Michael S. Brewer3, Jason E. Bond1

1 Auburn University Museum of Natural History & Department of Biological Sciences, Auburn, AL
2 Department of Biology, Millsaps College, Jackson, MS
3 Division of Environmental Science, Policy, and Management, University of California, Berkeley, CA

Species in the tarantula genus *Aphonopelma* are the most charismatic arthropod residents of the American southwest, yet the group has been woefully understudied. Only four major descriptive or revisionary works conducted over the past 75 years have evaluated the species diversity within *Aphonopelma*, none of which have employed a phylogenetic approach. Representing the highest nominal diversity of tarantulas in the world (90 described species), traditional morphological characters pose significant problems for species delimitation. Consequently, a history of overzealous taxonomy is hypothesized to have led to an over-description of species. The increased use of molecular information in systematic studies has been instrumental in uncovering tremendous diversity previously unrecognized using traditional morphological approaches. Our research focus was to evaluate the efficacy of popular molecular-based 'species-delimitation' methods (i.e. DNA barcoding) on the identification of known and unknown species, and test the sensitivity of random taxon sampling on the reproducibility of species boundaries between closely related species. By conducting the most comprehensive sampling of a single theraphosid genus to date (682 specimens), we developed an effective methodology that qualitatively and quantitatively evaluates species boundary limits, while incorporating previous knowledge of morphology, ecology, and biogeography. Based on these findings, our hypothesis of a massive over-splitting of species was confirmed wherein 54 of the described US species will be synonymized to 18. Alternatively, these data indicate that 16 species have been discovered that are new to science.

The dominance of seismic signaling and selection for signal complexity in *Schizocosa* multimodal courtship displays

Eileen A. Hebets1, Cor J. Vink2, Laura Sullivan-Beckers1, Malcolm Rosenthal1

1 School of Biological Sciences, University of Nebraska, Lincoln, NE, USA
2 Canterbury Museum, Christchurch, New Zealand

Schizocosa wolf spiders show tremendous diversity in courtship complexity, with different species employing varying numbers of components within and across sensory modalities. Using a comparative approach, we investigate the importance of each signaling modality in the courtship display of five Schizocosa species (3 stridulating/ 2 drumming) by assessing mating success under manipulated signaling environments. Irrespective of the degree of male ornamentation, the three stridulating species exhibit a dependence on the seismic, but not visual, signaling environment for mating success. Mating was independent of signaling environment for the two drumming species. We next ask whether the degree to which each species depends upon a signaling modality for mating is correlated with the estimated modality-specific signal complexity. We first calculate effect sizes for the influence of seismic versus visual signaling environments on the likelihood to mate for ten Schizocosa species and then use an element-counting approach to calculate seismic and visual signal complexity scores. We use a phylogenetic regression analysis to test two predictions: (i) the importance of seismic signaling is correlated with seismic signal complexity and (ii) the importance of visual signaling is correlated with visual signal complexity. We find a significant relationship between visual signal importance and visual signal complexity, but no relationship between seismic signal importance and seismic signal complexity. Finally, we test the hypothesis that selection acts on complexity per se by determining whether seismic and visual signal complexity is correlated across species. We find support for this hypothesis in a significant relationship between seismic and visual signal complexity.

A Precarious Existence: Conservation genetics of the federally endangered spruce-fir moss spider (*Microhexura montivaga* Crosby & Bishop)

Marshal Hedin1, Frederick A Coyle2

1Department of Biology, San Diego State University, San Diego, CA
2PO Box 1935, Cullowhee, North Carolina

In 1995 the spruce-fir moss spider (*Microhexura montivaga* Crosby & Bishop) was listed as federally endangered. This diminutive mygalomorph is endemic to high-

elevation spruce-fir forests of the southern Appalachian Mountains. Survey work by Coyle has shown that *Microhexura* is distributed on six isolated mountain massifs (Whitetop Mountain, VA; Grandfather, Black and Plott Balsam Mountains, NC; Roan Mountain and Great Smoky Mountains, TN and NC). Four of the largest massifs (e.g., Smoky Mountains, Blacks, etc.) include multiple outcrop populations within a larger metapopulation. Genetic analyses of this species - documenting how genetic diversity is partitioned within and among metapopulations - have never been conducted. Here we report on initial results based on DNA sequence data gathered for five newly-developed nuclear genes and a mitochondrial gene, from 47 samples representing all six massifs. Preliminary analyses of these data suggest the following: 1) Mitochondrial data resolves populations within larger metapopulations, thus indicating that female-based gene flow is limited in *Microhexura*. 2) Most nuclear genes do not resolve individual populations, but are consistent with mitochondrial data in resolving 5-6 distinct genetic units within *Microhexura*. A single unit is found west of the French Broad River basin (Great Smoky plus Plott Balsam Mountains), with 4-5 units found east of this basin (Whitetop, Roan, Grandfather, Blacks). The large Black Mountain metapopulation is genetically heterogeneous, and includes novel genetic lineages despite small geographic distances. Ultimately, these data will be fundamentally important in decisions regarding population reintroduction, augmentation and captive propagation efforts, particularly as precarious mountaintop populations face warming climates.

Double vision? Visual subsystems in jumping spiders

Elizabeth M. Jakob1, Skye M. Long2

1Department of Psychology, University of Massachusetts Amherst
2Graduate Program in Organismic and Evolutionary Biology, University of Massachusetts Amherst

Jumping spiders view objects in front of them with two different types of eyes. The principal eyes have fixed corneal lenses and tiny retinas at the end of internal moveable tubes. These retinas offer remarkably high spatial acuity. The fixed retinas of the anterior lateral eyes have a wider angle of view and lower, but still reasonably good, spatial acuity. To understand how these eyes divide up the acquisition of visual information, we use a combination of eye masking and a novel associative learning protocol. The anterior lateral eyes perceive looming objects; both eyes together are needed to identify moving prey; and experiments using stationary stimuli are now in progress. I will also briefly describe current work with our eyetracker that monitors the movement of the principal eyes.

