Apidologie 38 (2007) 482–504 © INRA/DIB-AGIB/ EDP Sciences, 2007 DOI: 10.1051/apido:2007030 Available online at: www.apidologie.org

Association of Institutes for Bee Research Report of the 54th seminar in Veitshöchheim 27–29 March 2007

Arbeitsgemeinschaft der Institute für Bienenforschung e.V. 54. Jahrestagung in Veitshöchheim vom 27.–29. März 2007

Association des Instituts de Recherche sur les abeilles Comptes rendus du 54^e congrès à Veitshöchheim 27–29 mars 2007

List of reports (* after the title indicates that no abstract of this report is published).

Verzeichnis der Referate (* bedeutet, dass zu diesem Titel keine Zusammenfassung aufgeführt ist).

Liste des communications (* après le titre indique que le résumé de la communication n'est pas publié dans ce numéro).

Invited talk Einführungsvortrag Conférence inaugurale

1. Microsporidia infections in Hymenopteran pollinators. *I. Fries**

Infektionen mit Mikrosporidien bei Bestäubern der Hymenopteren.

Infections à microsporidies chez les Hyménoptères pollinisateurs.

Praktische Imkerei/Monitoring

Bee management/Monitoring

Pratique apicole/Monitoring

2. Analysis of a mating season in the mountainous mating station "Torfhaus", Harz. O. Boecking*

Begleitende Analyse einer Paarungssaison auf der Gebirgsbelegstelle "Torfhaus" im Harz.

Analyse d'une saison d'accouplements à la station de fécondation d'altitude de Torfhaus dans le Harz.

3. Influence of location, colony management, climate and disease load on overwintering of honeybee colonies. *G. Liebig*, *A. Schroeder*, *E. Frey*, *J. Kiefer** Einfluss von Standort, Völkerführung, Witterung und Krankheitsbelastung auf die Überwinterung von Bienenvölkern.

Influence du lieu, de la gestion des colonies, du climat et de charge de maladie sur l'hivernage des colonies.

4. Results of the second year of "DEBIMO" (German Bee Monitoring) for insights in periodical incidences of bee colony losses. *AG Bieneninstitute Hohenheim**

Ergebnisse des zweiten Untersuchungsjahres im "DEBIMO" (Deutsches Bienenmonitoring) zur Aufklärung periodisch auftretender Völkerverluste.

Résultats de la deuxième année de "DEBIMO" (monitoring allemand des abeilles) pour expliquer les pertes périodiques de colonies.

5. The hive record project 2006-2007, an evaluation model for beekeeper practice. *R. van der Zee**

Das Völkerbeobachtungsprojekt 2006-2007, ein Modell zur Beurteilung von Bienenhaltungspraktiken.

Projet d'observation des colonies 2006-2007 : un modèle d'évaluation pour la pratique apicole.

6. Drone brood pressing for wax production. *K*. *Wallner**

Pressen von Drohnenbrut zur Wachsgewinnung.

Presser le couvain de mâles pour obtenir de la cire.

7. How can a beekeeper influence the quality of honey harvested from *Apis mellifera* colonies? *J. Radtke, E. Etzold*

Wie kann der Imker die Honigqualität beein-flussen?

Comment l'apiculteur peut-il influencer la qualité du miel de ses colonies ?

Article published by <u>EDP Sciences</u> and available at <u>http://www.apidologie.org</u> or <u>http://dx.doi.org/10.1051/apido:2007030</u>

8. Effects of colony splitting on population development and health status of honey bee colonies. *I. Illies, R. Siede, R. Büchler*

Effekte der Jungvolkbildung auf die Entwicklung und Krankheitsbelastung von Bienenvölkern.

Effets de la division des colonies sur le développement et l'état sanitaire des colonies d'abeilles.

9. Analysis of beekeeping practice in Syria for the development of a country wide breeding program. *K. Zakour*, *K. Bienefeld**

Analyse der Bienenhaltung in Syrien als Grundlage für die Entwicklung eines landesweiten Zuchtprogramms.

Analyse de la pratique apicole en Syrie pour mettre au point un programme de sélection à l'échelle nationale.

Physiologie, Verhalten

Physiology, behaviour

Physiologie, comportement

10. Analysis of individual brood temperatures and consequences for the lifespan of honeybee workers. *M. Becher*, *R.F.A. Moritz**

Analyse der Temperaturen individueller Honigbienenbrut und Auswirkungen auf die Lebensspanne der Arbeiterinnen.

Analyse des températures individuelles du couvain et conséquences pour la durée de vie des ouvrières d'abeilles.

11. The effect of group size on the mandibular gland secretions of honeybee workers. *H. Scharpenberg*, *R.F.A. Moritz**

Einfluss der Gruppengrösse auf die Sekretionen der Mandibeldrüsen bei Honigbienenarbeiterinnen.

Influence de la taille du groupe sur les secrétions des glandes mandibulaires des ouvrières d'abeilles.

12. The role of ecdysteroid hormones in young worker honeybees. J. Wegener, Z. Huang, K. Bienefeld

Zur Rolle von Ecdysteroid-Hormonen bei jungen Arbeiterinnen der Honigbiene (*Apis mellifera*).

Rôle des hormones ecdystéroïdes chez les jeunes ouvrières d'abeilles.

13. Specialised behaviours in brood incubation of honeybees (*Apis mellifera carnica*). *M. Kleinhenz*, *B. Bujok*, *S. Fuchs*, *J. Tautz*

Spezialisierte Verhaltensweisen bei der Regulierung der Brutnesttemperatur der Honigbiene *Apis mellifera*.

Comportements spécialisés lors de l'incubation du couvain d'abeilles (*Apis mellifera carnica*).

14. Potential of digital image processing for the observation of honeybee behaviour. U. Knauer, K. Bienefeld, B. Meffert

Möglichkeiten der Bildverarbeitung für Verhaltensbeobachtungen von Bienenvölkern.

Les possibilités du traitement d'images pour l'observation du comportement de l'Abeille domestique.

15. Larval movement and feeding behaviour of worker bees in *Apis mellifera*. *C. Heimken*, *W.H. Kirchner*

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16. Liquid nutrition within a honeybee colony – who feeds? R. Brodschneider, N. Hrassnigg, J. Vollmann, M. Petz, U. Riessberger-Gallé, K. Crailsheim

Einfütterung flüssiger Nahrung in ein Bienenvolk – wer trinkt?

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17. Solid nutrition within a honeybee colony – who feeds? U. Riessberger-Gallé, R. Brodschneider, J. Vollmann, R. Thenius, K. Uhl, S. Krainer, H. Kovac, K. Crailsheim

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19. High-power fuel for heavy duty workers – resource management in the bee hive. *R. Basile*, *CH.W.W. Pirk*, *J. Tautz**

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Du combustible haute énergie pour les ouvrières qui travaillent dur – gestion des ressources dans la ruche.

20. Temperature budgeting in a bee hive in a physically based model. S. Maier, H. Vierle, J. Tautz, J.A.C. Humphrey*

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Maintien de la température d'une colonie d'abeilles selon un modèle basé sur la physique.

21. Attempts to establish cell lines from the honey bee. R. Siede, M. König, R. Büchler, H.-J. Thiel

Versuche zur Etablierung einer Honigbienen-Zelllinie.

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22. Influence of prenatal provisioning on the properties of adult honeybee queens. *S. Al-Kahtani*, *K. Bienefeld**

Einfluss unterschiedlicher pränataler Versorgung auf Eigenschaften adulter Bienenköniginnen.

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26. Introgression of commercial bumbless (*Bombus terrestris*) into feral populations in Poland. *S.M. Rohde, M. Woyciechowski, R.F.A. Moritz**

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Introgression des bourdons (*Bombus terrestris*) du commerce dans les populations sauvages en Pologne. 27. Hans Staden – first record on Brazilian stingless bees 450 years ago. *W. Engels**

Hans Staden – erste Schilderung der Stachellosen Bienen Brasiliens vor 450 Jahren.

Hans Staden – première description des abeilles sans aiguillon du Brésil il y a 450 ans.

