

# Abstracts for the NAS – IUSSI Breakout Meeting

at Arecibo, Puerto Rico

Friday – Monday, 24-27 October, 2008

## SESSION 1. DIVISION OF LABOR

1. Invited Talk: **Using the sweat bee *Megalopta genalis* to investigate the transition from solitary to social.** *Adam Smith*; Smithsonian Tropical Research Institute, Panama.

Facultatively eusocial insects allow direct study of the transition between solitary and eusocial living. The facultatively eusocial and solitary tropical sweat bee *Megalopta genalis* is well-suited for such studies. Approximately one half of nests are solitary during the dry season at Barro Colorado Island, Panama, and social groups are small, usually only two or three individuals. Despite small groups, there is strong division of labor: queens perform <10% of foraging trips and foragers have undeveloped ovaries. Moreover, while all nests are apparently initiated by solitary foundresses, solitary nesting is not only a developmental phase: census of post-emergence nests showed that solitary nests tended to remain solitary, and social nests tended to remain social. Adult removal experiments showed that adults protect brood from ant predation, and census showed that social nests are less likely to fail than are solitary nests. Social nests also had higher productivity, and no difference in per-capita productivity, than solitary nests. On average foragers were smaller than both queens and dispersing nestmate females. The chloropid fly *Fiebrigella* sp. is a pollen-robbing cleptoparasite of *Megalopta*, and females parasitized by this fly were significantly smaller than their non-parasitized nestmates, suggesting that the parasite may influence caste expression. Queen removal experiments showed that foragers can reproduce: 16/17 isolated foragers enlarged ovaries, and 15 reproduced, four weeks after removal. Moreover, productivity matched that of unmanipulated solitary controls, suggesting that caste emerges from social competition.

2. **Genetic links between ovary size and social behavior support the reproductive ground plan hypothesis in honey bees.** *Olav Rueppell*; University of North Carolina-Greensboro, Greensboro, NC.

The Reproductive Ground Plan Hypothesis (RGPH) suggests that different aspects of social evolution in insects is derived from the reproductive control modules of their solitary ancestor. One of the most derived aspects of honeybee social evolution is the division of labor among foragers, biasing their foraging behavior towards either pollen or nectar. Genetic influences in form of quantitative trait loci (QTL) on foraging choice have been demonstrated but it has remained to be determined whether these QTL show a direct correlation with worker reproductive physiology, as predicted by the RGPH. This study was designed to test this prediction by investigating the genetic effect of the previously identified four pollen hoarding QTL (pln1-4) on worker ovary size in two reciprocal backcrosses of selected high and low pollen hoarding strains of honey bees. We report direct effects of two QTL (pln2 and pln3) and two new QTL affecting ovary size in worker honey bees. In addition, we report on candidate gene expression data that support the involvement of pln2 and pln3 in worker ovary size determination and tie the behavioral pollen hoarding syndrome to worker ovary size in support of the RGPH.

3. **Rate of behavioral development differences in three honey bee subspecies.** *Gun Koleoglu*(1), Aykut Kence(1), Meral Kence(1), Tugrul Giray(2); (1) Middle East Technical University, Ankara, Turkey; (2) University of Puerto Rico, San Juan, PR.

Worker rate of behavioral development underlies allocation to different jobs such as foraging vs brood care. We have measured rate of behavioral development in a co-fostering design in three experimental colonies each with focal bees from three races; *A.m. syriaca*, *A.m. caucasica*, and *A.m. carnica* from Turkey. This approach of co-fostering is useful to control and even quantify colony background and colony demography effects on rate of behavioral development of genetically distinct bees. The effect of race on becoming a normal age nurse or precocious forager was tested by comparing distributions of nurses or foragers with those of colony samples by two-way G-tests. *A. m. caucasica* bees were always under-represented in forager sample indicating slower rate of behavioral development. *A. m. syriaca* bees were over-represented in forager samples in 2 of 3 composite colonies. These differences in the rate of behavioral development are in line with the prediction that races from warmer climates have faster development rates.

4. **The role of vitellogenin in the regulation of honey bee worker behavior.** *Kate Ihle* (1), Robert E. Page Jr. (1), M. Kim Fondrk(1), Gro V. Amdam(1, 2); (1) Arizona State University, Tempe, AZ. (2) University of Life Sciences, Aas, Norway.

*Vitellogenin*, a yolk precursor gene, is proposed to have a role in pacing the foraging onset and in biasing the foraging loads of honey bee workers. The functions of vitellogenin are debated, but the reproductive ground plan hypothesis (RGPH) and the double-repressor hypothesis (DRH) provide potential frameworks for explaining the role of vitellogenin in the regulation of honey bee worker behavior. While these hypotheses potentially compatible, their direct relationship has been untested. We used selected strains of honey bees with different endocrine sensitivity to vitellogenin, combined with RNA interference (RNAi) to investigate the predictions of and relationship between the RPGH and DRH. *Vitellogenin* knockdown differentially affects the selected strains, demonstrating that the behavioral response to *vitellogenin* activity is conditional on the sensitivity of the endocrine repressor system as predicted by the DRH. We also show that *vitellogenin knockdown* influences foraging behavior in accord with the forager RGPH. These results demonstrate the compatibility of the DRH and RGPH, and show a consistent influence of *vitellogenin* activity on worker behavior.

5. **The relationship between spatial organization and division of labor in the bumble bee, *Bombus impatiens*.** *Jennifer Jandt*; University of Arizona, Tucson, AZ.

In social insects, task-related and reproductive division of labor may be correlated with spatial organization of individuals within the colony. Previous work on spatial organization and its relationship to division of labor, however, has been conducted on species that construct spatially organized nests. Bumble bees are unique in that their colony structure (i.e. brood and food storage areas inside the nest) is not organized in a predictable pattern. We examined space use and task performance of individual bumble bees (*Bombus impatiens*) inside the nest every day for three months. We found that individual bees tend to maintain a specific distance from the colony center over their entire lifetime and that individuals who are more likely to perform the in-nest task of larval feeding are also more likely to use less space inside the nest, whereas individuals who

perform other tasks, such as ventilating the nest, do not use a predictable area inside the nest. We also dissected workers to measure ovarian development, an estimate of reproductive potential, and found that individuals that perform larval feeding are less likely to develop ovaries; however, spatial organization has little to no effect on ovarian development overall. Because spatial segregation occurs in *Bombus impatiens* and there is some evidence to suggest it is related to the performance of in-nest tasks, spatial organization may be a colony-level adaptation. Furthermore, these patterns are not necessarily dependent on the layout organization of the nest structure or body size. Instead, these patterns may be considered a self-organized emergent property.

**6. Ant colony spatial structure and its implications for ant-plant mutualisms.** *Michele C. Lanan, Judith L. Bronstein; University of Arizona, Tucson, AZ.*

Ant protection of extrafloral nectar-secreting plants is a common form of mutualism found in most habitats around the world. However, very few studies have investigated the foraging behavior and spatial structure of the ant colonies that visit these plants. Here, we describe the spatial distribution of nests and trails of the Sonoran Desert ant *Crematogaster opuntiae* (Buren) in relation to the nectar-secreting cactus *Ferocactus wislizeni* (Engelm). To determine the size and position of ant colonies we used a combination of behavioral aggression assays, baiting, and observation of trails between nests from 2007 to 2008. We found that territories of *C. opuntiae* colonies are large, covering areas of up to three hectares. These ants appear to be highly polydomous, with up to twenty nest entrances interconnected by trails and dispersed throughout the territory. Ant workers from neighboring territories are hostile, and will attack each other upon contact. Each territory included many nectar-secreting barrel cacti, and within each territory we observed workers visiting between five and thirty-four plants. Because these cacti grow at different distances from ant nests and territory borders, the quality of ant protection they receive may vary depending on their location. We discuss the implications of the spatial structure of *C. opuntiae* colonies for this mutualism.

**7. Redefining worker caste systems of Pheidole ants with polymorphic major workers.** *Ming Hua Huang; University of Arizona, Tucson, AZ.*

In some ants, workers are morphologically divided into a single small-sized minor caste and one or more larger-sized major caste(s). Based on measurements of a single or a few specimens of over 600 species, Wilson (2003) determined that most New World Pheidole ants have a dimorphic caste system consisting of only one minor and one major worker caste. However, he described seven New World species as having a trimorphic caste system consisting of one minor and two discretely separated major worker castes. To test these designations, I performed a more rigorous sampling of workers of *P. spadonia*, *P. tepicana*, *P. obtusospinosa*, and *P. rhea*. My results show that *P. spadonia* is indeed dimorphic with a distinctly separate minor worker distribution and a unimodal narrow size distribution for major workers. However, *P. tepicana*, *P. obtusospinosa*, and *P. rhea* show three very different types of "trimorphic" worker caste systems. Furthermore, the size distributions of both *P. obtusospinosa* and *P. rhea* changes as their colonies develop from the founding to established stage. Field observations of *P. obtusospinosa* suggest that their large major workers assist in head blocking nest entrances in response to army ant raids. The exact roles of larger-sized major workers in *P. tepicana* and *P. rhea* have not been determined yet.

**8. Endocrinological factors and division of labor in *Pogonomyrmex californicus* founding.** *Adam Dolezal; Arizona State University, Tempe, AZ.*

Investigation into the origins of social insect division of labor can be approached by studying proximate controls of social organization. These investigations have shown that differing social phenotypes are often correlated with divergent hormone titers. This approach has been successfully applied to the honey bee (*Apis mellifera*) and has given rise to a framework proposing that social phenotypes emerged via co-option of the reproductive regulatory systems of solitary ancestors; however, similar investigations using other eusocial insects are necessary to better understand these relationships. Founding queens of *Pogonomyrmex californicus* exhibit behavioral changes during colony founding, with a dichotomy between nest-biased behaviors and field-biased behaviors. Pleometrotic (multiple-queen) founding is common in some populations, during which a division of labor can develop, with same-aged queens performing nest- and field-biased behaviors. To determine if this behavioral diphenism is associated with hormonal differences, we measured juvenile hormone (JH) and ecdysteroid content in i) single founding queens showing behavioral transition and ii) co-founding queens exhibiting a division of labor. We found significant differences in JH titer between nest-bias and field-bias in both single- and co-founding queens, suggesting that JH influences behavioral phenotype. These data provide evidence for the possibility of shared physiological control of behavior for both ants and honey bees and this supports the idea of a shared evolutionary route to social division of labor.

**9. Regulatory effects of reproductive physiology on foraging bias through modulation of sucrose response in honey bees (*Apis mellifera*).** *Adam J. Siegel, Osman Kaftanoglu, M. Kim Fondrk, Robert E. Page, Jr.; Arizona State University, Tempe, AZ.*

There is increasing evidence that elements of division of labor observed today in honey bees have origins in the reproductive signaling networks of solitary ancestors. Individual honey bee foragers usually bias their foraging efforts towards pollen (protein) or nectar (carbohydrate) collection. Ovary status may have a modulatory effect on this foraging bias. We have previously shown that European honey bee workers selectively bred to store larger amounts of pollen (High strain) have a higher number of ovarioles per ovary than workers from strains bred to store less pollen (low strain). Additional evidence from highly related African-European backcross bees that were not selected for foraging behavior supports the link between reproductive physiology and foraging bias. A potential pathway of how reproductive physiology could help regulate foraging behavior through the modulation of sucrose responsiveness can be built using evidence from these two groups of selected honey bees.