A molecular phylogeny of the sub-family Argyrodinae (Theridiidae) – tentative biogeographical patterns of kleptoparasites relying on islands (webs) within islands (forests) on islands

Heine Kiesbüy1, Ingi Agnarsson1

University of Vermont, Burlington, Vermont

In resolving biogeographical patterns, knowledge regarding the dispersal capability and habitat requirements of the species of interest, coupled with information of geographical barriers on a geological time scale, is the key to understanding the presence and possible speciation of the aforementioned species. The kleptoparasitic spiders of the genera *Argyrodes*, *Faiditus* and *Neospintharus* (Theridiidae:Argyrodinae) adds an extra dimension to the work of resolving such patterns as they not only rely on appropriate habitat variables, but also on the presence of suitable host species. This particular sub-family is not a common target of research in general and in particular from a phylogenetic point of view it needs more attention, as there currently is less than five species represented in the sequence databases. Collecting of kleptoparasites performed as a part of Dr. Agnarssons Caribbean Island Biogeography project combined with contributions gathered by collaborators across the globe, has been utilized in the construction of a phylogeny based on the CO1 and ITS2 genes. The phylogeny covers the Caribbean region extensively, but also North and Central America, Africa, the Indian Ocean, Asia and Australia are represented. The molecular phylogeny, combined with morphological traits (pedipalps and epigynes), will be used to establish and identify species, and will be used for tentative predictions regarding any biogeographical patterns that might be present in the molecular phylogeny.

Spatial binding of multimodal courtship signals in a wolf spider, *Schizocosa ocreata*

****Elizabeth C. Kozak**, George W. Uetz

University of Cincinnati, Dept. of Biological Sciences

Cognitive integration of information transmitted in more than one sensory mode (cross-modal integration) is essential for accurate perception of complex signals, and may have fitness consequences, i.e., females must be able to determine direction of male signals, and discriminate between multiple sources. In some vertebrate species, perceptual coupling of multimodal signals by receivers depends on precise spatial and/or temporal synchrony of modes. We used video/vibratory playback to examine the effect of spatial separation of isolated signal modes (visual and vibratory) from multimodal courtship of male *Schizocosa ocreata* wolf spiders on female detection and response. Spiders respond to spatially distant signals in different modes (90°-180° separation) as if they were from different individuals, but appear to treat more spatially congruent signals (0°- 45° separation) as if arising from a single individual. Results suggest spiders have the ability to perceptually bind signals from spatially close sources as multimodal, as well as discriminate individual modes from more disparate locations.

First record of an opilionid from the suborder Eupnoi on New Caledonia: further evidence for Gondwanan relicts?

Dallas Krentzel1, James Cokendolpher2

1Committee on Evolutionary Biology, University of Chicago, 900 E. 57th Street, Chicago, IL 2Invertebrate Zoology, Museum of Texas Tech University, 3301 W. 4th St. Lubbock, TX

Opiliones has increasingly been considered as a useful clade for testing biogeographical hypotheses, especially given their ancient diversification and poor dispersal abilities. The mite harvestmen (Cyphophthalmi) in particular are known for their markedly low vagility over deep time, and recent analyses support a model of vicariance to explain their intercontinental distributions. The cyphophthalmid family endemic to New Caledonia, Troglosironidae, was recently argued to be a Gondwanan relict, despite geological evidence for a full oceanic submergence of New Caledonia during the Paleocene and rising evidence that many New Caledonian taxa represent recent transoceanic dispersals. Here, we report the first records of a eupnoid from New Caledonia. This new taxon is small (11-14 mm adult length), with opalescent markings, extremely long legs, long chelicerae, and exhibits a large spur at the base of the pedipalpal femur. The new taxon was included in a morphological phylogenetic analysis based on a previously published character matrix for neopilionids, the predominate Eupnoi clade in the southern hemisphere. The analysis yielded nine most parsimonious trees (MPTs), five including the new taxon as sister to *Templar* and *Monoscutum* (two New Zealand enantiobunines), and four MPTs including the new taxon as sister to *Neopilio*, a South African genus that appears as sister to the remaining neopilionids. The congruence of this phylogeny with vicariance and dispersal models is discussed, particularly in light of known opilionid dispersal events. We find this evidence to be suggestive of Gondwanan relictualism; however, future molecular work will be required to corroborate this hypothesis.

On the issue of taxonomic status of *Latrodectus hasseltii* Thorell, 1870 (Araneae: Theridiidae) from India.

****Siddharth Kulkarni1**, Cor Vink2

1 Zoology Department, Yashavantrao Chavan Institute of Science
2 Canterbury Museum, Rolleston Avenue, Christchurch 8013, New Zealand

The taxonomic history of *Latrodectus hasseltii* reported from India is reviewed including the sub-species *L.h. indicus* Simon, 1897. Source of all reports being Pocock's Fauna of British India (1900), itself is adequate for species confirmation. Description of other species *L.erythromelas* Schmidt & Klaas, 1991 is observed to match with the description of Simon's *L.h indicus* morphologically and with the studied specimens. The questions: Is *L. erythromelas* a junior synonym of *L.hasselti indicus* ?; Can *L. indicus* be a valid species? are discussed.

Exotic brown widows versus native black widows in urban southern California

****Marty A. Lewis**, Dessie L.A. Underwood

California State University Long Beach, Biological Sciences

The introduced brown widow spider *Latrodectus geometricus* Koch 1841 was found in southern California in 2003, and anecdotal evidence suggested that native western black widow *Latrodectus hesperus* populations were declining due to *L. geometricus*. However, there were no quantitative studies that tested potential mechanisms. I related species abundances to temperature, humidity, human population size, number of human homes and elevation. I also conducted laboratory experiments on competition for dwellings, prey consumption and capture web properties.
L. geometricus and *L. hesperus* abundances were highest in coastal and desert parks, respectively. Unlike *L. hesperus*, *L. geometricus* abundances did not decrease from summer to winter, suggesting that *L. geometricus* populations are more reproductively active than *L. hesperus* during colder months. Furthermore, *L. geometricus* egg sacs were more abundant than *L. hesperus* at coastal parks, especially during the winter months. Multiple regression analyses showed that *L. geometricus* abundances and human population size were positively related, and *L. hesperus* abundances were negatively correlated with humidity. Lab results showed prey consumption and web structure did not differ between species, but *L. geometricus* juveniles were highly successful at displacing *L. hesperus* from dwelling spaces under warm conditions.
The decade-long replacement of *L. hesperus* by *L. geometricus* from southern California coastal cities may be explained by *L. geometricus'* usurpation of native widows and their ability to perennially reproduce, which would supply urban habitat dwellings with primogenial juveniles that may compete with increasing *L. hesperus* populations during warmer months.