28. Banana Wilt in East Africa: are stingless bees vectors of *Xanthomonas capestris musearum*? *F. Namu, D. Wittmann**

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29. Amplified fragment length polymorphism analysis on the genetic caste determination in a stingless bee (*Melipona quadrifasciata*). G.R. Makert, R.J. Paxton, K. Hartfelder

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36. A bee repellent endangers the quality of bee products. A. Schroeder, B. Fritz, D. Weber, K. Wallner

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40. Investigations of Shear, Oszillations and DSC in Honey. J. Smanalieva, B. Lichtenberg-Kraag, B. Senge*

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41. Strategies to reduce the input of pesticides in honey bee colonies demonstrated for the fungicide Cantus[®]. *I. Illies, K. Wallner, R. Büchler*

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42. Clearcutting: *Epilobium angustifolium* L. as a temporary resource for wild bees and other flower visitors. *J. Kühn, A. Hamm, D. Wittmann*

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47. Use of alternative pollen sources by *Colletes* hederae. R.C. Peruquetti, D. Wittmann

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55. Comparison of two sampling techniques for assessing the density of honeybee colonies in the field. *R.F.A. Moritz*, *V. Dietemann*, *R.M. Crewe**

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59. Molecular analysis of the candidate gene regulating parthenogenesis in honeybee workers. *E. Stolle, R.M. Crewe, R.F.A. Moritz**

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60. Differential expression of grainy head: a regulatory gene linked to the honeybee caste system. *A. Jarosch, R.F.A. Moritz**

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73. Radiofrequency – identification tags for measuring the reaction of foragers to the infestation with *Varroa destructor* in different honey bee lines. *Ch. Schneider, I. Illies, J. Tautz, S. Fuchs* *

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Ambiance refroidie dans le nid à couvain – recherches sur le comportement hygiénique d'*Apis mellifera*.

86. First report of *Nosema ceranae* in colonies of *Apis mellifera* in Austria. *I. Loncaric, I. Derakhshifar, H. Köglberger, R. Moosbeckhofer, R. Martin, M. Higes, A. Meana*

Ein erster Bericht über Nosema ceranae in Völkern von Apis mellifera in Österreich.

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87. Influence of virus infections on the overwintering abilities of honeybee colonies. *C. Garrido**

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Abstracts

7. How can a beekeeper influence the quality of honey harvested from *Apis mellifera* colonies? J. Radtke, E. Etzold (Länderinstitut für Bienenkunde, Friedrich-Engels-Str. 32, 16540 Hohen Neuendorf, Germany)

The water content of honey is a decisive parameter of quality. However, the 18% threshold indicative of especially high quality honeys is frequently exceeded. The conditions under which analysed honey samples had been produced were studied by means of a written questionnaire for beekeepers. The water content of honey harvested from rear-worked hives was significantly lower than that of honey from magazine hives (16.32 + 0.09%); n = 123 and 17.23 + 0.12%; n = 85, respectively; t-Test). No difference was discernable in the activity of the enzyme invertase. The average activity values of the honey from rear-worked hives was 110.88 + $3.92 U_{Sieg}/kg$ and was $113.83 + 4.31 U_{Sieg}/kg$ from magazine hives. A noteworthy influence was exerted by the comb material used in the honey supers: if the magazine hive was stocked exclusively with virgin wax in which the bees had never raised brood in, the water content was significantly higher (17.92 + 0.24%, n = 24) than if stocked with a combination of virgin and brooded (dark) comb, especially if these combs still contained brood at the start of the nectar flow (16.65 \pm 0.18%, n = 24). The difference was the same both in magazine hives made of wood (17.91 \pm 0.41%, n = 12 and 16.70 + 0.39%, n = 9, respectively) and polystyrene (17.76 + 0.16%, n = 8 and 16.71 + 0.12%, n = 13,respectively). The cause for this difference is believed to be a lower density of bee coverage on unbrooded comb, as demonstrated by population estimates in approximately equally strong colonies: while as many bees were found to cover brooded combs of a well-filled honey super, as was the case in the top brood chamber of a three story colony of a magazine hive, only half as many bees were found to cover unbrooded frames (P < 0.001; n = 16 resp. n = 18 Völker; t-Test). In contrast to magazine hives, in rear-worked hives the honey chamber is never filled solely with unbrooded frames.

8. Effects of colony splitting swarms on population development and health status of honey bee colonies. *I. Illies, Reinhold Siede, R. Büchler* (Landesbetrieb Landwirtschaft Hessen, Bieneninstitut Kirchhain, 35274 Kirchhain, Germany)

Nucleus colonies and artificial swarms are used to increase the number of honey bee colonies in apiaries. Colony regeneration this way simulates the natural colony reproduction by swarming. The new colonies are physically separated from the old, possibly pest- and pathogen loaded brood nest of the mother colony. The shook swarm method (i.e., one technique of producing an artificial swarm) has a disease preventing and sanitizing effect. It is used in the disease management of American foulbrood (AFB) and European foulbrood (EFB). Our study aims to test the effects of such manipulation practices on colony population dynamics, and on the number of Varroa destructor mites and secondary viral infestations. In July 2006 artificial swarms were generated from 12 honey bee colonies. Six swarms were shaken on to combs with unsealed brood (group A). Another six swarms were shaken on to fresh wax foundation (group B). The remaining brood from the swarm donating colonies was used to established six nucleus colonies (group C). Six untouched colonies were used as control (group D). Samples of adult workers were collected from the brood frames monthly from May till October. These samples were used to determine the level of parasitization by V. destructor and the load of Acute Bee Paralysis Virus (ABPV) and Sacbrood Virus (SBV). In November 2006 and March 2007 colony strength was estimated. In December, all colonies were treated with Oxuvar® against Varroa mites. The mite infection rate in October was significantly lower in A and B (A: 1.6 mites per 10 g bees. B: 0.9 mites per 10 g bees) than in the other two groups (C: 5.0 mites per 10 g bees, D: 5.7 mites per 10 g bees, P < 0.008 U-Test, Bonferroni corrected Plevel). Colony strength was significantly affected by the swarm simulating manipulations: The colonies in treatment groups A and B had 8000 individuals on average in November, while the mean of C and D was approx. 16000 bees (Bonferroni corrected *P*-level P < 0.008, U-Test). No winter fatalities occurred. The reduction of the colony strength over the winter months amounted up to 21% adult bee loss and was not significantly pronounced in a specific group (P > 0.05, U-Test). A real time PCR analysis revealed a general low ABPV load for all groups at all points of time. SBV was detected by end point PCR. The frequencies of positive SBV detection were high. Experiments with higher infected colonies are required to better elucidate the influence of colony splitting on viral infections.

12. The role of ecdysteroid hormones in young worker honeybees. J. Wegener¹, Z. Huang², K. Bienefeld¹ (¹ Länderinstitut für Bienenkunde, 16540 Hohen Neuendorf, Germany; ² Department

of Entomology, Michigan State University East Lansing, Michigan 48824, USA)

In many insects, ecdysteroids are involved in the regulation of reproduction. In Apis mellifera, this role seems to have been lost. In this species, pathways leading to egg formation and brood food production are linked by the protein vitellogenin, which is essential to both processes. We therefore examined whether ecdysteroids have switched to the regulation of brood food production in this species. In experiment 1, four queenright, broodless colonies with >34 days and 19 days old workers were established. Large amounts of brood were added to two of the colonies to induce a sudden rise in jelly production. Haemolymph samples of the younger workers were taken from all colonies before and after adding the brood. Ecdysteroid titers were measured by radio-immunoassay, and hypopharyngeal glands were rated. In a second experiment, eight groups of 40 ten-days-old workers each were caged. Twenty bees per cage were injected with 0.5 μg makisterone in 20% ethanol, the other half received the solvent alone. A third group of cages contained non-injected workers. Workers from all cages were frozen 0, 1 and 4 days post-injection, and hypopharyngeal glands dissected. The addition of brood in experiment 1 led to a strong increase in hypopharyngeal gland development, which was not accompanied by a change in ecdysteroid levels (before adding brood: $3.8 \pm$ 1.9 pg/ μ L; after adding brood: 3.8 ± 1.9 pg/ μ L; n = 56; P = 0.092; U-test). On all three days, there were positive, non-significant correlations between individual hormone titers and gland development (Rho = 0.03-0.22; n = 41-69; P = 0.06-0.77). In experiment 2, hormone injection led to an increase in Hypopharyngeal gland activation (n = 64, P = 0.008, Chi²-test). Some bees showed signs of hypertrophy of the glands. Our results indicate a possible role of ecdysteroids in jelly production. If confirmed, this would be another example of a hormone shifting to new functions as a result of eusociality.