**10. The genetic architecture of ovary size differences between Africanized and European honey bee workers.** *Allie Graham, Olav Rueppell; University of North Carolina-Greensboro, Greensboro, NC.*

The evolution of reproductively specialized castes and social behavior from a solitary ancestor may be explained by the reproductive ground plan hypothesis. To test the generality of the hypothesized genetic linkage between reproductive and social behavior, we investigated the genetic architecture of ovary size differences between Africanized and European honey bees. Two backcrosses of a hybrid queen and Africanized drones that resulted in transgressive worker ovary phenotypes were studied for pleiotropic effects of existing behavioral QTL and potential new QTL with a combination of SNP and microsatellite markers. Preliminary analyses showed small but significant effects on ovary size for some of the

behavioral QTL, as predicted by the reproductive ground plan hypothesis. In addition, we detected two new QTL of major effect on ovary size. We describe potential candidate genes for the QTL and suggest that the detected major and minor effects could reflect genetic control of caste divergence and worker division of labor, respectively, representing two distinct stages of honey bee social evolution that may be connected via female reproductive physiology.

11. **Slick dudes: venom and task specialization in *Termitopone commutata*.** Justin O. Schmidt; Southwestern Biological Institute, Tucson, AZ

*Termitopone commutata* is a large shiny ponerine ant species that raids *Syntermes* sp. The raiders quickly overwhelm the termite defenses, enter the termite tunnels, sting and inactivate the termites, and carry them back to their colony. Little natural history and nothing on task specialization – other than that some individuals participate in raiding parties – has been reported for this species. A large colony was excavated and totally censused with individuals separated into categories of raiders, nest defenders, and nest workers. The amount of venom per worker in the three specialities and in alates was measured and the respective lethality of their venoms to both insects and potential predators was determined. Venom activities paralleled needs of each task specialty indicating that physiology and behavior are closely linked in this species.

## SESSION 2. EVOLUTION

12. Invited Talk: **Sociometry: a field still in search of data.**

Walter R. Tschinkel; Florida State Univ., Gainesville, FL. In spite of my clarion call for the systematic collection of data on the basic attributes of social insect colonies (sociometry), there is little sign that the call has been heard, let alone answered. In my talk, I will discuss the importance of such data to understanding the evolution and functioning of the superorganism, as well as describe simple procedures for collecting such information efficiently. The accumulation of sociometric data for all stages of the colony life cycle and across species would give insight into both sociogenesis (superorganismal development) and the evolution of superorganismal life histories. Web-based databases offer an opportunity for the social insect research community to make headway in this important task.

13. **Evolution of colony size in the seed-harvester ant genus *Pogonomyrmex* (Hymenoptera: Formicidae).** Robert A. Johnson(1), Ehab Abouheif(2); (1) Arizona State University, Tempe, AZ; (2) McGill University, Montreal, Quebec, Canada.

Colony size varies by several orders of magnitude across species of social insects. Several evolutionary and ecological factors mold colony size, but few studies examine traits that influence it. This study examines five traits likely associated with the evolution of colony size (body mass of queens and males, ovariole number, number of sperm produced by males, number of sperm stored by queens) for 12 species of ants in the genus *Pogonomyrmex*. The Test for Serial Independence indicated lack of a phylogenetic signal in these traits, suggesting that colony size and the suite of reproductive traits evolved recently and rapidly, and that these traits are not developmentally constrained. Comparative analysis using Independent Contrasts and traditional statistical methods (raw tip data) showed a significant positive linear association of all five reproductive traits with colony size, and of all five traits with each other. We assessed the relative cost of reproductive traits by examining the slope of paired traits across our 12 species. For comparative purposes, we calculated number of sperm per ovariole, which displayed a positive linear association with ovariole number in

contrast to the exponential association that occurs across ant genera. These six traits displayed similar associations across three species of *Messor*.

14. **Sexual selection in social insects.** Jennifer Kovacs, Michael A.D. Goodisman; Georgia Institute of Technology, Atlanta, GA.

Sexual selection shapes mating habits and morphologies in many species. However, few sexually selected traits have been documented in highly social insects. Indeed, some have theorized that sexual selection should be largely absent in social insects, because of the overriding evolutionary importance of maintaining high relatedness among interacting group members. We tested if a form of sexual selection, male mate choice, occurs in the social wasp *Vespa maculifrons*. We first identified the morphological and genetic factors influencing female mating success and overwintering survival. We then determined whether female mating status was associated with survival. Our results reveal that female length is associated with mating success, suggesting that female length may be under sexual selection. Additionally, we found that longer females survived overwintering longer, and therefore female length is an honest indicator of fitness. However, we did not find that mated females survived overwintering longer than unmated females. Interestingly, we also found evidence for a genetic component to female mating success that was not associated with female length or survival. Thus, our data provide some evidence for sexual selection, specifically on female length, in this social wasp, and therefore suggest that social insects are not exempt from sexual selection.

15. **Rare sexual reproduction by a facultative asexual facilitates invasion of novel habitats.** Alexander S. Mikheyev, Stefan Bresson; University of Texas, Austin, TX.

Although numerous models purport to show short-term advantages of sexual reproduction, few data clearly demonstrate the benefits of sex or distinguish among the predictions of these models. Here we demonstrate short-term benefits of sex for adaptation to novel habitats in the invasive facultatively clonal little fire ant *Wasmannia auropunctata*. First, we showed that Central Africa has been invaded by a single queen clone of this Neotropical species at least 95, and likely more, generations ago. Second, we examined the fitness of sexually produced queen derived from the founding clone by following the trajectories of 23 independently evolving populations. While the clone originally introduced to Africa founded almost all of the populations, the sexually produced daughter clones had locally higher fitness and gradually replaced the founder lineage over time. However, the higher fitness of sexually produced lineages came at the cost of genetic diversity, which was lost due to inbreeding. Subsequently, sexually produced clones did not successfully establish new populations. Continuous human transport has assured the persistence of the original clone by selecting it from a background of sexually derived daughter lineages. Our results demonstrate that sex acts on existing genetic diversity to facilitate local adaptation.

16. **Intra- and inter-specific variability of recombination rate in *Apis*.** Emily Meznar, Olav Rueppell; University of North Carolina-Greensboro, Greensboro, NC.

Meiotic recombination is a product of natural selection designed to increase genetic diversity. Its frequency is known to vary both within genomes and between species. The western European honey bee (*Apis mellifera*) and a few other social insects studied in this regard demonstrate the highest recombination rates known among multicellular eukaryotes. To investigate the correlation between insect sociality and recombination rate further, we analyzed the recombination rate between select paired genetic markers in *A. florea*, which is a basal species in the genus *Apis*. We estimated local

and global recombination frequency in this species through direct comparison to *A. mellifera*. In a second experiment, we investigated the intra-specific variability of genetic recombination in *A. mellifera* by comparing recombination distances among two crosses between European and Africanized honeybees and two crosses within European honeybees, testing the prediction that the recombination rate co-varies with genetic similarity of the parental genomes. The results give first insights into the variability of recombination in the genus *Apis* and are discussed in the context of insect social evolution.

**17. Phylogenetic analysis of Range expansion of the fire ant *Solenopsis geminata*.** Heather Axen; University of Vermont, Burlington, VT.

The fire ant *Solenopsis geminata* is considered highly invasive and is found across the globe. Prior to invasions the range of *S. geminata* included parts of northern South America, Central America, Mexico, and the southern United States, though *S. geminata* may be a secondary invader into the southeastern U.S. *Solenopsis geminata* hybridizes with *Solenopsis xyloni* in Texas, where hybrid colonies exhibit genetic caste determination. The hybrid zone may have evolved as *S. geminata* expanded north from Central America and Mexico, or with westward expansion from the southeastern US following colonization there. We constructed a molecular phylogeny using a 653 base pair region of cytochrome oxidase 1 (COI) from workers across North, Central, and South America, and the Caribbean. The phylogeny shows Central America and southern Mexico *S. geminata* basal to two main clades, one clade including *S. geminata* from Texas (hybrid zone) and Mexico, and the other including specimens from Florida, South America, and the Caribbean. A haplotype network of Florida *S. geminata* showed a “star-like” pattern typical of recent introductions. These results suggest the hybrid zone evolved as *S. geminata* expanded from Central America, and southeastern US populations originated from secondary colonization, indicating *S. geminata* may have been invasive historically.

**18. Needle in the haystack – foundress number in *P. californicus* as model for group selection.** Zachary Shaffer; Arizona State University, Tempe, AZ.

Separate populations of *pogonomyrmex californicus* in San Diego county, California display different strategies in terms of foundress number: haplometrosis (a single foundress) predominates through most of the species’ range-- but in one area pleometrotic foundress associations (multiple foundresses) are the norm. Mixed associations of foundresses were created to explore the explanations for the maintenance of this life history strategy in each locale: Single pleometrotic queens, single haplometrotic queens, mixed pairs, five pleometrotic/one haplometrotic, and six pleometrotic foundress groups. Why does one behavioral type predominate in one area and another type in another area? It was found that larger groups had an advantage in terms of survival, brood numbers, and time to first worker eclosion. Contrary to expectations, the haplometrotic (aggressive) foundress type was found in both locales as was the pleometrotic (cooperative) type. The results of this early stage experiment show that this phenomenon may be useful as a natural world model of group selection and questions concerning the evolution of cooperative (seemingly altruistic) behavior.

**19. Innate immunocompetence in *Polistes dominulus*: A critical test of the “haploid susceptibility hypothesis”.** Noah Wilson-Rich, Faith Hester, Philip T. Starks; Tufts University, Medford, MA.

*Polistes* wasps engage in many behavioral interactions. Although there has been debate over the meaning of these interactions, these

stereotypical behaviors can be used to determine a colony's linear dominance hierarchy. Due to the implicit relationship between behavioral and reproductive dominance, behavioral interactions are commonly used to distinguish the reproductively dominant alpha foundress from the beta foundress. It has been suggested that, in order to maintain reproductive control, the alpha foundress is forced to remain at a physiologically constrained activity limit. This, in turn, may allow aggressive interactions to be used as determinants influencing reproductive partitioning between cooperating individuals. The energetic cost of the interactions, however, has not been measured. To address this, we measured the CO<sub>2</sub> production of 19 non-nestmate pairs displaying interactive and non-interactive behavior. The rate of energy use during interaction behavior was positively associated with published rankings of aggression. However, our results indicate that interactions are not very energetically costly in *Polistes*, particularly when compared to the likely cost of foraging. These data suggest that maintaining reproductive dominance is not very energetically expensive for the dominant, and that the dominant foundress expends energy at a lower rate than the subordinate foundress.

**SESSION 3. MATING AND REPRODUCTION**

**20. Choosing a new home: sensory coding of nest site value.** P. Kirk Visscher, Thomas D. Seeley; Cornell University, Ithaca, NY.

We investigated how a scout bee codes the value of a potential nest site in the waggle dances she produces to represent a potential nest site she has discovered. We gave honey bee swarms a choice between a high-value site and a medium-value site and recorded the behavior of individually identifiable scout bees as they reported on these two alternatives. Bees performed equally lengthy inspections at the two sites. However, on the swarm cluster, on average, the scouts for the high-quality site produced many more total dance circuits, though there was much individual-level noise in the coding of site value. The first bee to find a site had a high probability of reporting the site with a waggle dance, regardless of its value. This discoverer-should-dance phenomenon may help ensure that a swarm gives attention to all discovered sites. There was rapid adaptation in the dance response; the number of dance circuits produced by a bee after visiting a site decreased linearly over sequential visits, and eventually each bee ceased visiting her site. This adaptation improves a swarm's decision making ability by making the accumulation of bees at a site “leaky”, thereby helping a swarm avoid making fast-decision errors.