Life history, web ontology and cribellum use in *Tengella perfuga* Dahl 1901

Rachael Mallis

University of New Mexico

Using spiderlings hatched from egg sacs laid by field-caught female *Tengella perfuga,* I describe the web ontology of each life history stage, including the use of cribellate silk. There are between 10-12 instars for *T. perfuga*, depending on the sex of the spider, but typically 10 for males and 11 for females. Surprisingly, cribellate silk use was not observed until the 7th/8th instar, and full morphological development of the cribellum and calamistrum was not apparent until later instars. I also measured the size of each instar, using the standards of carapace length and width at widest point, as well as the length and width of femur I and tibia I. In adult spiders, leg I is dimorphic, with males having longer legs than females and these are used in courtship behavior. Finally, I calculated relative growth rates from instar to instar based upon these measurements. All of this information is working towards a better understanding of the recently rediscovered *T. perfuga,* and being applied to ongoing research into the phylogenetic status and relationships of *Tengella*, Tengellidaeand cribellate silk use evolution in spiders.

Morphological differences in the legs of three South Texas scorpions

C. Neal McReynolds, Oliver K Long

Texas A&M International University, Laredo, Texas

Centruroides, *Vaejovis*, and *Pseudouroctonus* are three genera of scorpions found in South Texas. Each genus occupies a different niche although all are arthropod predators in the same guild. Because of distinct preferences for microhabitats and foraging behavior we expected to find differences in the legs of each genus that might explain locomotion in its preferred habitat. The *Vaejovis* and the *Pseudouroctonus* had a ridge of spikes on the anterior part of the tarsus, and basitarsus while *Centruroides* had distinct pairs of setae in those areas. The ratio of the appendages to each other was also distinct in Centruroides when compared to the other two scorpions. These differences in morphology may explain the microhabitat preference and behavior of these three scorpion genera.

Phylogeny of Chilean *Porteria*, with comments on the families Desidae and Stiphidiidae

Elizabeth Morrill1,2, Charles Griswold2, Anthea Carmichael2

1San Francisco State University
2California Academy of Sciences

Porteria is a charismatic, monospecific genus of sheet-web building spiders and is known only from Chile. The genus was first described by E. Simon in 1904 to include the species *Porteria albopunctata* and was placed within Agelenidae. In 1967, *Porteria* was moved to the family Desidae. Recent morphological investigations have uncovered synapomorphic peculiarities of the male genitalia and spinneret features among these Chilean spiders and a group of Stiphidiid spiders from New Zealand and Australia, and have also led to the discovery of a handful of new *Porteria* species. Our research focuses on constructing a molecular phylogeny of *Porteria* using five loci (18S, 28S, H3, Actin, and CO1) and a broad range of Stiphidiid and Desid outgroup taxa. Our preliminary results resolve a northern clade within *Porteria* as well as a possible Chiloé clade. More work is still to be done on resolving outgroup relationships as well as combining the molecular analyses with morphological data.

Ontogenetic development of defensive behaviors in the western widow spider (*Latrodectus hesperus*)

****David R. Nelsen**, William K. Hayes

Department of Earth and Biological Sciences, Loma Linda University, Loma Linda, CA

The threats an animal must deal with changes during ontogeny, especially in relation to changes in body size, experience, defensive repertoire, and vulnerability to predators. Spiders offer a unique opportunity to test the ontogenetic development of antipredatory strategies, as they display a suite of defensive behaviors including the

use of both silk and venom, which are metabolically costly and, as limited commodities, should be used judiciously. We repeatedly subjected western widow spiders (*Latrodectus hesperus*) to low and high threat conditions over successive instar stages (sheds) to experimentally test three hypotheses. Spiders were poked by a taxidermy mouse specimen in the low-threat condition, and pinched by the same predator model against the wall of its home container in the high-threat condition. Consistent with our first hypothesis, spiders relied largely on non-combative behaviors early in life and switched to more combative behaviors, including silk flicking and biting, as they increased in size. Consistent with our second hypothesis, age exerted a much greater influence than sex differences for males and females within the range of equivalent body sizes. Consistent with our third hypothesis, spiders habituated to the repeated testing by exhibiting fewer combative behaviors than naïve spiders upon reaching adult size. Collectively, these findings suggest that selection has favored age-specific antipredator strategies that can be modified by experience.

South Dakota Spider Survey: Preliminary findings of field sampling and literature review.

L. Brian Patrick1, Kevin Pfeiffer2

1Department of Biological Sciences, Dakota Wesleyan University, Mitchell, SD

< The South Dakota Spider Survey (SDSS) was established in 2010 to inventory the spiders of the state, and catalogue the abundance, diversity, and distribution of spider species within the state. Previous to this current survey effort, only three publications directly examined spiders (Araneae) within the state, and a few additional publications included records from the state. Since the survey was established, we have increased sampling efforts within targeted areas of the state, most notably within the Fort Pierre National Grassland and areas of the Black Hills. To date, more than more than 250 species are now documented from South Dakota from either direct field sampling or from previously published literature, with more species added as sampling efforts expand. An online list is maintained, and digital imaging of the dorsal and ventral general habitus, relevant views of genetalia, and key identification characteristics (e.g., eye arrangement, spinnerets) has started. The goal of the imaging is to provide complete data sheets for each species that will be linked to Encyclopedia of Life and other online resources.

Consanguineous cannibals: Does relatedness reduce sexual cannibalism and mating frequency in the wolf spider *Tigrosa helluo*?