13. Specialised behaviours in brood incubation of honeybees (*Apis mellifera carnica*) *M. Kleinhenz*¹, *B. Bujok*¹, *S. Fuchs*², *J. Tautz*¹ (¹ Beegroup, University of Würzburg, Am Hubland, 97074 Würzburg, Germany; ² Institut für Bienenkunde, Polyt. Ges., J.W. Goethe-Univ. Frankfurt a.M., Karl-von-Frisch-Weg 2, 61440 Oberursel, Germany)

In honeybee colonies, specialised brood incubation behaviour occurs on the surface of sealed brood cells by motionless worker bees which press their thorax onto the brood caps, thereby enhancing heat transfer by means of conduction. Additionally (and similar to social wasps), heat production occurs inside "gaps", i.e. inside open cells which are scattered at low rates throughout the sealed brood area. Eggs or young larvae may be found in these open cells but their presence is not essential to trigger heat production to warm the adjacent sealed brood. During cell visits by worker bees with elevated thorax temperature, a raise in the temperature of adjacent sealed brood cells is detectable by implanted temperature probes and by infrared thermographic imaging. On the level of single cells, heating inside open cells is highly efficient because the thorax is deep in the comb surrounded by up to six sealed brood cells, and virtually all heat released by the thorax reaches the brood. In contrast, bees on the comb surface may cover only one brood cap (or parts of three caps) and transfer heat to them only via their ventral side. Multi-agent based computer simulations were performed to investigate the efficiency of heat production inside open cells on the colony level. Gap proportions from 0 to 50% in the sealed brood area were tested. For brood gap proportions of 5 to 10%, which are common to healthy colonies, the average incubation time per brood cell is significantly reduced by 17 to 32% of the time which would be required for this task in a completely sealed brood area without any gaps. High proportions of brood gaps (>20%) which indicate a poor condition of the colony render brood nest incubation inefficient by increasing the incubation time per brood cell.

14. Potential of digital image processing for the observation of honeybee behaviour. U. Knauer, K. Bienefeld, B. Meffert (Humboldt-Universität zu Berlin, Institut für Informatik, 10099 Berlin; Institute of Bee Research, 16540 Hohen Neuendorf, Germany)

Advances in the processing capabilities of modest computing equipment allow real-time tracking even in a crowd of marked bees inside an observation hive. We developed a tracking procedure based on the extraction of the contours of markers placed on the bees. For the association step between the measurements in two consecutive images, the Euclidian distance and a similarity measure based on 2-D cross-correlation are used. The successful tracking allows accessing the paths of the bees in the field of view. However, the identification of individual bees as well as classification of their behaviour has to be performed automatically to support a long-term observation. In our experiments the identification was based on the geometrical shape, the orientation, and a number printed on the markers. Preliminary results show that the ASM technique is capable of identifying the correct shape of the markers. The best feature that can be used to estimate the orientation of the markers is the position of the eyes. Finally, the identification number can be extracted with OCR methods based on the orientation data.

One class of long-term experiments is the selection of hygienic bees. In a one-week trial of the experiment all bees which performed hygienic activities such as the opening of *Varroa* infested brood cells were selected. An automatic observation of the individual brood cells was used to record even small changes of the combs surface. These changes were reported to a human operator. Preliminary results show extensive savings in time due to this kind of computer-aided analysis of the video data. Future work will focus on the fusion of the tracking, identification, and change detection data.

15. Larval movement and feeding behaviour of worker bees in *Apis mellifera. Ch. Heimken, W.H. Kirchner* (Ruhr-Universität Bochum, Fakultät für Biologie)

Honeybee (Apis mellifera) larvae rotate within their cells in order to obtain the surrounding food. We investigated how the movement of fifth instar larvae is affected by their food supplies and by the inspection and feeding behaviour of worker bees. Speed of movement was compared between food deprived larvae and a control group. Behavioural responses to feedings were examined under natural conditions as well as to manual feedings in the laboratory. In addition, the inspection behaviour of worker bees was investigated with regard to larval movement. The speed of larval rotation $(182 \pm 86^{\circ})$ / 30 min) increased after one to two hours of food deprivation (velocity increase: $+85 \pm 31^{\circ}$ / 30 min; n = 5, P < 0.05 ref. < 0.001; U-Test; number of larvae per trial: 67 ± 15 (food deprived), 56 ± 15 (control)). After three to four hours the speed of rotation did not differ from that of controls (n =5), and after six hours (n = 2) the larvae moved slower than untreated ones $(-53 \pm 74^{\circ} / 30 \text{ min})$; significant only for one of the two trials). In connection with feeding visits by worker bees, larval speed increased for about three minutes (P < 0.05; U-Test). Larvae reacted on manual feedings with increased activity, too (P < 0.01). Larval speed within 30 minutes was positively correlated with the number of feedings (r = 0.49 / P < 0.001; Spearmans rank order correlation) and with the number of inspections by worker bees (r = 0.45 / P < 0.001). Within 10-minute-intervals without feeding (additionally, the last feeding was at least 10 minutes ago), there also was a positive correlation between larval speed and the number of inspection visits (r = 0.38 / P < 0.05). Larval behaviour changes dependent on larval condition and needs. Therefore it potentially could act as a part in the communication system between larvae and worker bees.

16. Liquid nutrition within a honeybee colony – who feeds? *R. Brodschneider, N. Hrassnigg, J. Vollmann, M. Petz, U. Riessberger-Gallé, K. Crailsheim* (Institut für Zoologie, Karl-Franzens-Universität Graz, 8010 Graz, Austria)

After harvesting honey in summer, or after winter when colonies are growing, it is often necessary to supply bees with additional carbohydrates. In this study we compared the age of bees consuming sugar syrup within the colony among the known temporal castes such as nurses, food storers and foragers. The age of bees feeding on syrup was investigated over different times of the day and the food was presented in different regions of the hive. Observations were made using an eight-frame observation hive with 10 000 bees (Apis mellifera carnica) during daytime and around midnight. About 5000 bees were individually marked to identify their age. Syrup (water:sucrose; 1:1) was presented to the bees in an open dish. Syrup was given ad libidum by injecting it into the dishes without opening the hive. Location of the food was changed daily between the honey stores and the dance floor. Every 15 minutes the age of bees consuming the syrup and the age of bees remaining next to the food source was recorded. Corresponding regions on the other side of the combs served as control areas. Nurses, food storers and forager bees were identified by their behaviour and their ages were recorded. Bees consuming the liquid food were between 18 and 28 days old (1. and 3. quartile), and they were older than nurse bees and younger than forager bees (Fisher's least significant difference, P < 0.05). In this experiment they corresponded to the caste of food storer bees, both with respect to their tasks (storing and distributing food) and to their age. This correspondence was expected for bees consuming syrup at the dance floor where food receiver bees are allocated but not for the bees feeding in the honey stores. The location of the syrup and the time of the day showed no significant impact on the age of the bees consuming syrup. Bees consuming syrup within the colony were about the same age as the bees in the control areas. This finding leads to the conclusion that the

liquid food source does not attract a special group of bees to the feeder. Instead, bees of a certain age are stimulated to consume syrup.