**21. Social dominance enhances male mating opportunities in a paperwasp.** Yamile Molina, Sean O’Donnell; University of Washington, Seattle, WA.

In many animal societies, an individual's opportunities to reproduce correspond to their position in a social dominance hierarchy. Social aggression often regulates competition over food, and thereby affects variation in fecundity. Females of primitively eusocial insects exhibit these patterns: social dominance and direct reproduction are positively associated, and both are negatively correlated with leaving the nest. Unlike females that lay eggs in the nest, males gain reproductive opportunities by leaving the nest to find mates: mate competition and inbreeding avoidance promote male-biased dispersal in social Hymenoptera. *Mischocyttarus mastigophorus* males are exceptional in that they return to their natal nests for prolonged periods, and frequently direct aggression toward female nestmates. These males provide an unusual opportunity to test whether dominance plays a general role in enhancing fecundity in social Hymenoptera. If social aggression promotes male mating opportunities, male social aggression should be positively associated with nest departure. Males' rates of social aggression, food taking behaviour, and rates of nest departure were positively associated. We posit that social aggression favors nutrient acquisition, which then enables males to depart from the nest at younger ages. Nest departure

likely increases the chances of successful mating with unrelated females.

22. **Royal chemistry: Cuticular hydrocarbons and their possible role as a primer pheromone in a dampwood termite.** *Dorit Eliyahu*; Arizona State University, Tempe, AZ.

One of the main characteristics of eusociality is reproductive division of labor. Maintenance of reproductive dominance may be achieved by using primer pheromones that inhibit reproduction in nestmates. However, no such pheromones have yet been identified in termites. In the dampwood termite, *Zootermopsis nevadensis*, we identified four polyene hydrocarbons that occur solely on the cuticular surface of reproductive individuals. The amounts of these specific hydrocarbons, as measured by solvent extractions and gas chromatography analysis, are directly correlated with reproductive status, implying a potential role as a primer pheromone inhibiting reproduction in nestmates. One way a primer pheromone can be spread throughout a large colony is through eggs. Lipid extracts of *Z. nevadensis* eggs do show presence of the reproductive-specific polyene hydrocarbons. Therefore, the effect of eggs on the reproductive development of groups of 4<sup>th</sup> and 5<sup>th</sup> instar termite workers and on paired dewinged alates was tested. Preliminary results suggest that eggs do suppress reproduction. Future work will attempt to establish direct evidence for the role of these hydrocarbons as a primer pheromone.

23. **Are you my mother's? Learning and egg policing behavior in the ant *Camponotus floridanus*.** *Dani Moore*, Juergen Liebig; Arizona State University, Tempe, AZ.

Egg policing is one mechanism by which worker reproduction is regulated in eusocial insect societies. Workers of the ant *Camponotus floridanus* discriminate between queen-laid eggs and worker-laid eggs according to their surface hydrocarbons. However, the hydrocarbon profile of a queen-laid egg changes dramatically during a colony's life. We explore how workers accommodate changes in queen-laid eggs' surface hydrocarbons. We hypothesize that workers learn the profile of the most prevalent eggs in their nest and discriminate against eggs that do not match the familiar profile. We present two tests of this hypothesis. In the first, we provide worker groups with worker eggs, queen eggs, or no eggs and then test the workers' response to queen- and worker-laid eggs after several weeks of exposure. In the second study, we test the response of workers from incipient, intermediate, and established colonies to eggs from incipient queens, established queens, and workers. We find that repeated exposure to worker eggs does induce worker egg acceptance. This suggests learning influences egg policing behavior. However, learning does not appear to be the primary mechanism by which workers respond to the changes in queen-laid eggs' hydrocarbon profiles that accommodate colony growth. Instead we found workers in incipient colonies do not engage in egg policing at all. Future work will focus on the proximate mechanisms that trigger the switch from universal egg tolerance in incipient colonies to egg policing in established colonies.

24. **Cuticular hydrocarbons correlate with fertility, not dominance in a paper wasp.** *Amanda Izzo*; University of Michigan, Ann Arbor, MI.

Cuticular hydrocarbons (CHCs) are information-rich signals in social insects that coordinate a variety of complex behaviors within nests. Unfortunately, the exact information conveyed by CHCs is poorly understood. In particular, there is debate over whether CHCs signal dominance or fertility. This debate continues because 1) reproductive ranks within a nest are chemically distinguishable and ovarian development (i.e. fecundity) correlates tightly with rank; and 2) changes in rank are accompanied by both changes in fertility and in

the chemical profile. We show that fecundity signaling is plausible in paper wasps, yet chemical dominance signaling is unlikely. We found no relationship between cuticular hydrocarbon profiles and the ability to dominate rivals through aggression, suggesting that CHCs are unlikely to provide information about dominance. However, we did find a strong and significant correlation between ovarian development and the CHC profile, suggesting that CHCs could convey valuable information about their bearer's fertility. Furthermore, our data provide a potential mechanism for chemical signaling of fecundity. We discovered a link between endogenous juvenile hormone titer (a gonadotropin secreted during oogenesis), degree of ovarian development, and the CHC profile. Hormonal regulation of CHC profile expression would offer a physiological mechanism that coordinates behavior, physical state, and signal expression.

25. **Effect of Honey Bee Queen Insemination Quantity on Supersedure Rates.** *Elina L. Niño*(1), David Barnes(2), Tom Dowda(2), Joseph Flowers(1), Jerry Hayes(2), Christina M. Grozinger(1); (1) North Carolina State University, Raleigh, NC; (2) Florida Department of Agriculture and Consumer Services, Gainesville, FL.

Queen mandibular pheromone (QMP) plays a significant role in maintaining colony organization and health, and it prevents rearing of new queens (supersedure). Previous studies indicate that queen insemination quantity affects QMP production and therefore queen-worker interactions. In a choice test, workers preferred multi-drone over single-drone inseminated queens. Furthermore, workers exposed to a synthetic QMP are more resistant to starvation and have higher lipid stores. Our objective is to determine if the queen insemination quantity affects supercedure rates and worker physiology.

In this study queens will be instrumentally inseminated with either 1  $\mu$ l or 6  $\mu$ l of semen and placed in two-frame hives. Colonies will be monitored biweekly for four weeks. The number of queen cups and cells will be recorded and subsequently destroyed. At this point we will monitor hives weekly until the queens are superceded. We will also observe the worker retinue response to these differently inseminated queens. Lastly, we will assess the effect of QMP from differently inseminated queens on worker physiology, more specifically differences in lipid storage. Determining if queen insemination quantity affects colony health is important for improving queen rearing, insemination protocols, as well as colony management practices.

26. **The mass departure of a honey bee (*Apis mellifera*) swarm from its parental nest: Triggering signals and signaler identity.** *Juliana Rangel-Posada*; Cornell University, Ithaca, NY.

The timing of departures in group-living animals must be synchronized to assure group cohesion. The mechanisms used by a honey bee colony to organize the departure of a swarm from its nest remain a mystery. We examined the signals and signalers that trigger a swarm's exodus, and we documented the concurrent changes in bee density, mobility, and nest-site recruitment during the swarming process. Using videorecordings, as well as observations at potential nest sites, we analyzed how bees in three swarming colonies prepared for and then performed their sudden departures. We found that over the 60 min before swarm exodus, the production of piping signals gradually increased, and ultimately peaked at the start of the departure. Also, during swarm exodus, bee density dropped markedly, while the average bee velocity, and the production of buzz-run signals spiked dramatically. Neither waggle runs nor shaking signals showed increases before or during swarm exodus. Additionally, we found that bees start searching for potential nest

sites prior to departure, and that nest-site scouts are the first producers of piping and buzz-run signals inside the nest. This explosive swarm departure shows how animals can use the same communication signals in different contexts; we now know that nest-site scouts use piping and buzz-run signals to initiate both a swarm's departure from its nest and a swarm's take-off from its bivouac site. This study also demonstrates how a small minority of individuals in a honey bee colony operates as an oligarchy to decide when to swarm.

27. **Warranted aggression: the informational basis of policing in an ant society.** *Adrian A. Smith*, Bert Hölldobler, Jürgen Liebig; Arizona State University, Tempe, AZ.

Policing behaviors exist in ant societies as a means of regulating worker reproduction and maintaining a reproductive division of labor. Usually, the reproductive efforts of individuals outside of the reproductive caste are inhibited by physical attacks (physical policing) or the destruction of their viable eggs (egg policing). In the ant *Aphaenogaster cockerelli* inhibition of worker reproduction is limited to physical policing. Hydrocarbon blends present on the cuticle and egg surfaces are thought to be the informational basis of policing. We explain the presence of physical policing and lack of egg policing according to available hydrocarbon signals. By identifying and manipulating specific compounds that are unique to reproductive individuals, we provide the first experimental evidence of a causal relationship between cuticular hydrocarbons and worker physical policing. The observed reproductive conflict within societies of *A. cockerelli* is a direct result of the fertility signals utilized by both the queen and worker caste of this species.

#### SESSION 4. CASTES AND COLONY FOUNDING

28. Invited Talk: **Incest in the cold: geographic variation in inbreeding and colony breeding structure in subterranean termites.** *Edward L. Vargo*; North Carolina State University, Raleigh, NC.

Breeding systems of subterranean termites (Rhinotermitidae) are complex and poorly characterized. Colonies are founded by monogamous pairs of primary reproductives. Subsequently, colonies can be secondarily polygamous through production of neotenic reproductives that inbreed within the colony. Using molecular markers, we have characterized the colony breeding structure and levels of inbreeding in several populations in *Reticulitermes flavipes* in the U.S. and *R. grassei* in southern Europe. We found considerable variation among populations in both species in the proportions of colonies headed by primaries and neotenic and the levels of inbreeding. When plotted geographically, this variation showed a latitudinal trend, with populations further north being more inbred. These results suggest that colony breeding structure and degree of inbreeding are responsive to environmental conditions that vary along a latitudinal gradient. Possible reasons for this trend are discussed.

29. **A comparison of nest characteristics across three species of harvester ant that differ in colony founding strategy.** *Brittany Enzmann*, Univ. of California-Los Angeles, CA.

Harvester ants exhibit a trait termed "claustrality" that is characterized by the degree which foundresses rely on internal reserves to raise their first brood of workers. Claustrality is positively correlated with mass and stored reserve (claustral > facultative > semi-claustral) and is variable across species of *Pogonomyrmex* harvester ants. Foundresses of three species that differ in claustrality were observed initiating nests under laboratory conditions in the summers of 2005, 2007 and 2008. *P. rugosus* is

fully claustral, *P. salinus* is facultative, and *P. californicus* is semi-claustral. Preliminary results indicate a significant positive association between mass and digging rate in *P. salinus* during the first three days of colony initiation, but not in other species or time periods. Across species, degree of claustrality was positively associated with mean digging rate and nest depth. Nest depth and branching were qualitatively distinguishable across species: *P. californicus* dug shallow nests with few branches, *P. salinus* dug to intermediate depth with many branches, and *P. rugosus* dug deep vertically-oriented nests with intermediate branching. *P. rugosus* foundress mass was positively associated with nest depth by day two of colony initiation. Foundress size did not affect hole plugging behavior and neither size nor digging rate affected offspring production.