Matthew Persons, Gina Palazzi, Colin Sweet

Department of Biology, Susquehanna University

Kin recognition is common among animals since it may increase direct fitness through inbreeding avoidance or indirectly by helping non-descendent relatives survive or reproduce. Sexual cannibalism may be an adaptive foraging strategy for lycosids since eating an unacceptable mate provides a low cost energy source to the female; however, a potential evolutionary conflict exists when females consume a brother since this precludes future mating opportunities for that male and therefore may lower the inclusive fitness of the female. We examined the resolution of this conflict in the wolf spider, *Tigrosa helluo*. We tested whether adult *Tigrosa helluo* recognize siblings during courtship and mating and whether males show reduced courtship while females show less aggression and sexual receptivity toward siblings than non-relatives. We paired adult virgin males and females with either a sibling or non-sibling reared apart and measured male courtship intensity, female aggression levels, mating frequency and cannibalism frequency among pairs of siblings or unrelated spiders. Brothers showed significantly reduced courtship and mating compared to unrelated males and females showed a strong tendency to avoid attacking brothers compared to non-relatives. *Tigrosa helluo* has either an innate phenotype-matching system for kin discrimination or sexually imprints within four days of emerging from the eggsac. Male and female kin recognition mitigates inbreeding and enhances female inclusive fitness through sexual cannibalism avoidance of brothers.

***The Tom Sawyer effect*: the effect of keystone individuals on collective behavior starts when the keystone individual ceases participation**

Jonathan N. Pruitt1, Virginia Settepani2, Lena Grinsted2, Carl N. Keiser2

1Department of Biological Sciences, University of Pittsburgh, Pittsburgh, PA
2Department of Bioscience, Aarhus University, Denmark

Behavioral ecologists have long observed that, within social groups, some individuals have a disproportionately large impact on group dynamics relative to others. In this pair of studies, we first tested what environmental and demographic factors predicted inter-colony variation in collective foraging behavior in social spiders of genus *Stegodyphus*. Although numerous demographic factors were associated with colony-level foraging behavior, the single best predictor of inter-colony variation was the behavioral tendency of the single boldest individual. To test whether the behavior of these extreme individuals could actually cause differences in colonies' collective foraging behavior, we generated experimental colonies composed of an identical mixture of individuals bearing different behavioral tendencies and only varied the behavior of the single most extreme individual. Consistent with field observations, we found that colonies that harbored just one extremely bold individual exhibited more aggressive collective foraging behavior. However, these differences only emerged days after colony initiation and after these extreme individuals ceased involvement in collective behavior. We term this phenomenon here *The Tom Sawyer Effect*, where the effects of keystone individuals on their social groups are most palpable after they have ceased overt involvement in group dynamics.

Multilocus genomic analysis of the harvestmen superfamily Ischyropsalidoidea

****Casey H. Richart1**, Cheryl Hayashi2, Marshal Hedin1

1San Diego State University
2University of California, Riverside

Next-generation sequencing (NGS) technologies have revolutionized many aspects of evolutionary biology, with a particularly rapid impact on phylogenetics. For example, in only a few years modern molecular phylogenetic studies have gone from using a few candidate loci, to using hundreds of loci scattered throughout the genome. Here we report on a phylogenomic NGS pipeline that shows promise for elucidating ancient divergences within the harvestmen superfamily Ischyropsalidoidea. Using NGS technologies, comparative transcriptomics on multiple Dyspnoi harvestmen allowed for the generation, assembly and alignment of many homologous sequences. Loci annotated as "single copy, single exon" in *Ixodes* were targeted for PCR primer development. These primers were applied to a broader sample of ischyropsalidoid taxa in order to test current familial hypotheses. The higher level relationships within Ischyropsalidoidea have recently received taxonomic treatment, but these studies have resulted in low support for hypothesized families, or put forth hypotheses without testing them in a phylogenetic framework. We find that much of the controversy is associated with rapid branching events near the root node of Ischyropsalidoidea. In an attempt to elucidate this rapid radiation we use coalescent-based species tree analyses using hundreds of loci from exemplars representing each of three rapidly diverged branches, allowing for explicit tests of familial hypotheses.

Test for Temporal Stability of an Aggressive Syndrome in the Desert Spider, *Agelenopsis aperta*: Gregarious Phase Manipulations

Susan Riechert1, Jennifer Bosco1, Jonathan Pruitt1,2

1University of Tennessee
2University of Pittsburgh

Many animal taxa experience a gregarious phase early in the life cycle, though they are solitary and even territorial as adults. We partitioned newly emerged siblings of the desert spider, *Agelenopsis aperta*, into rearing conditions that differed with respect to group size, individual density and prey level to better understand how social (number of individuals) and competitive environment (spiderling density and prey availability) affect individual survival and rates of growth. In particular, we were interested in the extent to which behavioural aggressiveness influences early spiderling success under different environments. We found significant variation existed among families with respect to the probability of suffering mortality, rate of growth and behavioural aggressiveness. Nevertheless, both higher density and greater numbers of individuals present had significant effects on mortality, the probability of consuming sibs that have suffered mortality, and growth rates. Reducing prey levels further increased the magnitude of these effects. Contingency analyses completed on the first PC axis of individual rates of growth in behavioral aggressiveness space, demonstrated that the behavioural phenotype of individual sibs became increasingly important as reduction in prey availability was added to a double density treatment (competitive level manipulation). This pattern was even more pronounced in a social environment manipulation as numbers of individuals present doubled in successive treatments.

The effect of thermal environment and activity on anaerobic respiration in a wolf spider.