17. Solid nutrition within a honeybee colony – who feeds? U. Riessberger-Gallé, R. Brodschneider, J. Vollmann, R. Thenius, K. Uhl, S. Krainer, H. Kovac, K. Crailsheim (Institut für Zoologie, Karl-Franzens-Universität, 8010 Graz, Austria)

Supplemental Nutrition is often necessary for a honeybee colony. We investigated the number and age of bees feeding on a solid sugar pellet enriched with protein within the colony and measured the duration of individual feeding acts. Observations were carried out on a three-frame observation hive containing about 6000 bees (Apis mellifera carnica). About 50% of the bees were marked individually to identify their age. The food pellets $(3 \times 8 \times 1 \text{ cm})$ contained sugar (89.5%), fresh yeast (4.5%) and water (6%) and were presented to the bees in the middle of the honey stores and the middle of the brood nest at the same time. Corresponding regions on the other side of the combs served as control areas. Every 15 minutes the number and the age of consuming bees were noticed, duration of feedings were measured. During daytime the median age of bees feeding on the pellet was 12 to 15 days, depending on the region, during night time they appeared to be slightly younger. Bees feeding on the pellet were always older than bees staying in the control areas (Fisher's least significant difference, P < 0.05). From these observations we conclude that especially older bees actively consult the artificial food sources within the hive. There were always more bees feeding on the protein enriched food pellet compared to the number of bees staying in the control area (T-test, P < 0.01). There were more bees consuming on the pellet during night time than during daytime. Independent on the time of the day, there were always more bees on the brood nest pellet than on the pellet in the honey stores (T-test, P < 0.01). Both findings can be explained by the high density of bees during nights and within the brood area or by the temperature preference of the bees. The duration of the feeding acts was equal during days and nights, between 3 and 15 minutes (first and third quartile, U-test, no significant differences). Therefore, we assume that the amount of taken food is about the same.

21. Attempts to establish cell lines from the honey bee. R. Siede, M. König, R. Büchler, H.-J.

Thiel (Institut für Virologie (FB10), JLU Giessen. LLH, Bieneninstitut Kirchhain, Germany)

Cell lines from the honey bee would represent a significant milestone in the research of bee diseases. At the moment no established cell line is available. We intended to fill this gap by culturing bee cells in common insect cell culture media. Furthermore, a highly enriched medium on the basis of established insect media was designed. Sugars and amino acids were added in order to imitate the composition of larval haemolymph. The vitamin content of the medium was similar to the one of royal jelly. Apoptosis inhibitors and growth promoting substances were added to the media. Cells were obtained from bee eggs, embryos, neonates, larvae, young pupae and ovaries from adult queens. Tissues were disrupted either mechanically or enzymatically using trypsin or dispase. The primary cultures were incubated at 28 °C and at the brood nest temperature of 34 °C. Individual cells migrated from the explants. The formation of adherent cell clusters was observed after seeding primary cell suspensions. Frequently long, very fine fibroblast-like structures occurred around the explants. After initial proliferation the cells displayed a shrunken and granulated appearance. They finally died within several weeks. The set up of primary cultures was successful. However, subcultivation was not yet possible. With more than 140 primary cultures that were initiated we did so far not advance further than comparable studies. It remains a challenge for future work to obtain passageable bee cells which can be cultivated by standard techniques.

25. Management of bumblebee colonies for crop pollination: is a recirculation rearing program of young queens possible? A. Schlesinger, M.T. Almanza, D. Wittmann (Nutzpflanzenwissenschaften und Ressourcenschutz, FB Ökologie der Kulturlandschaft – Tierökologie, Univ. Bonn, Melbweg 42, 53127 Bonn, Germany)

Bumblebees are excellent pollinators of Solanaceae in greenhouses. Therefore, *Bombus terrestris* has been commercially reared since 1985 and sold all over the world to pollinate mainly tomatoes. However, global utilisation of commercial bumblebee colonies leads to ecological problems such as the alteration of bee fauna in countries where bumblebees do not occur naturally, and genetic changes in local races of bumble bees. Moreover, reared colonies have on average a more efficient production of new queens than wild colonies which could endanger native pollinators. These problems can be prevented by rearing and using local bumblebee species and regional races. To provide teaching material for courses on rearing local B. terrestris races, we compiled methods from the literature and from our own experiences. We also addressed whether it was possible to retrieve young gynes produced by colonies during pollination service and to integrate them into the rearing rotation. In actual practice bumblebee colonies are treated as disposable products after they have completed the job of pollination job. This means that young gynes are left to die in the nest boxes. Some of the new queens might escape and start new colonies in nature. The recirculation of young queens would avoid such unnecessary gyne mortality as well as the unwanted ecological effect of their establishment in the wild. Moreover, the breeder would get sufficient material to rear the next generation of colonies and would depend on only a few wild queens to renew the gene pool. In an experiment, we collected 314 young queens from six colonies of B. terrestris used in a greenhouses and re-integrated them into the rearing rotation. 37% were mated successfully. 47% of the mated queens started new colonies which grew to large sizes so that they could have been used for efficient crop pollination. However, collecting the queens manually out of their colonies is labourintensive. A method to retrieve young queens when they emerge has to be designed.

29. Amplified Fragment Length Polymorphism analysis of genetic caste determination in a stingless bee (*Melipona quadrifasciata*). *G.R. Makert*¹, *R.J. Paxton*², *K. Hartfelder*¹ (¹ Faculdade de Medicina de Ribeirão Preto, Universidade de São Paulo, Ribeirão Preto, Brazil; ² School of Biological Sciences, Queen's University Belfast, Belfast, UK)

The role of genetic factors in the determination of caste in the genus Melipona is a controversial issue for already more than half a century. Since the hypothesis of a genetic predisposition for caste in Melipona still has not passed a critical experimental test (so far it is supported primarily by circumstantial evidence) we adopted an AFLP method to address this intriguing question. This analysis, performed with three primer sets on 474 bees taken from four colonies of Melipona quadrifasciata, revealed that $\sim 25\%$ of the females carry genetic markers exclusively linked to the queen caste. Another ~15% of the females also carry these queen markers, but in addition also show markers linked to the worker caste. Approximately 30% of the females carry exclusively worker markers and another ~30% of the females carry none of these markers linked to caste. We take this to mean that only the $\sim 25\%$ females that carry queen markers may develop into queens, provided they encounter the appropriate nutritional conditions. All the other females (~75%) would become workers. Thus, based on our AFLP results we propose a mechanistic model that involves various loci (a multilocus system) to explain the genetic basis of caste determination in M. quadrifasciata, and we favor this over the traditional Mendelian two-loci-two-alleles system. This multilocus system could form a system or network of pleiotropic interactions, including effects on other genes (for instance by silencing their expression), that would determine the caste phenotype in M. quadrifasciata.

30. "Devoted" males – Holding and adhering structures in carpenter bees. *A.M. Muffert, D. Wittmann* (INRES-Tierökologie, Univ. Bonn, Melbweg 42, 53127 Bonn, Germany)

Xylocopa torrida, X. flavorufa and X. latipes males have specific holding and adhering structures on their legs to hold the females during copulation. With the help of the reconstructed copulatory positions of the three species, the exact function of these structures was derived. Holding structures are hooks or other adaptations of the cuticle, which are measured in mm. They operate mechanically through the muscle power of the bee. By means of holding structures the male holds fast onto the female and significantly restricts her movements during copulation. Adhering structures on the other hand are extensively haired pads on the ventral surfaces of some leg segments of the male bee. These structures are of μm and nm size. Because of the specific character of the contact surfaces, the adhering structures provide additional adhesion between male and female during copulation. They also serve to conserve muscular energy.

32. Post industrial areas – difficult terrain for bees. *L. Trein*¹, *M. Schindler*¹, *A. Hamm*¹, *D. Wittmann*¹, *W. Schumacher*² (¹ Rheinische Friedrich-Wilhelms-Universität Bonn, INRES Abt. Tierökologie Melbweg 42, 53127 Bonn, Germany; ² Rheinische Friedrich-Wilhelms-Universität Bonn, INRES Abt. Geobotanik und Naturschutz, Karlrobert-Kreiten-Str. 13, 53115 Bonn, Germany)

Post industrial areas show extreme conditions: a variety of often technogene soils, low nutrient status, contamination and warm microclimates. Bees are typically adapted to open habitats with warm microclimates. Do they use post industrial areas as habitats? On Echium vulgare, an important key forage plant for bees, pollen-production was investigated to find out how conditions on post industrial sites influence reproduction. The investigated sites "Waldteichgelände" and "Sinteranlage" differed in their soils and stage of succession. Bees were caught on these sites from April to September 2006 along transects and also on flowers. Ripe anthers of E. vulgare were taken from the post industrial areas and a control-site and analysed in the laboratory. At both sites a total of 42 species of solitary bees were found: 39 at "Sinteranlage", 13 on "Waldteichgelände". At "Waldteichgelände" only three of the species were obligate ground nesting bees. The sandy areas at "Sinteranlage" provided habitat for 16 ground nesting bees, e.g. Andrena and Lasioglossum species. Only few of the species were highly plant specific, e.g. Colletes similis or Osmia adunca. The plants at the post industrial areas produced nearly twice the amount of pollen (about 2300 ± 250 (sd) pollen grains in one anther, n = 10) than the plants at the control-site (about 1000 ± 125 , n = 10). The percentage of collapsed pollen was 28% in the control-site and 28-33% in the post industrial areas. Post industrial areas can provide habitat for bees, but their appropriateness highly depends on their stage of vegetational succession: highly structured areas with floristic diversity and a mosaic of different successional stages contain most species. Difficulties may occur for ground nesting bees. At the post industrial areas E. vulgare produced more pollen than at the control-site. The high amount of collapsed pollen could have negative effects on reproduction of both bees and plants.