30. **A role for maternal effects on caste determination in a facultatively social sweat bee (*Megalopta genalis*)?** *Karen M. Kapheim*(1), Adam R. Smith(2), Kate Ihle(3), Gro V. Amdam(3), William T. Wcislo(2), Peter Nonacs(1); (1) University of California-Los Angeles, CA; (2) Smithsonian Tropical Research Institute, Panama; (3) Arizona State University, Tempe, AZ.

Facultatively social species are particularly informative for studies of social evolution, because they remain closely linked to the evolutionary processes responsible for the origins of eusociality. It is generally thought that caste determination in primitively eusocial insects is largely based on dominance interactions among adults, while pre-imaginal processes are more important in highly eusocial species. Yet, the connection between developmental nutrition and reproductive physiology is ubiquitous among insects. It is thus possible that the outcomes of dominance interactions in primitively eusocial species are influenced by developmental processes.

Although all female *Megalopta genalis* are apparently capable of reproducing independently, many forego dispersal to remain on their natal nests as subordinate workers with a strong division of labor.

Preliminary inclusive fitness estimates suggest *Megalopta* sociality may have evolved through selection on maternal effects, rather than altruistic workers. As mass provisioners, foundresses can potentially manipulate the reproductive potential of their daughters through differences in nutritional quality of individual pollen masses. We investigated the role of maternal effects on caste determination in *M. genalis* by quantifying differences in ovary development, juvenile hormone, ecdysteroids, and vitellogenin titers among females raised in social isolation and subsequently compared these to females of known caste.

31. **Larval castration in a ponerine ant: a mechanism for the suppression of larval queen development through aggression.** *Clint Penick*, Arizona State University, Tempe, AZ.

While conflicts over reproduction arise amongst adults in eusocial insect colonies through aggression and egg policing, an earlier conflict occurs between adult workers and developing larvae—when a larva has the potential to develop into either the worker caste or reproductive caste. In the ponerine ant *Harpegnathos saltator*, larvae can choose their developmental fate because they are not fed by adults but feed on their own within the nest. This conflict over larval development became evident when queen development was induced in larvae and adult workers responded. Application of a juvenile hormone analog to larvae induced partial queen development, with major morphological changes occurring at pupation. When treated larvae were returned to the nest, nurse workers began to attack the larvae within 24 hours by biting them and restricting larval feeding. This worker response provides a mechanism for sisters to suppress the development of "cheater" larvae who attempt to develop into reproductives instead of workers. Instead of sacrificing cheater larvae, these ants have developed an alternative mechanism to suppress queen development and provide a unique behavior to help understand the conflict over caste determination in insect societies.

32. **Origin and evolution of the dependent lineages in the genetic caste determination system of *Pogonomyrmex* spp.** *Anu Vihavainen* (1), Pekka Pamilo (1,3), Robert A. Johnson (2), Robert E. Page (2), Jurgen Gadau (2); (1) University of Oulu, Linnanmaa, Finland; (2) Arizona State University, Tempe, AZ; (3) University of Helsinki, Finland.

We combined population genomics and linkage mapping using app. 1200 nuclear markers and partial DNA sequence of 600 bp of the mitochondrial gene COX1 to characterize the genetic basis of a genetic caste determination system in *Pogonomyrmex*. A total of 94 *P. barbatus*- and *P. rugosus* individuals (a virgin queen and worker from 47 colonies) derived from previously studied GCD populations were genotyped for 17 AFLP primer pairs. These resulted in 1147 polymorphic markers. Principal Coordinate Analysis for these nuclear markers gave us a tool to detect admixture of genomic DNA of dependent lineages. Our results confirmed the existence of four main lineages that corresponded well with the branches of mitochondrial Neighbor Joining tree constructed on the basis of ~600 bp of *cox1* gene for all samples. Statistically significant differences between workers and queens identified 165 candidate genetic caste determination markers. To determine the distribution of these 165 candidate markers within the genome we constructed a linkage map based on AFLP markers for 92 haploid non GCD-*P. rugosus* males derived from the same queen. Nineteen of the previously identified 165 GCD markers segregated in this mapping population and could be assigned to a specific map location. The 19 markers were distributed throughout the genome making a single locus or two locus model for GCD unlikely.

33. **Nutritional levels of wasps during the different stages of the colony cycle.** *Timothy M. Judd*, Roxane Magnus, Matthew Fasnacht; Southeast Missouri State University, Cape Girardeau, MO.

In wasps, nutrition plays a vital role for colony cohesion and caste determination. However, there is no baseline data set for the nutritional levels of wasps during the different stages of the colony cycle. Here we examined the levels of carbohydrates, lipids, protein, Ca, Cu, Fe, K, Mg, Mn, Na, and Zn in the wasp *Polistes metricus* at different stages of the wasp's lifecycle. Wasps were collected at the following stages 1) spring gynes, 2) foundress colonies, 3) early worker colonies, 4) late worker colonies, 5) emerging reproductives (gynes and males), 6) early fall reproductives, and 7) late fall reproductives. All eggs, larvae, pupae and adults were analyzed for their nutritional content at each stage to determine if there were any differences between the nutrient levels in the different castes. In addition, changes in nutrient levels during the lifetime of the reproductive females and males were also measured. Overall, the results show there are distinct differences between the levels of macromolecules and cations between the different castes and between early and late workers. In addition, the reproductives the nutrient levels changed during their life time. This presentation will highlight the differences between the adults of each caste and the changes in nutrient levels for the adult reproductives during their lifetime.

34. **The trophic ecology of castes in an ant colony.** *Chris R. Smith*, Arizona State University, Tempe, AZ.

Despite an historic knowledge of the role of diet in determining castes in honey bees, little work has examined diet quality and caste in ants. We analyzed the elemental (C:N) and isotopic (<sup>13</sup>C and <sup>15</sup>N) composition of all castes in *Pogonomyrmex badius*, the Florida seed harvester ant, in colonies at each of two sites. Each of the four castes (male, reproductive female or gyne, major worker and minor worker) is distinguishable based on the diet they received as larvae. Colonies at the two sites differed in their relative trophic position, but within colonies the largest individuals (by mass: gynes>major workers>males>minor workers) were fed a better diet as judged by more insect/protein relative to seeds. This result suggests that the largest castes are preferentially fed a higher quality diet by nurses,

who control larval feeding. Solitary insects sampled at these sites had similar C:N as male and female reproductives, supporting the historic notion that workers are nutritionally castrated. In terms of diet similarity among castes the males and gynes were most similar suggesting that seasonal changes in food availability are a mechanism for controlling caste production, even when caste determination is partially genetic.

35. **Lineage ratios in the *Pogonomyrmex* seed-harvester ants with genetic caste determination.** *Kirk E. Anderson*; University of Arizona, Tucson, AZ.

In insect societies, worker versus queen development (reproductive caste) is typically governed by environmental factors, but some *Pogonomyrmex* seed-harvester ants exhibit strict genetic caste determination, resulting in an obligate mutualism between two reproductively isolated lineages. Same-lineage matings produce queens while alternate-lineage matings produce sterile workers. As both worker and queen phenotypes are required for colony growth and reproduction, colony fitness is influenced by the relative frequency of each lineage and queen mating frequency. Theory predicts that negative frequency-dependent selection will lead to stable lineage frequencies at 0.5/0.5. We surveyed the lineage ratio of 46 populations and found that the majority deviated significantly from equal lineage frequencies. To understand factors that may enable the maintenance of asymmetric lineage frequencies, we constructed a simulation model that incorporates realistic colony growth and reproduction processes. Our model demonstrates that an ecological advantage of one lineage can lead to the maintenance of stable but asymmetric lineage frequencies. Further analysis of our population survey revealed that an ecological factor, elevation, was strongly correlated with lineage frequency. Thus, one lineage appears to have an ecological advantage over the other at higher elevations, and this ecological factor may explain why asymmetric lineage frequencies are widespread, and apparently stable.

36. **Antennal drumming and larval development in *Polistes*.** *Robert L. Jeanne*; University of Wisconsin, Madison, WI.

Paper wasps produce vibrational signals in a variety of contexts. Here I focus on antennal drumming (AD), a mechanical signal produced by female *Polistes* in the context of feeding the larvae. Of the several existing hypotheses on the function(s) of AD, all assume that it is a releaser stimulus, and none is well supported. I propose that AD is instead a modulatory signal that induces biochemical changes that mediate gene-expression differences, which in turn bias larvae to develop into adults with worker-like traits. I argue that differences in food quantity alone is not enough to cause observed developmental patterns, and that AD provides the necessary additional input. It is well known that stressors of various kinds, including mechanical ones, induce significant biochemical changes in insects and vertebrates. Thus, AD may well trigger a cascade of events linking biogenic amines, through hormones, to gene expression, to patterns of development. Support for the hypothesis is circumstantial so far, but includes the fact that the first performance of AD in a colony coincides with the appearance of the first 3rd instar larvae in the nest. This is the same instar in which the caste-determining switch occurs in *Apis* and in some vespines.

37. **From conflict to cooperation: colony founding by unrelated ant queens.** *Rick P. Overson*, Jennifer Fewell, Stephen Pratt, Rebecca Clark, Jurgen Gadau; Arizona State University, Tempe, AZ.

Most cooperating associations consist of relatives and their origin and maintenance is described by kin selection theory. Those cases in which non-relatives form stable cooperative associations are thus particularly interesting. Unlike cooperation between relatives, to remain stable through evolutionary time, cooperation among non-relatives must provide direct benefits to each group member which outweigh each individual's fitness costs from cooperation. In order to reach a state in which cooperation can occur however, individuals

## SESSION 5. LIFE HISTORIES

must first aggregate in groups which tolerate one another. Much research has been done exploring the costs and benefits of cooperation at the level of the individual and of the trait-group to understand how these associations are maintained. However, comparatively little work has been done on the behavioral mechanisms which accompany the switch from a solitary to cooperative life history. The California harvester ant *Pogonomyrmex californicus* is ideal for studying the behavioral mechanisms involved in this switch because populations of strictly haplometrotic queens (solitary) as well as a localized population of pleometrotic queens (communally social) coexist in close proximity to one other. We investigated the behavioral differences between queens from haplometrotic and pleometrotic populations in *P. californicus*. A significant difference in queen behavior between these two distinct groups is their tolerance for conspecific queens during the founding stage of colony development. We argue that the evolution of tolerance towards co-foundresses is the essential mechanism enabling a transition from solitary to communal social foundresses in *P. californicus*.

### 38. **My house, my rules: Acceptance of alien conspecifics in the paper wasp *Mischocyttarus mexicanus*.** Floria Mora-Kepfer; University of Miami, Miami, FL.

In primitively eusocial wasps, little attention has been paid to the effect of colony developmental stage in recognition and acceptance of alien conspecifics. Here I analyze recognition behavior of nestmates according to alien age in newly founded nests and more established nests of *Mischocyttarus mexicanus*. I performed field experiments in a subtropical population in Miami, where I filmed the introduction of alien wasps to foreign newly founded nests and more established nests. Nests with offspring were excluded from this study. My results demonstrate that residents of newly founded foreign nests accept alien conspecifics more than in established nests. Alien behavior also influences acceptance into a foreign nest. Older alien wasps are more aggressive when put in contact with residents of a foreign nest. Number of resident wasps in a nest did not have an effect on acceptance or rejection of an alien. In this experiment we obtained similar results to previous studies that demonstrated that young aliens are more accepted into foreign colonies due to lack of an established chemical profile. However, my results show that both colony developmental stage and alien behavior play an active role in acceptance or rejection into a foreign colony.