J. Andrew Roberts1, Michael Wilhelm2, Ian Ackers2

1,2Department of Evolution, Ecology, and Organismal Biology; 1The Ohio State University at Newark, Newark OH; 2The Ohio State University, Columbus OH

Behavioral performance (i.e. vigor), has often been tied to likelihood of reproduction across a wide array of taxa. Performance is obviously tied to the underlying physiology of the performer and may be temperature sensitive, especially in small ectotherms (like spiders) where behavior is largely dependent on surrounding temperature. Despite the great number of species, wide distribution, and ecological importance, the amount of data available on spider physiology is limited. This is especially true of otherwise well-studied behavioral model systems, such as the brush-legged wolf spider, *Schizocosa ocreata*. In insects, temperature has been demonstrated to have a significant, negative effect on basal metabolic rate, that is, rate declines as ambient temperature increases. This has been suggested as cold adaptation common to small ectotherms. Our goal was to explore the influence of thermal environment on anaerobic respiratory capacity (tissue lactate concentration) in *S. ocreata*. Results establish lactate concentrations at rest and following maximal activity, but suggest that there is no significant differential effect of temperature on maximal lactate concentration in this species. Thermal environment may, instead, influence the rate of accumulation by limiting behavioral vigor and suggest that there may be an optimal thermal environment for behavioral activity.

The effect of food resource heterogeneity on male mating success in the wolf spider *Schizocosa stridulans*

****Malcolm F. Rosenthal**, Eileen A. Hebets

University of Nebraska-Lincoln

Variability in food resource acquisition over a lifetime may differentially affect a variety of a mature male's traits. The effects of food resource variability on the components of a complex courtship display may provide perceiving females with information about a male's foraging history. Males of the wolf spider species *Schizocosa stridulans* attract mates via multimodal courtship consisting of a seismically transmitted “song”, and a visual display with both movement (leg tapping) and structural (ornamented foreleg) components. The leg tapping also produces one of the acoustic components of the seismic display. Previous research in this species has shown that the seismic component is both necessary and sufficient to elicit copulation, that courtship rate (i.e., leg taps/minute) predicts copulation likelihood, and that leg ornamentation affects copulation success only through its interaction with courtship rate. In a closely related species, *S. floridana,* juvenile and adult diet have been shown to interact with courtship rate to affect female mate-choice decision making. Given this, and the evidence for interactions between multiple display components in *S. stridulans*, we examined the effect of variable food abundance on male *S. stridulans* mating success by independently manipulating juvenile and adult food availability and subsequently assessing male phenotype expression and mating success. Results will be discussed.

Temporal and spatial positioning for foraging optimization in subsoical *Anelosimus studiosus* (Araneae: Theridiidae)

****Chelsea R. Ross a,b**, J. Colton Wattsa, Edith Seierb, Michele Joynerb, Thomas C. Jonesa

a Department of Biological Sciences, East Tennessee State University
b Department of Mathematics and Statistics, East Tennessee State University

In subsocial colonies of *Anelosimus studiosus,* females and their maturing offspring share a web and cooperate in web maintenance and prey capture. As both predators and prey, spiders must balance efficient foraging and effective avoidance of predators. We hypothesized that the positions of the spiders in the web would change over the course of the day reflecting the known fluctuations in prey/threat densities. We also hypothesized that the spatial distributions would change over the course of spiderling development as they became capable foragers. Colonies were observed in webs built on a controlled substrate over the daily cycle as well as over the course of their development. New statistical tools were developed to analyze the data. Patterns in positioning including distance to nearest neighbor and distance to the nearest edge were shown to be affected by time of day, age of spiderlings, and whether or not the matriarch was present. When the matriarch was present, the degree of clustering around her was shown to change over the course of spiderling development. These findings support our hypotheses that the spatial distribution of the spiders changes with respect to time of day and developmental stage. Daily changes in spatial distributions are similar to patterns in prey/threat densities and patterns of aggression found in previous studies. This suggests that individuals benefit from changing their behavior in accordance with fluctuations in local prey/threat densities. Future studies will focus on determining the optimal spatial distribution with a stochastic differential equation model by accurately describing prey capture interactions, and comparing this with the known spatial distributions.

Info-disruption and the function of courtship in the wolf spider, *Pardosa milvina* (Araneae, Lycosidae)

Ann L. Rypstra, Alex W. Webb

Department of Biology, Miami University, OH

Conspicuous courtship displays help females identify males, which is particularly important in cannibalistic species where females may attack males as prey. Males of the wolf spider, *Pardosa milvina* (Araneae, Lycosidae), use chemotactile cues (silk, feces, excreta) to identify the mating status and condition of prospective mates. However, glyphosate-based herbicides interfere with accurate detection of those female signals. We hypothesized that including herbicide along with female cues would obscure information about the females and affect the frequency and intensity of courtship. We used old mated females that are not typically receptive to males. We varied the risk posed by females by placing them on different feeding regimes with the expectation that hungry females would attempt to attack and cannibalize males. We observed male courtship in the presence of female cues with and without herbicide present and then released the females and observed how the interaction between the male and female played out. Males initiated courtship more quickly and courted more intensely when female cues were coupled with herbicide. Once females were released, the effects of herbicide disappeared but, surprisingly, males courted food-limited females more vigorously than well-fed individuals. Thus, males engaged in courtship when female information was masked by herbicide and when they were with hungry female. We suspect males are attempting to reduce the risk of cannibalism by courting food-limited females. These findings reveal an understudied function of courtship and also reveal an unexpected cost of life in human-dominated environments where anthropogenic chemicals interfere with information flow.

Mechanical performance of spider orb webs is tuned for high speed prey

Andrew T. Sensenig1, Sean P. Kelly2, Kimberly A. Lorentz2, Brittany Lesher2, and Todd A. Blackledge2

1Department of Biology, Tabor College, Hillsboro, KS

2Department of Biology and Integrated Bioscience Program, University of Akron, Akron, OH

Small prey are easily stopped by webs but provide little energetic gain. While larger prey offer substantial nourishment, they are also challenging to capture and can damage the web if they escape. We therefore hypothesized that spider orb webs exhibit properties that improve their probability of stopping larger insects while minimizing damage when the mechanical energy of prey exceeds the web's capacity. We used high speed video to visualize the impact of wood pellets fired into orb webs to simulate prey strikes and tested how capture probability varied as a function of pellet size and speed. However, successful captures do not directly measure the maximum possible energy dissipation by orb webs because these events include lower energy impacts that may not significantly challenge orb web performance. Therefore, we also compared the total kinetic energy removed from projectiles that escaped orb webs by breaking through the silk, asking whether more energy is removed at faster speeds. Over a range of speeds relevant to insect flight, the amount of energy absorbed by orb webs increases with the speed of prey (i.e. the rates at which webs are stretched). Orb webs therefore respond to faster, and hence higher kinetic energy, prey with better performance, suggesting adaptation to capture larger and hence faster flying insect prey. This speed dependent toughness of a complex structure suggests the utility of either the intrinsic toughness of spider silk and/or features of the macro-design of webs for high velocity industrial or military applications, such as ballistic energy absorption.