33. Potential of infrared spectroscopy to classify honeydew honeys from fir and spruce. *K. Ruoff*¹, *A. Schroeder*², *K. von der Ohe*³, *W. von der Ohe*³ (¹ Inframiel, Herrenmatt 8, 5200 Brugg, Switzerland; ² Universität Hohenheim, Landesanstalt für Bienenkunde, August-von-Hartmann-Str. 13, 70593 Stuttgart, Germany; ³ Nds. Landesamt für Verbraucherschutz und Lebensmittelsicherheit, Institut für Bienenkunde Celle, Herzogin-Eleonore-Allee 5, 29221 Celle, Germany)

The potential of Fourier-transform mid-infrared spectroscopy (FT-MIR) using an attenuated total reflectance (ATR) cell was evaluated for the classification of different honeydew honeys (fir (*Abies* spp.), spruce (*Picea* spp.), metcalfa (*Metcalfa pruinosa*), oak (*Quercus* spp.) and honeydew honey from mixed origins; n = 151). Chemometric evaluation of the spectra was carried out by applying principal component analysis (for data reduction) and linear discriminant analysis (to build the classification models). In jackknife classification, high rates were found for the unifloral honeydew honeys (fir 84%, spruce 90%, metcalfa 100% and oak 100% correct). However only 58% of the honeydew honeys of mixed origins were correctly classified, indicating that numerous samples are misclassified into the groups of the unifloral honeydew honeys. The same problem has been encountered with floral honeys and may be overcome by a two step procedure (Ruoff et al. (2006) J. Agric. Food Chem. 54, 6873–6880). Hierarchical cluster analysis (Algorithm: Ward; Distance: Pearson correlation) applied on spectra of characteristic spruce and fir honeys (n = 39) showed that the fir honeys were more closely related than the spruce honeys. The more uniform composition of the fir honeys may be explained by the smaller number of honeydew producing aphid species on fir trees compared to the ones living on spruce. This preliminary study indicates that ATR-MIR spectra may be useful for classification of different honeydew honeys.

35. Rheological process analysis of processing honey with creamy consistency. J. Smanalieva¹, B. Lichtenberg-Kraag², B. Senge¹ (¹ TU Berlin, 14195 Berlin, Germany; ² LIB Hohen Neuendorf e.V., 16540 Hohen Neuendorf, Germany)

Honey is a highly viscous and supersaturated multi-component carbohydrate solution in form of a crystalline mixed phase system. The evaluation of honey structure during its processing may play an important role on sensory acceptance and texture attributes. Therefore, a rheological process analysis was used to study crystallized honey properties during processing. Accordingly, the rheological measurement was used to evaluate the effect of each processing step such as warming and agitation on honey structure. Steady shear rheological measurements were performed using a rheometer MC 120 with computer control to obtain shear viscosity $(\eta(\gamma))$ and shear stress $(\tau(\dot{\gamma}))$ in a range of shear rates $(\dot{\gamma})$ of 0, 1, ..., 50 s⁻¹ at 20, 25, 30, 35, 40 °C. Generally, the warming process T(t) caused a partially melting of the honey. Additionally, the viscosity of honey dramatically decreased from 58.87 to 7.15 Pas with increasing temperature from 20 to 34 °C. However, the agitating process at 34 °C is required to ensure the formation of a homogenized particle structure of the honey with a slight decrease in viscosity from 7.15 to 6.34 Pas and to avoid a temperature gradient and phase separation. In this study, the agitating power, shear rate and the structure development were determined by using a "Carl Fritz" agitator. The results showed that the summer flower honey had a creamy consistency when an agitating speed of 25 rpm with agitating power of 800 W/m³ was applied. The results can be used to explain clearly the effect of shear rate by agitation at Reynolds number lesser than 50 (Laminar flow) on the structure formation of honey.

36. A bee repellent endangers the quality of bee products. *A. Schroeder*, *B. Fritz*, *D. Weber*, *K. Wallner* (State Institute of Apiculture, University of Hohenheim, 70593 Stuttgart, Germany)

In several European countries a minority of beekeepers use a bee repellent spray for colony management and honey harvesting. The effective substance DEET (N,N-Diethyl-m-toluamid) is a well known repellent against mosquitoes. The spray was introduced in the market in 1988. DEET is classified as a pesticide, which automatically results in an European MRL of 0.01 mg/kg in honey. Some beekeepers used the spray as a substitute for smoke, which causes residues in honey above the legal limit. In addition, the use of DEET leads to a contamination of the beeswax, especially the uncapping wax, which is intensively exposed to the spray during the honey harvest. We investigated the migration tendency of DEET from wax into honey. Residue-free wax was spiked with 0.5 mg/kg, 5 mg/kg, 10 mg/kg and 20 mg/kg DEET. Four glass petri dishes (Ø 20 cm) were prepared with a thin layer (< 1 mm) of contaminated wax. 30 grams of DEET free honey was poured on each of the wax surfaces in layers of about 2 mm. The honey was steadily distributed by circling the petri dishes. The dishes were covered, sealed with tape and incubated with 30 °C in a brood chamber. After 25 days, the four honey samples were poured off and got analysed for DEET with a LC-MS-MS method (Thanks to Dr. Kurt-Peter Raezke, Applica GmbH, Bremen, Germany). The results showed that even the lowest level of 0.5 mg/kg in wax results in honey with DEET residues in the range of the tolerated limit. The 20 mg/kg DEET wax caused honey with a 20 times higher residue level than tolerated. A further 89 German beeswax samples were investigated (LoQ: 0.1 mg/kg). In 8 samples (9%), DEET was found at levels higher than 0.5 mg/kg (min-max: 0.5-18 mg/kg). Honey stored in wax combs containing such levels of DEET would endanger its marketability. DEET accumulates in the wax with the years. It is recommended that beekeepers that used the spray regularly in the past, should exchange the wax in their colonies completely.

39. Bees and farmers in the Kakamega forest: Economics of pollination. *M. Kasina*^{1,2}, *D. Wittmann*¹, *Ch. Martius*² (¹ Department of Ecology of Cultural Landscapes -animal ecology-, University of Bonn; Melbweg 42, 53127 Bonn; ² Centre for Development Research, University of Bonn; Walter-Flex Str. 3, 53113 Bonn, Germany)

Kakamega forest is the most important tropical forest in Kenya. It is a main source of wild bees that pollinate crops in the adjacent farmland. However, pollination may be at risk due to agricultural intensification and continued human pressure on the forest. To raise awareness of this dilemma, we quantified the reliance of crops on bee pollination. We found that most crops grown in the study area depend on bee pollination. In addition, we calculated the monetary contribution of bee pollination to crop production for the year 2005 and established that 48% of the total annual market value of the crops in Kakamega region was due to bee pollination. We then assessed the knowledge of farmers about bees and their pollinating effect. More than 98% of the 352 farmers interviewed had knowledge of different bee species but only about 50% were aware of the function of pollination for high yields and quality of crops. This awareness was positively correlated to education of the farmer. We reminded them of the role of bees and pollination in crop production, and asked whether they were willing to conserve bees. The positive effect of pollination convinced the majority (97%) of the farmers to conserve bees. Based on our findings, we suggest that policies be created that can enhance education of farmers and other stakeholders on why and how to conserve bees, and support agricultural landscape management in a manner suitable for bees. This would complement the forest as a reservoir of crop pollinators.