### 39. **Nest building in *Polybia occidentalis*; using a model to understand task coordination.** Teresa I. León; University of Wisconsin, Madison, WI.

Nest building in the Neotropical wasp *Polybia occidentalis* requires the task coordination between three specialized worker groups. Water foragers collect water that is used by pulp foragers and builders to moisten pulp. Pulp foragers collect the pulp used by builders. There is no central control for this process; rather it is thought that one or both of two regulating mechanisms are used. The system may be demand-driven in that the builders drive the pace of construction, with water and pulp foragers responding to the demand from builders. This is thought to coordinate construction and minimize queuing delays during material transfer. Alternatively, the system may be supply-driven in that the pace of construction relies on the availability of either pulp or water. Material foragers work as fast as availability allows, and the other worker groups match their efforts to this pace. To investigate the implications of these two contrasting hypotheses for task coordination, I constructed a model to predict patterns of worker activity under different assumptions of worker behavior. By comparing the model output with data, I can identify which hypothesis is most likely to explain observed *P. occidentalis* nest building behavior.

### 40. **Life history trade-offs in social insects.** Bert Rivera-Marchand; InterAmerican University, Bayamon, PR.

Defense is one of the most important factors affecting life history. The relationship of defense to life history traits as well as its possible costs has been reviewed extensively for many groups, including plants. However, defense in social insects, such as honey bees, has never been examined from a trade-off perspective, although defense in honey bees, *Apis mellifera* L. (Hymenoptera: Apidae), has been widely studied. Here I discuss the life history traits of honey bees, particularly traits related to defense. I then examine trade-offs in the context of resource availability. Lastly, I offer suggestions for future research on trade-offs in honey bees and other social insects.

### 41. **Resin collection as a colony-level immune defense in honey bees.** Mike S Simone (1), Jay Evans (2) and Marla Spivak (1); (1) University of Minnesota, St. Paul, MN; (2) USDA-ARS Bee Research Lab, Beltsville, MD

This study aimed to determine if the use of resins, complex plant secretions with diverse antimicrobial properties, acts as a colony-level immune defense by honey bees. The harvesting and incorporation of antimicrobial compounds as propolis in nest architecture is an exciting, but relatively unexplored behavior. We hypothesized that propolis reduces in-hive microbes lowering a bee's physiological investment in its immune system. In year one of the study, colonies were enriched with extracts of Brazilian propolis, Minnesota propolis or deprived of propolis. Larvae and age-marked bees were collected from each of 18 colonies. We measured gene transcript levels of several immune-related genes (abaecin, apidaecin, defensin1, hymenoptaecin, AmEater). We also measured gene transcripts of the 16S rRNA loci to quantify eubacteria and vitellogenin to indicate stress. Significant differences in hymenoptaecin and AmEater gene transcript levels were found in 1-week old bees with those from propolis enriched colonies having reduced expression. Subsequent studies in year two examined how propolis affects immune-gene transcript levels of bees in healthy and pathogen-challenged colonies naturally collecting more propolis compared to colonies that collected little or were enriched with propolis. This is the first known evidence of the nest environment affecting immune expression in honey bees.

### 42. **Life history and social factors constrain independent reproduction in a primitively eusocial wasp.** Hans Kelstrup, Daniele Fanelli, Seirian Sumner; University of Copenhagen, Denmark.

In cooperatively breeding primitively eusocial insect societies and vertebrate societies, helpers are capable of leaving the group and reproducing independently, and yet do not. A fundamental question, therefore, is why helpers do not leave and start their own group where they may be the dominant reproductive. Independent nesting is often constrained by life history and ecological factors. Most studies to date have considered these constraints separately, but it is likely that a combination of these factors constrain independent nesting (the broad constraints hypothesis). We tested the broad constraints hypothesis in a primitively eusocial paper wasp (*Polistes canadensis*) where all females are able to become egg-layers. We relaxed the constraints on independent reproduction and gave 31 helpers the opportunity to become dominant egg-layers on established nests containing related brood by removing all their nestmates. We found that helpers were likely to remain on their nest if new adults were about to emerge and if they were young. This indicates that independent reproduction is constrained by a combination of life history (age-dependent fertility) and ecological (group living benefits) factors. A holistic approach to testing constraints

hypotheses with relative quantification of the component parts may reveal key differences in how sociality evolves.

43. **Effects of food limitation and the environment on colony performance in the Fungus Gardening Ant, *Trachymyrmex septentrionalis*.** Jon Nicholas Seal, Universität Regensburg, Regensburg, Germany.

Recent phylogenies indicate that the leaf-cutting ants (*Atta* and *Acromyrmex*) shared its most recent ancestor with several temperate species of *Trachymyrmex*. *Trachymyrmex septentrionalis*, a widespread fungus-gardening ant, has become a model system in understanding the evolutionary ecology of higher fungus-gardening ants. Populations of this species in northern Florida often exceed 1,000 nests per hectare. When colony size and performance were estimated from excavations and the weight of sand in the tumulus, nests were larger and more productive in open treeless habitats than wooded areas, probably because soils of open sites stimulate worker activity and colony growth and cool soils depress performance. Moreover, wooded sites exhibited evidence of recruitment limitation, since the number of ant nests correlated positively with colony size. Food supplementation experiments in wooded areas partly support this, since supplemented colonies greatly increased production of ant and fungal biomass. The results indicate that the population is food limited and colonies could compete for relatively rare fungal substrate. However, intraspecific competition in this species is not obvious and if it occurs, it is restricted in open, barren sites. One possible explanation is that, according to theory, their mode of colony founding is poorly adaptive to competitive environments.

44. **The energetic costs of stereotyped behavior in the paper wasp, *Polistes dominulus*.** Susan A. Weiner, William A. Woods Jr., Philip T. Starks; Tufts University, Medford, MA.

*Polistes* wasps engage in many behavioral interactions. Although there has been debate over the meaning of these interactions, these stereotypical behaviors can be used to determine a colony's linear dominance hierarchy. Due to the implicit relationship between behavioral and reproductive dominance, behavioral interactions are commonly used to distinguish the reproductively dominant alpha foundress from the beta foundress. It has been suggested that, in order to maintain reproductive control, the alpha foundress is forced to remain at a physiologically constrained activity limit. This, in turn, may allow aggressive interactions to be used as determinants influencing reproductive partitioning between cooperating individuals. The energetic cost of the interactions, however, has not been measured. To address this, we measured the CO<sub>2</sub> production of 19 non-nestmate pairs displaying interactive and non-interactive behavior. The rate of energy use during interaction behavior was positively associated with published rankings of aggression. However, our results indicate that interactions are not very energetically costly in *Polistes*, particularly when compared to the likely cost of foraging. These data suggest that maintaining reproductive dominance is not very energetically expensive for the dominant, and that the dominant foundress expends energy at a lower rate than the subordinate foundress.

45. **Social scaling in the seed-harvester ant *Pogonomyrmex californicus*.** Tate Holbrook, James S. Waters, Jon F. Harrison, Jennifer Fewell; Arizona State University, Tempe, AZ.

Body size has profound consequences for the structure, function, and ecology of organisms. Scaling relationships may also shape life at higher levels of biological organization, including insect societies, which vary tremendously in number of individuals and biomass. We asked whether colony size is associated with changes in metabolism, brood productivity, and/or organization of work in the seed-harvester

ant *Pogonomyrmex californicus*. Whole-colony metabolic rate, brood-to-worker ratio, and task performance were quantified in one-year-old laboratory colonies ranging from approximately 30 to 600 workers. Metabolic rate scaled to the  $\frac{3}{4}$  power of colony mass, indicating that individuals in larger colonies consume less energy on average. This effect could not be explained by differences in worker body size, per capita brood productivity, activity, or division of labor – none of which was significantly associated with colony size. The remarkable similarity in metabolic scaling between *P. californicus* colonies and unitary organisms may reflect a fundamental principle that spans levels of organization, though the underlying mechanism(s) remains elusive.

46. **Enhanced colony growth rates in GCD populations of *Pogonomyrmex harvester* ants.** S. Helms Cahan, A.B. Daly, T. Schwander; University of Vermont, Burlington, VT.

Reproductive caste is typically determined by environmental cues rather than by genetic predisposition. Environmental caste determination is predicted by kin selection theory and for ergonomic reasons, as offspring biased toward a particular caste regardless of colony needs are expected to waste resources and reduce colony growth. Nevertheless, genetic caste determination (GCD) is known from several populations of *Pogonomyrmex harvester* ants over a wide geographic region in southwestern North America. We tested whether GCD reduces performance by comparing colony growth rates of two GCD lineage pairs (H and J) with two closely related non-GCD congeners, *P. rugosus* and *P. barbatus*, under controlled laboratory conditions over a range of temperatures. In contrast to expectations, GCD lineages displayed significantly higher colony growth rates than *P. rugosus* across all temperatures, and equivalent growth rates to *P. barbatus*. Analysis of possible mechanisms indicated that the primary cause of growth rate differences is that GCD workers, which are of inter-lineage hybrid ancestry, hatch earlier and move through larval stages more rapidly than non-GCD workers. This suggests that the unusual genetic composition of GCD workers provides an individual-level advantage that can counteract colony-level costs of the GCD system and allow such populations to persist over the long term.

## SESSION 6. ADAPTATION

47. **Reorientation after swarming: makes a hive a home?** Elizabeth Capaldi Evans; Bucknell University, Lewisburg, PA.

48. **Spatial analysis of foraging activity of the polygynous red imported fire ant *Solenopsis invicta* (Hymenoptera: Formicidae).** Charito Orenco Rodríguez; University of Puerto Rico, San Juan, PR.

The polygyne form of *Solenopsis invicta* has been poorly studied in terms of foraging behavior. External marking ultraviolet fluorescent ink of foragers and recruits will allow to map complete polygyne foraging areas and also observe co-occurrence of colonies at baits. Some basic questions about polygyne foraging organization will be answered: 1) How far do polygyne ants forage and how is the mound volume related to their foraging areas? 2) Do colonies co-occur at a single bait? 3) Can bait distance and/or colony biomass determine food acquisition success? 4) Do polygyne foraging areas overlap? 5) If so, to what extent? 6) Does quantity of neighbors affect polygyne foraging area and/or shape?

49. **Behavioral responses of two subterranean termite species (Isoptera: Rhinotermitidae) to instant freezing or chilling temperatures.** Xing Ping Hu; Auburn University, Auburn, Alabama.

The behavioral responses to instant freezing or chilling temperatures and survivorship of the Formosan subterranean termite, *Coptotermes formosanus* Shiraki, and the Eastern subterranean termite,

Reticulitermes flavipes (Kollar), were studied using a novel experimental design that closely simulated subterranean termites' natural in-ground environment. Both termite species responded to changes in temperature by exhibiting a downward mass movement from the cold to warmer area of constant temperature. However, the degrees of response were specific to the species and temperature regimen. Approximately 88 and 96% of *R. flavipes* escaped from instant 0°C and chilling regimens (from 24 to 0°C at a rate of 1°C/h or 1°C/12 h), respectively, compared with 77 and 91% of *C. formosanus*. No significant difference was detected between the two cooling regimens in either termite species. Controls resulted in a relatively even distribution within test tubes in both termite species. The small portion of the termites that did not escape endured a cold coma at a 24-h 0°C and had low mortality of 2.2 and 1% in *R. flavipes* and 5.2 and 3% in *C. formosanus* at instant and chilling regimens, respectively. This result may have implications for understanding group intelligence and decision making evolved by subterranean termites to survive temporary freezing cold.