Temperature affects an orb-weaving spider's viscous droplet responses to humidity

****Sarah D. Stellwagen**, Brent D. Opell, Kelly G. Short

Virginia Tech

Sticky viscous prey capture threads retain insects that strike araneoid orb-webs. The thread's two axial fibers support a series of glue droplets, each featuring a core of adhesive viscoelastic glycoprotein covered by an aqueous solution. After sticking, the glue extends, transferring the adhesive force and, in the process, dissipates some of the force generated by a struggling prey. As a day progresses, these threads experience a drop in humidity and an increase in temperature. Hence, these environmental variables have the potential to alter thread and web function. This study tests the hypothesis that viscous threads have evolved to function effectively under these fluctuating conditions. We test it by examining threads spun by *Argiope aurantia*, a species that occupies weedy habitats where humidity drops and temperature increases during the afternoon and predict that threads compensate for these opposing changes in a manner that stabilizes their performance over the course of a day. We equated a droplet's energy dissipation capability as the product of tension on the droplet filament, gauged by axial line deflection, and the duration of its extension under a load. This product was greatest under afternoon (hot and dry) conditions, less under morning (cool and humid), and least under hot humid afternoon conditions, suggesting that both high temperature and high humidity decreases glycoprotein viscosity. These results support the hypothesis and show that *A. aurantia* threads are adapted to perform optimally during the afternoon when these spiders capture large orthopterans, which are the most important prey source for this species.

Visual imprinting for a secondary sexual characteristic in *Schizocosa ocreata* (Hentz)

****Brent Stoffer**, George W. Uetz

University of Cincinnati

In many animals, including spiders, female mate choice is often based on the presence or size of male secondary sexual characteristics. However, recent studies suggest that experience (juvenile exposure) and the social environment (e.g., male density, sex ratio, and male quality) also impact mate choice decisions. This may be especially true in species that experience an asynchrony of maturation, providing a period of time in which sexual imprinting may occur. The brush-legged wolf spider, *Schizocosa ocreata*, is an excellent model organism to study the effects of sexual imprinting because females may encounter mature courting males in the field prior to sexual maturity. We tested whether or not females demonstrated plasticity in their preference for male leg tuft size, a secondary sexual characteristic indicative of male quality. Penultimate females were exposed to videos of courting males with reduced tufts, average tufts, enlarged tufts, a mixture of male qualities, or no males at all. As adults, each female was presented a choice between a courting male with reduced tufts and a courting male with enlarged tufts. Results provide evidence for visual imprinting, such that females previously exposed to courtship from small-tufted or large-tufted males demonstrated a stronger preference (i.e., greater number of receptivity displays) for small-tufted or large-tufted males, respectively. Further, females exposed to a mixture of male qualities also demonstrated a stronger preference for large-tufted males. This study demonstrates the effects of an individual's social environment and the role of sexual imprinting.

Insights into the communication of the "purring" wolf spider, *Gladicosa gulosa*

****Alexander L. Sweger**, George W. Uetz

Department of Biological Sciences, University of Cincinnati

Communication research over the past century has suggested that both vibrations and airborne sound play a large role in conspecific communication in animals. Among insects and spiders, vibratory communication is prevalent, even in species where airborne signals are used. It has thus been suggested that vibratory communication is the ancestral state, and may lead to the evolution of airborne signals. Early observations of the "purring" wolf spider *Gladicosa gulosa* (Araneae: Lycosidae) describe an audible, airborne sound produced by males that can be heard up to several meters away. Although coupled with a simultaneous vibratory signal used in courtship, the production of this signal is unusual in that the spider itself does not possess any known structures for detecting airborne sound. This poses several questions regarding this behavior and the pressures that may have driven its initial and continuing evolution within the communication network of this species. We conducted isolated vibratory and acoustic recordings of individual male spiders, and playback of male spider signals to conspecific males and females. Our preliminary results show that the airborne signal in this species is significantly affected by the vibration of the substrate through which the vibratory signal is sent. Additionally, we compared the morphology of the sound-producing organs of this species with several other genera within the family Lycosidae. These data may shed some light on the mechanisms underlying both the physical production of the sound and its potential role in the sexual behavior of this species.

Color use by tiny predators: jumping spiders exhibit biases and color learning during foraging

Lisa A. Taylor, Emily B. Maier, Zarreen Amin, Kevin J. Byrne, and Nathan I. Morehouse

University of Pittsburgh, PA

Understanding the design of prey defenses, such as aposematic colors, involves understanding the perceptual and cognitive abilities of the predators that drive their evolution. Research in this area has largely focused on 'key' predators, such as birds, with less attention to smaller invertebrate predators. To understand the selection

pressures that jumping spiders exert on the color patterns of their prey, we first examined natural biases by offering field-collected *Habronattus pyrrithrix* arrays of artificially colored crickets. We found population-wide color biases with the lowest attack rates on red and yellow prey (colors commonly used as warning colors) and highest attack rates on blue prey. We then went on to examine whether we could 'train' naïve spiders to develop color biases by manipulating their exposure to naturally-occurring combinations of prey. Spiders were randomly assigned to one of three diet treatments: (1) white-eyed *Drosophila* and unpalatable red milkweed bugs (reared on milkweed seeds), (2) white-eyed *Drosophila* and palatable red milkweed bugs (reared on sunflower seeds),or (3) white-eyed *Drosophila* only (control group). When tested with artificially colored crickets, we found that the spiders in group 1 demonstrated an aversion to red, the spiders in group 2 demonstrated a preference for red, and the control group showed no color bias. We discuss the implications of such color biases and learning for the evolution of prey coloration, as well as the implications for sexual selection and the bright red facial coloration of male *H. pyrrithrix*.