41. Strategies to reduce the input of pesticides in honey bee colonies from the fungicide Cantus[®]. *I. Illies*¹, *R. Büchler*, *K. Wallner*² (¹ Landesbetrieb Landwirtschaft Hessen, Bieneninstitut Kirchhain, 35274 Kirchhain, Germany, ² Universität Hohenheim, Landesanstalt für Bienenkunde 70593 Stuttgart, Germany)

The fungicide Cantus[®] is registered for use during the winter bloom of oilseed rape. In 2005 residues of its active component, Boscalid, were detected in many spring-time honey samples. The aims of our study were to collect data on the residue level of rape honeys from fields treated with Boscalid, and to develop beekeeping methods to minimize these residues. In May of 2006, nine colonies were placed at the edge of an isolated field of oilseed rape, and three colonies each were placed at 200 m, 400 m and 800 m from the same field. All colonies used this field as their main nectar and pollen source. Three days after installing the colonies, the field was treated with the maximum dosage of Cantus® (0.5 kg/ha) during full bee flight. The hive entrances of six colonies placed at the edge of the field were closed in the morning before the fungicide was sprayed and opened again after 24 h and 48 h (3 colonies per time), respectively. Eight days after application, honey samples were taken from a central comb of the honey stores. The honey from the colonies that were left open, placed at the edge of the field, contained the highest concentration of Boscalid ($21.6 \pm 6.2 \,\mu g/kg$). The residue levels in honey from colonies placed 200 m away were considerably lower (6.1 \pm 1.3 μ g/kg). In honey from colonies in 400 m away, no Boscalid residues were detected, but residues were found in honey from colonies in 800 m (12.0 \pm 13.6 μ g/kg). Compared to the open colonies, residues in colonies that were closed for 24 h were lower (9.3 \pm 8.4 μ g/kg), and no Boscalid residues at all were detected in honey from colonies closed for 48 h. However, the weight increase of these colonies during the test period was lower $(1.1 \pm 0.3 \text{ kg})$ compared to colonies with open hive entrances $(5.2 \pm 0.8 \text{ kg})$.

42. Clearcutting: Epilobium angustifolium L. as a temporary resource for wild bees and other flower visitors. J. Kühn, A. Hamm, D. Wittmann (Fachbereich Ökologie der Kulturlandschaft - Tierökologie Univ. Bonn, Melbweg 42, 53127 Bonn, Germany)

In this study we examined three aspects of the importance of Epilobium angustifolium - a widely distributed plant that grows in areas that have been clearcut - for flower visiting insects. (1) We recorded the frequency and foraging behaviour of insects on this plant over 27 days; (2) We quantified seed set after a single flower visit by different insects (n = 144 flowers); and (3) We counted the number of pollen grains of E. angus*tifolium* in brood cells (n = 20) of the oligolectic bee, Megachile lapponica, to determine the number of grains necessary for the development of one larva. Individuals of 31 insect species (13 families, 5 orders) visited the flowers of E. angustifolium. Among them were Apis mellifera (49.5%), 8 different hover flies species (23.2%) and 8 wild bee

species (11% bumble bees, 5% M. lapponica., 1% Lasioglossum spec. and Halictus spp.). Most solitary bees used E. angustifolium as a pollen resource while the honey bees collected primarily nectar. The hover flies stayed on the flowers longer than all other visitors and emptied the anthers completely. Thus, from a single pollination visit of a syrphid fly, 304 seeds/flower were produced. One visit of a bumble bee resulted in 289 seeds/flower, single visits of honey bees rendered 256 and a visit of M. lapponica generated, on average, 232 seeds/flower. One anther of E. angustifolium contains approx. 700 pollen grains (sd = 122, n = 40). The analysis showed that approx. 512.250 pollen grains of E. angustifolium are necessary to rear one larvae of M. *lapponica* (sd = 247.227, n =20). This is an equivalent of the total pollen production of one plant with 92 flowers. These data show that E. angustifolium is an important pollen and nectar source for many flower visiting insects in areas that have been clearcut. Each E. angustifolium plant represents the equivalent of one larvae of the oligolectic bee M. lapponica.

43. Honey consumption in Germany – factors of influence and effects in their interlinkage. K. Schneider, K. Forchmann, K. Friedrichs, E.M. Haas, M. Interthal, K. Jänicke, T. Kühn, B. Mergler, E. Mertens, J. Raehse, Y. Schrüffer, N. Seelinger, K. Sölch, C. Weißenborn, I. Hoffmann (Professur Ernährungsökologie, Institut für Ernährungswissenschaft, Justus-Liebig-Universität Gießen, Wilhelmstr. 20, 35392 Giessen, Germany)

Despite the high per-capita consumption of honey (1.3 kg/year) the number of honey bee colonies in Germany is declining. According to the approach of nutrition ecology, the goal of this project is to capture aspects affecting honey consumption and effects of honey consumption on the four dimensions of nutrition (health, environment, society and economy) and to demonstrate their interlinkages. Based on cited literature causal and purely statistic associations between aspects of honey consumption in Germany were composed as a causal loop diagram with hypertext structure (see www.uni-giessen.de/fbr09/nutr-ecol/ forsc_honig.php). The modelling reveals amongst others the interrelatedness and multidimensionality of the topic. The dimension environment focuses on the honey bee as the provider of honey. The bee colonies are threatened by bee diseases, pesticides and intensive agriculture. In the dimension economy is observable that the demand for honey has effects along the product chain. The profitability of beekeeping and therefore their continuance are affected by the interaction of supply, demand and price. The dimension society demonstrates the meaning of honey for different cultural epochs. The honey consumption depends on consumer preferences and different dietary patterns. For the dimension health it can be shown that honey may both prevent and induce diseases. By modelling procedures direct and indirect influences and effects as well as side effects of planned interventions become apparent. Thus the model may support decision making for planned measures, for example to secure the future pollination of plants.

45. Analysis of enzyme activity in unifloral honey from *Robinia pseudoacacia*. *B. Lichtenberg-Kraag* (Länderinstitut für Bienenkunde Hohen Neuendorf e.V., Friedrich-Engels-Str. 32, 16540 Hohen Neuendorf, Germany, lichtenberg.kraag@rz.hu-berlin.de)

Enzymes are an important indicator of the ripening process of honey. But they are also an indicator of heat damage of honey samples. Limits of enzyme activity are given by legislation. A minimum value for the enzyme diastase is prescribed by the EU-Council directive 2001/110/EC, for the enzyme invertase by the German beekeepers association and some bio-directives. Furthermore lower limits are prescribed for honey types with low natural enzyme activity; e.g. honey from Robinia pseudoacacia. The aim of our study was to investigate the causes of this natural low enzyme activity. About 60% of the Robinia honey from local beekeepers showed an invertase activity higher than 64 U_{Sieg}/kg. Less than 10% did not fulfil the directives of the German beekeepers association (45 Usieg/kg). The mean was $60-70 U_{\text{Sieg}}$ /kg and therefore clearly higher than the mean of European Robinia honey. Thirty-one samples were also analysed for diastase activity. None of the samples fell below the threshold for Robinia honey according to the EU-Council directive. We found a correlation of the enzyme activity of invertase and diastase (r = 0.72; P < 0.0001). To exclude that the enzyme might be inactivated we tried to remove potential inhibitors of the invertase by dialysis. No effects could be measured. Honey directly from combs was harvested at the beginning and end of the ripening process and analysed. As expected, moisture and sucrose content decreased and diastase activity increased. But the invertase activity was also reduced during this period although the honey was more concentrated. Whether this is an effect of the high summer temperatures or other parameters has to be investigated.

47. Use of alternative pollen sources by *Colletes hederae. R.C. Peruquetti, D. Wittmann* (Ecology of Cultural Landscapes – Animal Ecology, Univ. Bonn, Melbweg 42, 53127 Bonn, Germany)

Colletes hederae is traditionally termed monolectic. Hedera helix (ivy) would be its unique pollen source. It visits other plants, but only for nectar. Field observations and pollen analyses have confirmed this. Despite this, we report that C. hederae can use alternative pollen sources, even when ivy is present and blooming abundantly. We studied two populations of C. hederae during all of its flight season in 2006. Apart from ivy flowers, females also visited flowers of Polygonum aubertii and Solidago canadensis. We suspected that they gathered both nectar and pollen from the flowers. In order to confirm this, females were caught and their pollen load analysed. Additionally, 71 completed brood cells, containing eggs and larvae in different growth stages, were sampled and analysed. We verified that females of C. hederae harvested pollen of at least 14 plant genera in seven different families. Pollen of S. canadensis ranged from 0-100% (mean 74.6, sd = 35.38%) and of *P. aubertii* from 25-99% (mean 66.3, sd = 36.2%). Around 42% of the sampled brood cells had pollen grains from plant species in addition to ivy (range: 2.1-79.6%). We conclude that C. hederae cannot be named monolectic of ivy or even oligolectic under any of the proposed classification for pollen use by bees. This bee seems to be quite opportunistic, using what is available. This may guarantee females' reproduction through food supply and increase their colonization potential. But, it was not possible to say which factor promoted the use of one or more pollen hosts. Probably longer than normal blooming is necessary, since C. hederae starts to fly late in summer or beginning of fall. Climate change and its effects on plant phenology may play some role in determining which plants will be available to C. hederae.