**50. *Melittobia* parasitoid wasps display unusual behavior and life history attributes that preage eusociality.**

*Robert M. Matthews*, University of Georgia, Athens, GA.

Behavior and life history parameters of a gregariously developing parasitoid wasp are very ant-like. Comparisons between ants and *Melittobia* (Eulophidae) reveal that foundress females in both initially produce morphologically distinct daughter "castes" who remain with mom (overlapping adult generations) assisting to produce more offspring. Adult offspring of both cooperate to achieve tasks that benefit the group and that individuals working alone are incapable of doing. Although highly inbred, a given clutch of female *Melittobia* offspring display varying degrees of genetic relatedness. Implications suggest that antecedents of eusocial behavior arose early in the Hymenoptera clade.

**51. Consequences of a cryptic invasive ant (*Pachycondyla chinensis*)- Local extinctions mediated by behavior?**

*Benoit Guénard*, Rob R. Dunn, Jules Silverman; North Carolina State University, Raleigh, NC.

Invasive species and in particular ants are one of the most important threats for biodiversity, but are mostly restricted to human disturbed environments. Invasive species remain rare in most undisturbed habitats. The Asian ant, *Pachycondyla chinensis*, was introduced no fewer than 75 years ago to the United States. Recently it was noticed that this species is increasing in abundance and expanding into not only disturbed but also undisturbed forest ecosystems of the southeastern US. We evaluated the impact of this species on the native ant communities in four forests in North Carolina and in paired sites in the species native range in Japan. We also conducted behavioral observations of *P. chinensis* and native species. Our results show that *P. chinensis* has a strong negative impact on most of the native species, with apparent local extinction, but has at some density a positive impact on a few other species. We speculate that these effects are mediated by aspects of the behavior of *P. chinensis* and offer speculation as to what these might be.

**52. Dispersal and mate choice in the Formosan subterranean termite.** *Claudia Husseneder*; Louisiana State University, Baton Rouge, LA.

A subterranean termite colony is initiated by a monogamous pair. Partner relatedness and heterozygosity determine the genetic diversity within the colony, which may be important for colony founding success. We tested whether swarming individuals select partners according to genetic diversity and whether they avoid pairing with relatives. Pairing was random with respect to

relatedness, showing no evidence of kin avoidance. Interestingly, females forming tandems with males had a higher degree of heterozygosity than females that remained single. Although there was no kin discrimination during partner selection, there was little inbreeding due to long dispersal distances and due to genetic differentiation between the sexes. The dispersal distance of alates was sufficient to provide mixing of an average of 13 colonies within swarm aggregations. Genotypic frequencies differed significantly between males and females. This could not be explained by sex-biased dispersal but by sex-biased investment depending on colony breeding structure. Inbred colonies produced predominantly males, while outbred colonies produced predominantly females. This discrepancy could be caused by the pairing advantage of heterozygous females.

**53. Intraspecific color pattern polymorphism and individual recognition abilities in paper wasps.** *Michael Sheehan* and Elizabeth Tibbetts; University of Michigan, Ann Arbor, MI.

Social insects rely heavily on chemical signals during social interactions to communicate many important types of information such as colony membership and fertility state. Among paper wasps some species have variable color patterns used for individual recognition, though most species have uniform, species-typical color patterns. The information content of a signal correlates with its variability; more variable signals provide more information. In the case of identity signaling, increased polymorphism allows each individual to be easily recognizable. Species with low levels of color pattern variation, however, are predicted to lack individual recognition. We tested the extent of recognition abilities in three *Polistes* species with varying levels of intraspecific color pattern polymorphism. *P. fuscatus*, the most variable species, remembered individuals for a week despite interacting with ten other wasps. This is the most robust social memory demonstrated for an insect. Less variable species, *P. dominulus* and *P. metricus*, did not individually recognize conspecifics. Collectively, these data confirm that individual recognition in *Polistes* requires the sort of highly variable color patterns seen in *P. fuscatus*. This study demonstrates that relatively small amounts of phenotypic variability are unlikely to be used for individual recognition, underscoring the need to behaviorally test for recognition abilities.

**54. Volatiles from diseased brood elicit honey bee hygienic behavior.** *Jodi Swanson(1), Baldwyn Torto(2), Steve Kells (1), Marla Spivak(1)*. (1) University of Minnesota, (2) USDA-ARS Chemistry Research Unit, Gainesville, FL.

Honey bee hygienic behavior is a form of social immunity in which the behaviors of individual bees comprises a colony-level defense to pathogens and parasites. Bees within colonies selected for hygienic behavior detect and remove diseased brood before the pathogen reaches an infectious stage, thus reducing disease transmission within the nest. Laboratory experiments demonstrated that hygienic bees have high olfactory sensitivity to the odors produced by diseased brood. Here we isolated the volatile compounds from diseased brood that elicit the behavior. We collected and analyzed volatiles produced by honey bee larvae infected with *Ascosphaera apis*, a fungal pathogen. Using gas chromatography coupled with electroantennographic detection (GC-EAD) we determined that adult honey bees detected three compounds: benzyl alcohol, phenyl alcohol and phenethyl acetate. Two field bioassays showed that application of phenethyl acetate a) induced removal of topically treated, healthy larvae and b) inhibited capping of paraffin larval dummies. The responses were significantly correlated with the degree of hygienic behavior of the colony (i.e. rapid-hygienic colonies removed more topically treated larvae and capped fewer paraffin dummies compared to slow-hygienic colonies in a concentration dependent manner). These experiments support the hypothesis that hygienic behavior is mediated by olfactory cues and is based on a threshold response in which bees with the highest

olfactory sensitivity initiate the hygienic response more quickly compared to bees with lower olfactory sensitivity.

## POSTER SESSION

### 1. **The Birds and the Bees: Nest Site Competition between Honey Bees and Cavity-nesting Birds.** *Kyla Ercit and Gard W. Otis*; University of Guelph, Ontario, Canada.

A number of bird taxa obligately nest in cavities. Larger cavity-nesting bird species compete with honey bees for nest sites. This is especially true for larger species of parrots; some owls, falcons, and ducks; and hornbills. Because of their larger population densities and smaller nest sizes, Africanized honey bees compete more intensely with birds than do European bees. The strongest evidence of negative competitive effects of honey bees on birds comes from Puerto Rico. Since the arrival of Africanized honeybees, every nest box put out for use by the endangered Puerto Rican Parrot becomes occupied within a year by Africanized bees. Continuous maintenance of boxes and removal of bees is essential to provide nest sites for the parrots. Our survey suggests that where bees are having the greatest effects on birds they are introduced species (e.g., European bees on cockatoos in Australia; African bees on parrots in tropical America).

### 2. **Space invaders: nest-site competition between an introduced paper wasp, *Polistes dominulus*, and cavity-nesting Birds.** *Chris Earley, Gard W. Otis*; University of Guelph, Ontario, Canada.

The populations of many species of cavity-nesting birds are limited by the number of nest sites available. Low numbers of available nest cavities can cause competition with conspecifics, other bird species, mammals and even insects. Introduced honey bees (*Apis mellifera*) are known to compete with, or at least occupy the nest sites of, many cavity-nesting birds around the world. The European Paper Wasp (*Polistes dominulus*) is an invasive species that was introduced to North America in the late 1970's. This wasp builds its nests in a variety of locations including human-made nest boxes, making it a possible competitor with cavity-nesting birds that utilize nest boxes. Many naturalists and researchers who monitor nest box lines now complain about the presence of these wasps, but little research has been done on the real impacts of the wasps on the birds that use nest boxes. Boxes where wasp nests were removed continuously were found to fledge more young birds than boxes where wasp nests were left intact. This suggests that the wasps could be a problem in natural cavity nest sites as well.

### 3. **Factors influencing mating behavior in *Lygus hesperus*.** *Colin Brent*, USDA-ARS, Arid-Land Agricultural Research Center, Maricopa, AZ.

The timing and frequency of mating events in insects is often determined by the physiological state and previous interactions of both females and males. Not only can these factors affect an individual's proclivity to mate, but they can also influence the behavior of their prospective partners. To determine the regulatory mechanisms governing reproduction in the western tarnished plant bug, *Lygus hesperus*, the mating behavior of both sexes was examined. Hour-long pairings of potential mates were conducted inside covered glass Petri dishes. To determine receptivity to mating, individuals were used that varied in age from the newly emerged to one week after emergence. It was found that males were ready to mate just three days after emergence while females were receptive after five days. Males were only attracted to females that were at least three days old, and when given a choice preferred those that were at least five days old. Males also preferred females that were unmated

over those that had mated within the previous 24 hours. Males appear to be assessing female acceptability based on chemical cues collected by antennation, although additional experimentation is necessary to confirm this.

### 4. **Standard sampling plan for *Varroa destructor*.** *Katie Lee, Marla Spivak, Eric Burkness, Roger Moon*, University of Minnesota, St. Paul, MN.

The parasitic mite *Varroa destructor* is a significant pest of the honey bee, *Apis mellifera*, being the major cause of colony death in the US. To keep colonies alive, beekeepers often treat all of their colonies once or twice a year, irrespective of mite level. The aim of my research is to create a standard and efficient method to quantify colony and apiary levels of *V. destructor* to help beekeepers make educated treatment decisions. My long term goal is to encourage beekeepers to reduce the amount of in-hive use of chemicals to prevent evolution of miticide resistance and contamination of honey and wax. I sampled 30 apiaries, each containing 22-57 colonies, to determine how the mites are distributed within and among colonies and apiaries. A nested analysis of variance showed the largest source of variation in mite infestation level was among colonies. Taylor's Power Law and Green's Plan were used to determine the optimal sample size. Results show that 210 bees should be sampled per colony from one comb containing brood, and samples from eight colonies within an apiary will give an accurate estimate of mite level within an apiary.

### 5. **The biogeography of sex in the facultative thelytokous ant *Platythyrea punctata*.** *Katrin Kellner, Jon N. Seal, Jürgen Heinze*; University of Regensburg, Regensburg, Germany.

Of all the species of social Hymenoptera, only six ants and the Cape Honeybee are capable of thelytokous parthenogenesis (the production of diploid females from unfertilized eggs). One of these ants is the ponerine ant, *Platythyrea punctata*. This species occurs in the neotropics from South Texas to at least Costa Rica on the American mainland and on nearly all Caribbean islands, from south Florida to Grenada. Previous work suggested that colonies from Puerto Rico, Barbados and Florida are predominantly thelytokous, whereas colonies from Costa Rica produced sexual offspring from mated workers ("gamergates"). Since then we have increased our sample size by collecting colonies from eleven other islands and mainland sites ( $N > 200$  colonies). By performing analyses with five highly polymorphic microsatellite loci, we investigate the occurrence of sexual and thelytokous reproduction over a large portion of the range of *P. punctata* and the resulting population structures. We also have a preliminary phylogeny based on a 1450kb fragment of the COI gene. It appears that thelytoky evolved once and some of the mainland populations are sexual but some may have mixed strategies.