Consistencies and Differences in Araneae/Bacterial associations detected from 2006-2012.

Cathy R. Tugmon 1, Norman Horner2

1 Department of Biology, Georgia Regents University, Augusta, GA

2 Director of Natural Laboratories, Midwestern State University, Wichita Falls, TX

From 2006 to 2012 538 spiders from 24 families have been collected in and around the Augusta, GA area. The environments chosen for collection incorporated both urban and rural. From these spiders, approximately 1,215 aerobic bacteria have been isolated. To date, 447 bacteria have been identified and tested for antibiotic sensitivities. The overall data will be discussed concerning urban versus rural bacterial patterns, antibiotic resistance, and consistencies seen in bacterial spider associations from year to year as well as potentially emerging patterns. These data are currently being used to create a data base that will allow us to answer more specific questions concerning the spider/bacterial relationship.

Multimodal signals increase active space of communication by wolf spiders in a complex litter environment

George W. Uetz1, J. Andrew Roberts2, David L. Clark3, Jeremy S. Gibson1,4, Shira D. Gordon1,5; A. Kluckman1

1Department of Biological Sciences, University of Cincinnati, Cincinnati, OH
2 Dept. of EEO Biology, The Ohio State University at Newark, Newark, OH
3 Department of Biology, Alma College, Alma, MI
4 current address: Department of Biological Sciences, University of Missouri, Columbia, MO
5 current address: Royal College Building, University of Strathclyde, Glasgow, UK

We examined the influence of complex microhabitats on transmission of vibratory and visual signals of courting male *Schizocosa ocreata* (Hentz) with Laser Doppler Vibrometry (LDV) and behavioral observations in lab and field. We measured maximum potential detection distance of visual and vibratory signals by females in laboratory mesocosms, recorded vibration signal attenuation on different substrates, and estimated transmission distances for male vibration signals in the field. We also determined effective line of-sight visual detection distances in the field with laser distance measures. Together, these data were used to estimate the potential and effective active space of multimodal signals.
LDV measures show leaves are highly conductive substrates for wolf spider vibratory signals compared to other substrata (soil, wood, rock). For both visual and vibratory modes, lab estimates of maximum potential distance for signal transmission and detection (behavior studies) exceeded estimates of effective active space (signal attenuation,"vanishing point" and "line-of-sight" measures). Field estimates of transmission distance for signal modes overlap, such that in close range (<20 cm), vibratory signals are more likely to be detected, while farther away, visual signals are more likely to be seen. These findings support current hypotheses regarding how multimodal communication might extend the range of overall signal active space or compensate for environmental constraints.

Arachnophobic entomologists: when two more legs makes a big difference

Richard S. Vetter

Dept. Entomology, Univ. California, Riverside CA 92521

Entomologist?
Then one must often handle
Six-legged beasties

Day in and day out
Insect processing occurs
With no thought of fear

Yet spiders evoke
Adverse repellent response
How wondrously strange!

Add just two more legs
The result is disgust, fear
Arachnophobic?

Just how can that be?
Why a different response
To the eight-legged?

Chronoethology of *Cyclosa turbinata* (Araneae: Araneidae)

****J. Colton Watts**, Ashley Herrig, William D. Allen, Darrell J. Moore, Thomas C. Jones

Department of Biological Sciences, East Tennessee State University

Studies examining diel patterns of behavior in spiders are relatively few despite the need to consider consistent behavioral variation when conducting experiments in the laboratory and field. Furthermore, diel patterns of behavior may be adaptive, allowing individuals to coordinate behaviors with ecologically appropriate times of day. We describe web-building, antipredator behavior, and locomotor activity over the diel cycle in *Cyclosa turbinata* (Araneae: Araneidae) under controlled laboratory conditions. We consider these behavioral patterns in an ecological context by considering these data with respect to continuous observations of spiders in the natural habitat. We found that female *C. turbinata* perform web maintenance behaviors (e.g. sticky spiral replacement) several hours before the onset of photophase in both controlled conditions and natural conditions. In either setting, individuals remained in the web hub over the diel cycle and never sought a retreat. Under controlled conditions, we found strong diel patterns in antipredator behavior. During photophase, spiders exhibited more defensive behavior in response to simulated predator cues and devoted more time to defensive posturing. We also found nocturnal rhythms of locomotor activity in the laboratory and field. Individuals generally remained motionless throughout photophase and became active during scotophase. In the laboratory, we found that these nocturnal activity patterns persist in constant conditions, suggesting that circadian mechanisms regulate activity in *C. turbinata*. Our descriptive results lay the groundwork for investigating the ecological significance of diel rhythms of behavior

in orb-weaving spiders.

Effects of season and prey consumption on *Schizocosa* cold tolerance

****Thomas D. Whitney1**, Benjamin N. Philip2, & James D. Harwood1

1Department of Entomology, University of Kentucky, Lexington, KY
2Department of Biology, Rivier University, Nashua, NH

Some temperate forest spiders are considered to be winter-active, including members from the genus *Schizocosa*. While the presence of ice nucleators in the gut is deemed to increase the probability of freezing, and thus mortality in most ectotherms, previous work has shown *Schizocosa* to regularly feed during winter. Therefore, this research sought to elucidate their physiological capacity to remain active in cold temperatures and to determine if predation affects their cold hardiness. *S. ocreata* and *S. stridulans* were collected monthly from a deciduous forest in Kentucky from August 2012 to March 2013 and were subjected to supercooling point (SCP) determination assays. Contrary to other invertebrates, their average SCP remained constant throughout the study, which we later determined was their lower lethal temperature; a laboratory experiment resulted in 100% mortality when spiders were frozen and thawed, while only 53% mortality occurred when spiders were exposed to temperatures near their SCP without freezing. Interestingly, daily low temperatures within the leaf litter fell below the average spider SCP multiple times during winter, exposing them to considerable risk of freezing. To determine if this high risk was a result of winter predation, we fed spiders varying numbers of prey items, but found no significant association with consumption amount and cold tolerance. Despite facing substantial seasonal changes in temperature, our results show that *Schizocosa* do not augment their cold tolerance to better survive. This suggests an ecological tradeoff, where risk of mortality in the population is potentially exchanged for winter growth and thus reproductive advantages in spring.