48. To what extent are *Colletes*-plant interactions specialized? *R.C. Peruquetti, D. Wittmann* (Ecology of Cultural Landscapes – Animal Ecology, Univ. Bonn, Melbweg 42, 53127 Bonn, Germany)

Questions like "which bees visit this plant?" are commonly addressed in bee-plant interaction studies. Here we want to know with which plants (families, genera and species) Colletes species interact preferably and how specialized this interaction is. Colletes was chosen because some of its species thought to be oligolectic are in fact polylectic. Though oligolecty relates to pollen use, we used single presence-absence records to measure preference. An interaction matrix (550 plant species, 68 Colletes species) was set up and analysed using a "networking" approach and basic statistical methods. The same was done for two equally robust datasets comprising Andrena and Syrphidae species. These two groups and Colletes may theoretically explore the same flowers, hence they are comparable. We found that Colletes, Andrena and Syrphidae are quite specialized regarding the plants they visit. They share only 16 plants species, all known as "cornucopia flowers", e.g. Heracleum sphondylium and Angelica sylvestris. Andrena interact with more plants than Colletes and Syrphidae (Kruskal-Wallis Test, $H_{2,2023} = 45.3 P <$ 0.0001). But generalists and specialists exist in all the three groups. We hypothesize that alternative pollen sources for oligolectic species of Colletes should be found among intensively visited hosts already in the Colletes' roll. Therefore, new pollen hosts should be found mainly among Asteraceae members. Considering plant species, strong candidates are Daucus carota (Apiaceae), Melilotus albus, Trifolium repens (both Fabaceae) and Tanacetum vulgare (Asteraceae). The case of C. hederae and C. cunicularius corroborate this hypothesis.

50. Effects of sunflowers on colony development and bee health. *S. Berg*¹, *R. Siede*²

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The quality of sunflowers as a bee forage plant is unclear. Beekeepers often claim that their bees suffer damages when foraging on sunflowers. To demonstrate the effect of sunflowers on honey bee colony development we compared three groups of 6 honey bee colonies each (A, B and C) in northern Bavaria (Germany) during 2005. Groups A and B collected nectar and pollen from sunflowers. Colonies of group B were deprived of pollen. Group C had no access to profitable crops at all. Colonies in groups A and B wintered on the sunflower supplies, and C wintered on sugar solution. Estimates of population dynamics, honey yield and bee diseases were recorded before bloom, after bloom and after wintering. All colonies were of the same size before bloom, but after bloom the colonies in groups A and B were significantly stronger than the colonies in C. The average weight in groups A and B increased during bloom by +9.8 kg (A) and +10.8 kg (B) while the colonies of C lost -0.5 kg. Varroa destructor infestation, recorded as natural mite fall onto a sticky board, was comparably low in all groups before bloom but was higher in A and B after bloom. In all colonies the ABP-virus load was low and there were no significant differences (P = 0.755) among groups during the whole observation period. Before bloom all colonies were heavily infested with Nosema spp. as detected by light microscopy. After bloom the load of spores decreased only in groups A and B. Therefore, under our conditions sunflowers had a positive effect on colony development and the reduction of Nosema spp. compared to colonies without substantial nectar flow. The higher infestation with Varroa destructor may be an effect of the greater amount of brood reared by colonies visiting the sunflowers. However, ABP-virus load as a Varroa associated secondary infection was not boosted by sunflowers.

52. Spatial distribution of drones and queens (*Apis mellifera*) during mating flights in lowland areas (Wesermarsch, Germany) *C. Färber*¹, *N. Koeniger*¹, *R.F.A. Moritz*² (¹ Institut für Bienenkunde (Polytechnische Gesellschaft) Fachbereich Biowissenschaften der J.W. Goethe-Universität Frankfurt am Main, Karl-von-Frisch Weg 2, 61440 Oberursel; ² Institut für Biologie, Martin Luther Universität Halle-Wittenberg, Hoher Weg 4, 06099 Halle/Saale, Germany)

To investigate the spatial distribution of drones and queens during mating flights in lowland areas we set up two test apiaries 4 km apart, consisting of drone and nucleus colonies with unmated queens in the district of Wesermarsch in northern Germany. The distribution of drones in the test area was monitored using pheromone balloon traps. 3870 Drones were caught at 19 randomly chosen places. The density of drones on mating flights decreased with increasing distance from their colonies. This spatial distribution of drones suggested the absence of Drone Congregation Areas (DCA). Nevertheless the mating of test queens was successful. The mean mating distance of drones was calculated to be 1000 m. DNA microsatellite analysis of the queen offspring showed that significantly more bees were fathered by drones from one of the two test-apiaries. We used four closely linked loci as an economical and accurate method to obtain diagnostic results. Based on the assumption of a correlation between drone density and probability of queen mating, we estimated a relatively low mean mating distance compared to previous results using our data on distribution of drones and the results of the patriline analysis. We suggest that drones patrol the area surrounding their colonies to copulate with virgin queens as soon as they leave for mating flights. The mating behaviour in the flat test area was different to that in mountainous regions. Without the formation of DCAs one important factor for avoidance inbreeding of *Apis mellifera* appears to be absent.

53. Mate finding in the European hornet *Vespa crabro*: Involvement of sex attractants. *S. Spiewok*¹, *E. Schmolz*², *J. Ruther*³ (¹ Martin-Luther-Universität Halle-Wittenberg, Institut für Zoologie/Molekulare Ökologie, Halle (Saale), Germany; ² Umweltbundesamt – Federal Environmental Agency, FG IV 1.4, Berlin, Germany; ³ Freie Universität Berlin, Institut für Biologie, 12163 Berlin, Germany)

The mate finding strategy of drones of the European hornet Vespa crabro and the possible involvement of sex attractants were studied in free flying drones in their natural habitats. We presented pairs of caged, hardly visible living gynes and empty cages to the drones (N = 7). In a further experiment, the attraction of drones to pairs of dead untreated workers and workers treated with gyne extracts were investigated (N = 8). The extracts of drones workers and gynes were analysed by coupled gas chromatography-mass spectrometry (GC-MS). Males patrolled the nest site and nearby nonresource-based sites without showing territorial behaviour. Since drones alighted on caged queens but not on empty cages (Z = 2.201, P = 0.028), optical cues seem to play only a minor role in mate finding. They were significantly more attracted to dead workers treated with gyne extracts than to untreated ones (Z = 2.37, P = 0.018) indicating the presence of a female-produced sex attractant. Furthermore, treated workers but not untreated ones elicited copulation attempts by the attracted drones (Z = 2.20, P = 0.028). The solvent alone proved to be not attractive to the drones. Extracts from gynes, workers and drones contained exclusively cuticular lipids and the profile from gynes was much more diverse than those of workers and drones. The most striking differences observed related to the alkenes, monomethyl-, and dimethylalkanes. Although these chemicals possess only a low volatility they are good pheromone candidates in Vespa crabro since comparable hydrocarbons have been shown to be also involved in mate finding in other hymenopterans.