### 6. **The effect of juvenile hormone on temporal polyethism in the paper wasp *Polistes dominulus*.** *John R. Shorter*, Purdue University, West Lafayette, IN.

Juvenile hormone (JH) has an important role in the behavior of eusocial Hymenoptera. Previous work has shown that JH influences aggression and dominance behavior in primitive eusocial insects that lack discrete queen and worker castes (e.g. *Bombus* bees and *Polistes* wasps). In contrast, JH is one of the factors that mediates temporal polyethism among workers in advanced eusocial insects that have reproductive castes (e.g. *Apis* bees and *Polybia* wasps). Therefore, initial observations suggest that JH may have different roles in primitive and advanced eusocial taxa. Here, we use detailed behavioral observations of marked individuals to test whether JH influences temporal polyethism in the primitive eusocial

wasp *Polistes dominulus*. First, we show that workers in *P. dominulus* have an age-related division of labor, as workers switch from nest work to foraging as they mature. Then, we show that application of JH accelerates the onset of foraging behavior. Workers treated with JH start foraging at a younger age than control workers. Therefore, JH mediates temporal polyethism in the primitively eusocial insect *Polistes dominulus*.

7. **Does *Varroa jacobsoni* reproduce on worker brood of *Apis cerana* in India?** Jessica Burtness (1), Raghavendra Gadakgar (2), Marla Spivak (1); (1) University of Minnesota, St. Paul, MN; (2) Bangalore University, Bangalore, India.

A parasitic mite (*Varroa destructor*) is one of the most serious pests currently infesting honey bee colonies worldwide. The original host of the mite is the Asian honey bee (*Apis cerana*), which is able to tolerate mite infestation without any intervention. One of the mechanisms by which *Apis cerana* keeps the mite populations low is by suppressing mite reproduction in worker brood. To determine whether *Apis cerana* uses hygienic behavior to remove reproducing mites from worker brood, worker pupae were artificially infested with mites. After a period of nine days, the infested worker cells were checked for removal of the infested pupae, removal of the introduced mites, and reproduction of remaining mites. No mites introduced onto worker pupae reproduced.

8. **Element flow through colonies of the desert leafcutter ant *Acromyrmex versicolor*.** Rebecca Clark, Kimberly Shaffer, Jennifer Fewell; Arizona State University, Tempe, AZ.

Agriculture, the act of growing one's own food, has evolved only a handful of times in social organisms, including in fungus-growing ants. We do not yet understand how the evolution of the ant-fungus mutualism affects trophic dynamics within leafcutter colonies. One method for understanding trophic dynamics involves comparing stoichiometric relationships (relative elemental composition) between organisms, their food sources, and their waste. Three key elements, Carbon (C), Nitrogen (N), and Phosphorus (P), regulate many organismal and ecological processes, and N and P are found in much higher relative concentrations in insects compared to plants. To determine if the leafcutter fungus improves relative N and P availability, we measured the relative elemental compositions of: workers of the desert leafcutter ant *Acromyrmex versicolor*; the ants' fungus; and leaves of one of the ants' major food sources (Palo brea, a desert tree species). Relative N and P concentrations were two times higher in ants than in both leaves and fungus. Thus, in contrast with expectations, the leafcutter ant fungus does not appear to concentrate N and P, and further research is needed to understand how leafcutter ants obtain adequate quantities of these resources. An examination of colony refuse materials should provide further insight.

9. **Gene expression and phenotype association in social wasps.** Brendan G. Hunt, Soojin V. Yi, Michael A.D. Goodisman; Georgia Institute of Technology

Social insects are well suited for the study of mechanisms by which genotype and environment mediate behavioral and physiological phenotypes through differential gene regulation. The usefulness of social insects stems from the fact that individuals belong to distinct castes which undertake different behavioral tasks. Caste-differentiation is environmentally triggered and linked to variation in gene expression, resulting in discrete adult phenotypes. In this investigation, we determine whether gene expression patterns are conserved across social insect species. We present comparative gene expression data for four eusocial Vespidae wasps, including three species that exhibit clearly dimorphic queen and worker castes and

one species that does not exhibit morphological castes. Using quantitative real-time PCR, we address gene expression patterns between adults of different castes and sexes. Gene-specific primers which amplify across species were developed using sequences generated from the wasp *Vespula squamosa*. Several genes were found to be differentially expressed between castes and sexes of multiple species, indicating a conserved relationship with phenotypic class. Many other genes exhibit divergent expression patterns across taxa with respect to sex and caste. Overall, our research helps us to understand how variation in gene expression is associated with phenotypic differences.

10. **Ant engineering: nest building challenges in variable granular media.** Laura Levy, Michael Goodisman, Daniel Goldman; Georgia Institute of Technology, Atlanta, GA.

Almost all social insects construct multi-functional nests that provide refuge for young, storage for food and defenses against the elements. Over the colony's lifespan, continual nest modifications accommodate growing numbers and resist general wear. Most species of ants dwell in subterranean nests which require significant effort for excavation, maintenance and defense. The morphology of these underground systems is highly influenced by the physical materials available for construction. We are interested in understanding variable nest phenotypes as they pertain to the different mechanical properties of granular media. Using the red imported fire ant (*Solenopsis invicta*) as a model to study nest building behavior, we will alter particle size and moisture content to determine how these factors jointly affect nest construction. Abundant moisture results in higher adhesion forces between particles that allows for stronger tunnel walls. Large sized particles often have higher percolation rates that drain water quickly from the soil creating a difficult building environment. Using two-dimensional ant farms, queen-right colonies will excavate nests in 50, 300 and 700 micron glass beads with varying moisture levels. These experiments ultimately will examine optimal building conditions in regards to moisture content and media size, providing insight into how nest infrastructure affects inhabitant viability.

11. **Social apoptosis in the super-organism?** Miranda Hayworth, Nathan Ross, Olav Rueppell; University of North Carolina-Greensboro, Greensboro, NC.

Cellular apoptosis is crucial for multicellular organisms in development and homeostasis by allowing organisms to safely dispose of malfunctioning or surplus cells. Social insect colonies are highly integrated units that can be compared to a super-organism and workers readily kill themselves to fulfill an essential colony function (e.g. defense). However, it is unknown whether workers kill themselves to prevent damage to the colony caused by their presence, analogous to apoptosis. The purpose of this study was to theoretically analyze biological conditions under which such behavior should occur and evaluate the theoretical predictions with an empirical test. We focused on sick worker that could harm their colony by spreading disease. Theoretical modeling predicts social apoptosis under a wide range of parameters in this case, i.e. the workers that perceive serious illness should leave their hive immediately to prevent spreading the disease. We tested this prediction by comparing multiple behavioral variables between control workers and workers of two artificially sickened treatment groups. The first group was treated with Hydroxyurea and the second one was anaesthetized with carbon dioxide for two hours. Both treatments result in significant mortality and may simulate serious illness. The results will be discussed in light of social evolution.

12. **Patterns of expression and abundance of Gp-9, a protein linked to social form in the fire ant, *Solenopsis invicta*.** Emily Matthews, University of Georgia, Athens, GA.

Social form in fire ants (*Solenopsis invicta*) appears to be regulated by genotype at the *Gp-9* gene or linked genes. When all workers are

*BB* at *Gp-9*, colonies accept a single *BB* queen (monogyny); in contrast, colonies with *b*-bearing workers accept multiple *b*-bearing queens (polygyny) but reject any queens without the *b* allele. However, the function of *Gp-9* and its specific role in social organization remain unknown. *Gp-9* gene expression and protein abundance were examined across life stages and castes, and *Gp-9* protein localization was further studied within individual ants. *Gp-9* was absent in all larval and pupal stages and present in all adults, with a much higher concentration in workers and female sexuals than in males. Interestingly, *Gp-9* was present in eggs but not produced by the eggs themselves. The protein occurred throughout the adult worker body and was also present externally on the cuticle. This last finding suggests that *Gp-9* could potentially play a direct role in communication, and these results provide a direction for further study to determine *Gp-9*'s role in the maintenance of social form.

13. **Africanized and European honey bee pollen foraging thresholds.** *Dina Leslie Grayson* and Jennifer H. Fewell, Arizona State University, Tempe, AZ.

Variation in foraging strategy has been linked to the success of some invasive organisms. However, we know little about how invasive animals achieve these shifts in foraging behaviors. The Africanized honey bee (AHB) provides an ideal system to examine these questions, for the highly invasive AHB has out-competed closely related European honey bees (EHB) in areas with warm climates across the Americas. Evidence suggests AHB's success could be related to their higher colony-level pollen intake. Yet, the specific individual-level mechanisms that lead to these colony differences remain unclear. This study tests the hypothesis that AHB foragers have a lower shifted distribution of response thresholds to pollen foraging stimuli. To evaluate this hypothesis we observed the foraging behavior of co-fostered individually marked AHB and EHB in response to graded changes in pollen stores. If AHB are more sensitive to stimuli for pollen foraging, then they should increase pollen foraging at higher pollen storage levels (lower pollen foraging stimulus level) than EHB. Surveys of foragers indicated that the number of AHB foragers collecting pollen was higher across pollen storage treatments, indicating a higher intrinsic propensity to pollen forage in AHB workers. As we gradually increased the stimulus for pollen foraging, AHB tended to increase their pollen foraging effort earlier (at a lower stimulus level) than EHB, as predicted by the hypothesis that AHB have a lower-shifted distribution of pollen foraging response thresholds.

14. **Putative native source of the invasive fire ant *Solenopsis invicta* in the USA.** *David DeWayne Shoemaker*, USDA-ARS, Center for Medical, Agricultural and Veterinary Entomology, Gainesville, FL.

The ecological and evolutionary dynamics of newly introduced invasive species can best be understood by identifying the source population(s) from which they originated, as many species vary behaviorally, morphologically, and genetically across their native landscapes. We attempt to identify the source(s) of the red imported fire ant (*Solenopsis invicta*) in the southern U.S.A. utilizing data from three classes of genetic markers (allozymes, microsatellites, and mitochondrial DNA sequences) and employing Bayesian clustering simulations, assignment and exclusion tests, and phylogenetic and population genetic analyses. We conclude that the Mesopotamia flood plain near Formosa, Argentina represents the most probable source region for introduced *S. invicta* among the ten localities sampled across the native South American range. This result confirms previous suspicions that the source population resides in northern Argentina, while adding further doubts to earlier claims that the Pantanal region of Brazil is the source area. Although finer-scale sampling of northern Argentina and Paraguay combined with the use of additional genetic markers will be necessary to provide a highly

precise source population assignment, our current results are of immediate use in directing future sampling and focusing ongoing biological control efforts.

15. **Lack of scent-marking of food sources in the swarm-founding wasp, *Polybia occidentalis*.** *Benjamin J. Taylor*, Robert L. Jeanne; University of Wisconsin-Madison, Madison, WI.

Food-site marking, either repellent or attractive, is widespread in the social bees, and is expected to enhance foraging efficiency. In contrast, other than one species of wasp, *Vespa mandarinia*, there has been no evidence for scent-marking of food sources in the social wasps. We tested for food site marking in the swarm-founding wasp, *Polybia occidentalis*, which chemically marks sites during colony emigration. Foragers from 11 nests were given a choice between a previously visited feeder containing a rich sucrose solution and an unvisited one. There was no difference between the number of visits to the visited feeder and an unvisited control. We conclude that foragers of this species of swarm-founding wasp do not deposit scent marks, either attractive or repellent, on food sources.