Female receptivity of *Schizocosa ocreata* wolf spiders (Araneae: Lycosidae) towards potential mates of varying quality in sequential male encounters

****Maggie Williams**, Brent Stoffer, George W. Uetz

Biological Sciences, University of Cincinnati

In the wolf spider, *Schizocosa ocreata,* males possess dark pigmented tufts on their forelegs that are a conspicuous secondary sexual characteristic. In previous simultaneous choice studies, females demonstrated higher receptivity towards males with larger tufts over males with smaller tufts. Even so, in live mating trials with two males, females preferentially mated with the males that courted first, regardless of tufts. However, whether female receptivity changes when males of different quality are encountered sequentially has yet to be investigated. This is particularly interesting in the context of male eavesdropping behaviors, as males may exploit the courtship of others to find females and initiate courtship themselves. We used video playback experiments to introduce females to a 'virtual' courting male (the 'primary' male), followed by a virtual "interloper" thirty seconds later. The interloper male had tufts of greater, lesser, or equal size relative to the primary male. When the primary male had average-sized tufts (control), there was no subsequent effect on female receptivity toward the interloper, regardless of tuft size. In subsequent studies comparing responses to all sequence combinations, females showed more receptivity displays toward interloper males with larger tufts. Moreover, as long as the interloper male had larger tufts, females' latency to approach that male video was significantly shorter. These results suggest that eavesdropping behavior might benefit an interloper male, depending on relative differences in male quality.

Limb-Loss, Regeneration, and Orange Bitey Things (*Pterinochilus murinus*): The impact of autotomy on metabolism

****Brenan Wilson**, Cara Shillington

Eastern Michigan University

Autotomy (voluntary limb-loss) is employed by spiders to escape predation and discard damaged appendages. With successive molts, a lost limb is regenerated and grows to become indistinguishable from those left intact. Despite its frequent occurrence among spiders, many physiologically and ecologically important aspects of autotomy have not been investigated; specifically, the metabolic consequences of autotomy and subsequent regeneration remain unknown. In this study, we tested the effects of autotomy and the relative size of prey on the metabolism, growth, development, and feeding behavior of orange baboon tarantula (*Pterinochilus murinus*) spiderlings under laboratory conditions. Autotomy was induced among half of the spiderlings, while the legs of the other individuals remained intact. Each spiderling was then randomly assigned to a feeding treatment, which was fed prey weighing either ~10% or ~30% of the spiderling's mass, bi-weekly. The metabolism of each spiderling was measured by quantifying the rate of CO_2 production, and a spiderling's willingness to feed was determined from its prey-capture times (the latency between first contact with the prey and capture of the prey). Our results suggest that autotomy and the relative size of prey may have an interactive effect on metabolism, while neither stressor appears to affect metabolism alone. An organism's metabolic response to a combination of stimuli may have indirect, but critical, effects on its fitness.

The role of different eye types in mate recognition and prey detection by female *Schizocosa ocreata* (Hentz) wolf spiders (Araneae: Lycosidae)

****Rebecca Wilson**, Tess Piening and George W. Uetz

Department of Biological Sciences, University of Cincinnati, Cincinnati, OH

Female *Schizocosa ocreata* wolf spiders use visual cues for both prey recognition and capture as well as mate recognition and assessment. Since previous research has shown differences in structure and function of the eye rows, we tested the hypothesis that the anterior and posterior eyes play different roles in detection and decision-making processes involved in mate choice and prey detection. We occluded the different sets of eyes of individual females using an opaque, non-toxic paint, and presented them with either video playback of a courting male stimulus or a live cricket. With courting male video stimuli, latency (seconds) to orient and respond with receptivity were shortest with unmanipulated controls, but when the posterior eyes were occluded, there was a significantly longer latency compared to (unmanipulated) controls. In addition, a composite receptivity score (sum of female displays) was significantly reduced when posterior eyes were occluded. This suggests the posterior eyes are involved in mate detection and recognition. In female responses to live prey, the latency to orient and approach crickets was again shortest for unmanipulated controls and significantly longer when posterior eyes were occluded, but intermediate when anterior eyes were occluded. Taken together, these data suggest that the two sets of eyes may have different roles in detection and identification of prey and courting males. Given differences in apparent use of anterior and posterior eyes in prey and mate recognition/detection, ongoing research is investigating electrophysiological and anatomical capabilities of the two sets of eyes. Results of these studies will be discussed.

The phylogenetic scale of venom variation in Haplogyne spiders

Pamela A. Zobel-Thropp, Emily Thomas, Jennifer Mullins, Miles Dale, Andrew Wood, Greta J. Binford

Lewis & Clark College, Dept. Biology, Portland, OR

Haplogynes includes many taxa of interest with respect to venom composition, however outside of Sicariidae little comprehensive venom work has been done. With a goal of analyzing the phylogenetic scale of venom variation in spiders, we are comparing venom gland transcriptomes and proteomes from representatives of this group selected based on their phylogenetic position. Our dataset allows some comparisons among relatively closely related taxa of sicariids (common ancestor within the last 30 million years) and among other more distant relatives (common ancestor more than 200 million years old). Our center of focus is comparisons among lineages of sicariids, that been evolving for over 100 million years in the context of being generalist, ground-dwelling predators of arthropods. We compare sets of sicariid venom components with toxins from Scytodidae, Pholcidae and non-haplogyne archaeid spiders. We have discovered venom peptide toxin families that appear to be distinct for sicariidae, and others that are expressed across haplogynes. While some peptide and protein toxins are phylogenetically widespread, there are striking differences among lineages in relative abundance, and numbers of paralogs of toxins we capture in transcriptomes. We analyze and compare patterns of gene duplications and positive selection within some of the most common and widespread toxin lineages. Together these data help illuminate the evolutionary dynamics of venom functional complexes in spiders.