16. **Genetic caste determination in harvester ants: If you are going to mate with many males, does it matter who they are?** *Brendon Mott*, Arizona State University, Tempe, AZ.

The majority of ants rely on environmental caste determination (ECD), but many populations of the seed-harvester ant *Pogonomyrmex barbatus* rely on a system of strict genetic caste determination (GCD) resulting from the interbreeding of two distinct yet interdependent lineages. Same-lineage fertilizations result in queens while alternate-lineage fertilizations produce workers. In these dependent lineage populations, costs at colony founding and reproduction can be predicted from the relative frequency of each lineage and the mating frequency of the queens. Rare lineage queens are expected to mate more often, increasing their probability of acquiring intralinear sperm for new queen production. Common lineage queens are expected to limit their mating to decrease costs from excess queen production during periods of colony growth. Five microsatellite loci were used to compare queen mating frequency and caste-specific patriline diversity in two GCD lineages as well as a separate ECD population. Our results suggest that queens in mature colonies mated randomly, and the lineage ratios of sperm acquired by queens during mating do not predict survival to maturity. Thus the predictions of previous studies are not supported; queens of the common lineage are not less likely to survive to maturity due to an excess of queen destined sperm.

17. **Dominance hierarchy and nutrient acquisition in the slave-making ant *Protomognathus americanus*.** *Jason Carbaugh*, Timothy M. Judd; Southeast Missouri State University, Cape Girardeau, MO.

A reproductive dominance hierarchy exists among the workers of the slave-maker ant *Protomognathus americanus*. The hierarchy can be observed by the interactions among the slave-maker workers, how the workers interact with the slave-maker queen, and how the slaves interact with the slave-maker workers. One interaction is that has been previously observed was that the dominant workers receive more food from the slaves than the subordinates. It has also been observed that the dominant workers will interrupt the transfer of food by trophallaxis between subordinates and slaves. This study determined the relationship between the amount of macronutrients obtained by a slave-maker worker and their rank in the dominance hierarchy. Slave maker colonies were collected from the field and housed in artificial arenas. The dominance hierarchy was determined for the slave-maker workers for each colony. Once the hierarchy was determined, the individuals were analyzed to see if the more

dominant workers had higher levels of protein, carbohydrates and lipids than their subordinates.

18. **Differences in sugar preferences in several populations of the termite *Reticulitermes flavipes* during different seasons.** Bruce A. Wallace, Timothy M. Judd; Southeast Missouri State University, Cape Girardeau, MO.

Studies have shown that the termite *Reticulitermes flavipes* prefer food with certain types of sugars. However, in each study the types of sugars that are preferred by the termites differ. The differences between the results of each of these studies might be explained by differences in populations or changes in feeding preferences during the active season. To address these variables, we examined the preference for food containing glucose, sucrose, or xylose versus a food source without sugar in several populations of termites and if these preferences changed during the year. Termites were collected from colonies from four field sites in Missouri during the spring (May and June), summer (July and August) and fall (September and October) and tested for their preference for all three sugars under laboratory conditions. Results show there are distinct differences in sugar preferences between populations.

19. **Worker and weapon size and nest defense in *Solenopsis invicta*.** Kevin Haight; Arizona State University, Tempe, AZ.

Workers of the fire ant, *Solenopsis invicta*, come in a continuous range of sizes. Such size variation is thought to enable a more efficient division of labor, as some worker sizes may be better suited to performing particular tasks than others. Here I focus on the task of nest defense, where efficiency and effectiveness may depend on having appropriately-sized workers respond to particular types of threats. Specifically, I test whether the size of responding workers differs between two magnitudes of nest disturbance (light, insect-like vs. heavy, vertebrate-like), and I investigate how lancet length, a key defensive trait, varies with worker size. Workers responding to heavy, vertebrate-like nest disturbances were larger than those responding to light, insect-like ones. Between the light and heavy disturbances the proportion of minor workers responding dropped, while that of majors increased five-fold. Though the relationship between lancet length and worker size is not directly proportional, the average lancet length of the average major is ~ 0.20mm greater than that of the average minor, and there is little overlap in range. So, vertebrate-magnitude nest disturbances are met with greater proportions of large *S. invicta* workers than are insect-magnitude disturbances, and these larger workers bring with them longer lancets which may increase their relative effectiveness against such thicker-skinned threats.

20. **Locating and characterizing drone congregation areas (DCA) of *Apis mellifera* in Puerto Rico.** Carlos J. Rivera Rivera, Carlos M. Huerta Done, Laura Caicedo Quiroga, Rafiné Moreno-Jackson, Alberto Galindo Cardona, Tugrul Giray; University of Puerto Rico, San Juan, PR.

DCAs are zones that drones and queens frequent for mating. DCAs are thought to have certain geographical characteristics preferred by bees. We raised a helium-filled balloon carrying a wire with a lure (black material with queen bee pheromones) attached to it, in order to attract drones flying close by. A DCA was identified when large densities of drones were attracted to the pheromone lure. We marked the coordinates, and described the geographical characteristics of the area. Based on previous descriptions of DCAs we have examined 16 localities, finding a DCA in half of these localities. Through further study of these 8 locations we will define the general characteristics of a DCA, and we will predict new DCAs over large areas using a geographical analysis software. Reliably predicting DCAs will be an

invaluable tool to study honeybee mating behavior, hybridization, and colony health.

21. **Homing in male honey bees (drones): If I don't come back, don't wait for me.** Rafine Moreno Jackson, Carlos M. Huerta Dones, Alberto Galindo Cardona; University of Puerto Rico, San Juan, PR.

The orientation of the male drones of honey bees (*Apis mellifera* L.) has not been studied much. A worker bee has a high probability of disorientation if it gets farther than 2.5 Km from its hive. It has been reported that some drones can travel more than 7 Km but only for reproductive means. It is important to study the way populations act to understand the spatial behavior of organisms. The purpose of this study is to measure the return rate and orientation of drones released at different distances from their hives (1, 2 and 4 Km.), from the four cardinal points (from the apiary). We captured and marked 720 drones (120 per distance per cardinal point) and we observed the number of drones that returned to their hives after being released. It was expected that more drones would return from the closer releasing sites but did not expect to find differences between the orientations of the releasing sites. The results show that there is a difference on the time it took the drones to return to their hives from different distances and from the different cardinal points. There could exist known routes for the drones that make it easier to return faster to their hives.

22. **Aggressive behavior of fire ants (*Solenopsis invicta*) in Puerto Rico.** Vilmarie Figueroa-Nieves and Bert Rivera-Marchand; Inter American University of Puerto Rico, Bayamón Campus \*vilmastephanie@hotmail.com  
*Solenopsis invicta*, known as fire ants, arrived to Puerto Rico in the 1970's. This ant considered an invasive species, impacts biodiversity, economy and human health. One of its negative characteristics is aggressive behavior. The objective of our study was to evaluate the aggressive behavior of this ant on the island. As with Africanized bees on the island, we expected reduced aggressive behavior in these ants. We first classified the nests of origin of each sample as monogyne or polygyne using a PCR based assay. Then we evaluated intraspecific aggressiveness by observing the interactions of ants from different nests placed in an arena. Observed interactions between ants from the same nest were considered a negative control while interactions between *S. invicta* and *S. geminata* were considered positive controls. Assays with all possible combinations of monogyne and polygyne samples were considered experimental groups. There was no significant difference between the negative control and experimental groups. There were, however significantly high levels of aggressive behavior between the positive control and experimental groups. The results of the experiments suggest that the intraspecific aggressive behavior of the studied fire ants is low. These results are similar to studies of invasive Argentine ants in California which were also found to be relatively unaggressive. This study supports the hypothesis that island species tend to be less aggressive than continental ones and extends our knowledge in conservation as well as evolution on islands.

23. **Division of labor in the little fire ant (*Wasmannia auropunctata*).** Rafael Fernández-Casas, Bert Rivera-Marchand; Inter American University of Puerto Rico, Bayamón, PR.

Division of labor in ants may be related to different morphologies or differences in ages of the worker castes. Division of labor in

monomorphic ants is known to be age related. Younger workers perform tasks inside the colony and older workers perform tasks outside. Behavioral plasticity within division of labor has been found in some species of ant, as well as honey bees, where older individuals can perform tasks of younger individuals and younger workers can precociously perform tasks of the older. We performed two experiments to determine division of labor and behavioral plasticity on the little fire ant *Wasmannia auropunctata* an invasive species from the tropics. Although the little fire ant is considered monomorphic we found differences in coloration, where lighter colored ants performed nursing tasks and darker ants foraged. Prior to the all experiments we will determine the age at which different behavior occur by painting the ants. In the first experiment we removed foragers to determine if the younger ants would forage precociously. In the second experiment we removed foragers to determine if they would revert to nursing duties. After removing foragers the nurses did not forage precociously, instead they cannibalized larvae and pupae. After removal of nurses the foragers assumed the role of nurses, forgoing foraging. The seemingly lack of precocious foraging in these ants may be an important adaptation to tropics. In the tropics large and intermediate disturbances are frequent. A nest that loses its foragers due to a stochastic event decreases the risk of losing all workers by retaining them within the nest.

24. **Behavioral plasticity of queens in the little fire ant (*Wasmannia auropunctata*).** Yarira Ortiz-Alvarado, Bert Rivera-Marchand; InterAmerican University of Puerto Rico, Bayamon, PR.

One of the most important traits of social insects, such as ants, is reproductive division of labor, where queens lay eggs and sterile worker castes perform nest duties including brood care and foraging. Many species of social hymenoptera have plasticity in division of labor. This plasticity is typical of the sterile worker class yet rare in queens. The little fire ant, a monomorphic species with age related division of labor, has recently been found to have plasticity of worker division of labor where older ants, that typically forage, return to the nest to perform nursing duties when these are absent. We evaluated behavioral plasticity of queens the little fire ant *Wasmannia auropunctata*. When we removed nurses from the colony queens performed nursing tasks including brood tending and nest cleaning. The behavior of queens performing worker duties is rare in ants yet common in primitively social hymenoptera such as paper wasps. This may be an example of convergent evolution where the queens of these ants, similar to the wasps, have behavioral adaptations important in survival of this invasive species.

25. **Methoprene influences wing beat frequency increase and troponin T expression of honey bees with accelerated development of foraging behavior.** Daliris Ramirez Burgos(1), Devrim Oskay(2), Tugrul Giray(1); (1) University of Puerto Rico, , San Juan, PR; (2) Washington State University, Pullman, WA.

Adult honey bees show a stereotypical behavior development that underlies colony division of labor. This temporal polyethism has regulatory correlates, including the Juvenile Hormone (JH). It has been shown that workers show higher levels of this hormone at onset of foraging and hormone analog application leads to precocious foraging. As the honey bee moves from non-flying tasks in the hive to the energetically costly task of foraging, their physiology changes as well. We hypothesize that JH is associated to muscle development. Our prediction is that in a group of bees treated with JHA, changes in contractile protein isoforms will occur sooner than in the control group. Bees treated with JHA started foraging two days sooner than the control group (6 and 8 days, respectively) and Wing Beat

Frequencies increased significantly by age in JHA group of bees. Moreover, preliminary analysis of flight muscle motor proteins by SDS-PAGE and Western Blots show that control individuals of age 3 have one band for TnT, whereas one of the JHA group has two, resembling older bees. Further assays should be performed to analyze this differences.