61. Influence of relatedness of a foreign queen on the acceptance by honey bee workers in a laboratory bioassay. *R. Alkattea*¹, *H. Steidle*², *P. Rosenkranz*¹ (¹ Apicultural State Institute, University of Hohenheim, 70593, Stuttgart, Germany; ² Institute of Zoology, University of Hohenheim, 70593, Stuttgart, Germany)

The mechanisms by which honey bee workers recognize "own" and "foreign" queens are not completely understood. In this work, we investigated the role of queen relatedness and mating status in the recognition and acceptance of a foreign queen by honeybee workers. We had 4 groups of queens: Sister/virgin and sister/mated queens and unrelated/virgin and unrelated/mated queens. 80 tests were conducted where a queen and about 30 of her own bees were put in a laboratory bioassay (wooden box $13 \times 12 \times 6$ cm with glass boards and a wax comb). Queens of two test-boxes were exchanged and both benign and aggressive contacts of workers towards the foreign queen were observed and recorded. The total number of benign contacts was higher toward sister queens compared to unrelated queens. These differences were significant for the virgin queens but not for the mated ones (P = 0.05). Similarly, the total number of aggressive contacts was higher toward unrelated queens compared to sister queens, again only significant for the virgin ones (P = 0.03). By using live queens and a "mini-colony" our bioassay represents a new approach for the quantitative evaluation of queenworker interactions. We know that worker bees are able to learn the odour of their own queen. Our results indicate that this odour has a genetic basis as virgin sister queens had significantly better acceptance compared to non-related ones. In mated queens, the high amount of queen pheromone may mask individual differences in kin specific odours.

63. Effects of body size on reproductive success of *Colletes hederae*. *R.C. Peruquetti*, *D. Wittmann* (Ecology of Cultural Landscapes – Animal Ecology, University of Bonn, Melbweg 42, 53127 Bonn, Germany)

Differential body size in many bee species relates to reproductive success. We report the effects of body size on mating success and offspring production in the solitary bee *Colletes hederae*. We sampled males and females of this bee in a large nest aggregation in Dirmstein (Pfalz region). Width of bee heads and forewing length were used as parameters of body size. We verified the relationship among these parameters and male male success and females' capacity for pollen transport. Males may adopt three different strategies to achieve mates: staying at the nest aggregation and searching for females; entering nest burrows and copulating with recently emerged females; or leaving the aggregation and searching for females at the food plant. When a female is located, a ball-shaped cluster of males is formed. But only one male succeeds in mating with her. Our data indicate that males that succeed in mating are neither among the larger ones (Mann-Whitney U Test, $Z_{34,86} = -0.87$; P = 0.40), nor do they have advantages in physical conflicts for females (Mann-Whitney U-test, $Z_{22,34} = 1.52$; P > 0.05), nor do they have access to larger females (Mann-Whitney U-test, $Z_{12,12} = -1.49$; P >0.05). Furthermore, males with different searching strategies are similar in size (Kruskal-Wallis Test, $H_{2.227} = 4.88, P > 0.05$). However, larger females carry larger pollen loads than smaller ones (linear regression, P < 0.05; n = 47). Thus large females have an advantage, and their brood cells would be better provisioned, resulting in offspring with large body size. But male body size does not affect reproductive success in C. hederae. In such a scenario, further investigations are necessary to verify how sexual allocation and maternal effects are linked in this bee.

64. Causes for delayed initiation of egg laying by inseminated honey bee queens. *R. Büchler* (LLH – Bieneninstitut Kirchhain, Erlenstrasse 9, 35274 Kirchhain, Germany)

Data from 9869 honey bee queens reared during 1985-2004 by the bee institute in Kirchhain showed that about 5% of all queens initiated egg laying when they were over 3 weeks of age. This occurred with open mated queens, and more often with artificially inseminated queens. Within the group of inseminated queens (n = 6.081), those with a delayed initiation of egg laying (later than 21 days after insemination) were compared to the rest. The age of queens at the moment of insemination, the amount of semen used for insemination and the date of queen emergence did not differ significantly between both groups (T-test). However, the number of queens with delayed egg laying was extremely high in specific years and for specific genetic groups of queens and drone sources (F-test, P < 0.01). In 2005 and 2006, the effects of different factors on the initiation of egg laying were investigated: origin of the nurse bees, food supply of the mating nuclei and the age of drones at the moment of semen collection. No differences were found between nurse bees taken from open brood combs and those from colonies without open brood (P = 0.89), or between

mating nuclei fed with pure sugar cake and nuclei fed with Nektapoll[®], a protein enriched sugar cake (P = 0.98). Insemination with semen from very young (about 2 weeks) or very old drones (7–8 weeks) had no negative effect on the start of egg laying compared to medium age drones (3–6 weeks). However, the investigation in 2006 confirmed a genetic influence of the queen line. Highly significant differences in the start of egg laying after insemination occurred in a comparison of 3 different groups of queens (P = 0.001).

70. Should I stay or should I go? Infestation level of *Varroa destructor* and its impact on honey bee and mite population structure. *P. Aumeier*¹, *J. Lipka*¹, *G. Liebig*², *W.H. Kirchner*¹ (¹ AG Verhaltensbiologie und Didaktik der Biologie, Ruhr-Universität Bochum, Germany; ² Landesanstalt für Bienenkunde, Universität Hohenheim, Germany)

investigated whether the infestation-We dependent behaviour of foragers may serve not only as a resistance mechanism of the honey bee (Kralj and Fuchs, 2006) but also could be adaptive for the mite Varroa destructor. Experimental honey bee colonies of different infestation levels were split into a queenright part, which remained at the original site and after several hours contained most of the old forager bees, and a queenless part, which contained brood and mainly younger hive bees. The number of bees and brood cells and their degree of infestation was evaluated for both parts of each original colony. Hive bees were always found to be twice as infested with Varroa destructor as foragers (Spearman r = 0.77, P < 0.001, n = 55). About 70% of all mites (3.500 ± 2.900) were found to be in the reproductive phase. The more highly infested colonies had more foragers (r = 0.49, P < 0.01, n = 31). However, the strength of the queenright part of the colonies (4 to 89% of the original hive population) was also affected by the time elapsed since establishment (10 h: $33 \pm 21\%$, 24 h: 49 \pm 22%; 48 h: 83 \pm 6%) and by the strength of the original colony (Spearman r = -0.42, P < 0.001, n = 55). Bees leaving the hive were not more highly infested by Varroa destructor than returning bees. However, more highly infested colonies lost more bees per day (r = 0.74, P < 0.001, n = 28). In a laboratory bioassay, mites infesting forager bees did not show any preference for hive bees or foragers. In contrast, mites originating from hive-bees had a significant preference for hive bees (P < 0.001, χ^2 -test, n = 688), but lost this strong bias with increasing infestation degree (r =0.45; P < 0.05). This host selection behaviour of phoretic mites might be adaptive for the parasite: at low infestation levels, parasitization of hive bees guarantees a high probability of being transported to brood cells whereas high infestation levels favour the parasitization of forager bees which can infest other colonies.

74. Invasion of *Varroa* mites into mite-free colonies at a military training area. *E. Frey*, *P. Rosenkranz* (Apicultural State Institute, University of Hohenheim, 70593 Stuttgart, Germany)

Honey bee colonies should be treated against varroosis in late summer to guarantee the production of healthy winter bees. However, the invasion of Varroa mites from non-treated colonies may wreck the treatment effect. To quantify the invasion rates we used a former military training area which is not accessible to other beekeepers. Four heavily Varroa infested honey bee colonies served as "donator" colonies. In distances of 1, 30, 400, 1300 and 1500 m from the donator colonies we placed two mite-free monitoring colonies each which were continuously treated with different acaricides. The "mite free status" was confirmed by the analysis of bee and brood samples. From August-October 2006 the number of Varroa mites on the bottom boards of the monitoring colonies were counted at weekly intervals to record the Varroa invasion rate. The average Varroa infestation rates of the adult bees in the "donator colonies" increased from 18% in August till 38% in October. Two colonies died at the beginning of October due to the high Varroa infestation. In total, 2029 mites were recorded in the bottom boards of the 10 monitoring colonies during the 8-week period. The average invasion rates per colony were 283 at 1 m distances from the "donator colonies", 201 at 30 m, 155 at 400 m, 122 at 1300 m and 255 at 1500 m. Under normal beekeeping conditions the density of bee colonies and, therefore, the invasion pressure would be considerably higher. Our data confirm that highly infested colonies represent a risk for treated colonies over distances up to 1.5 km. For beekeeping practices, the treatment of all colonies at the same time during the late summer period is an urgent need for treatment concepts.

80. FISH-analysis and putative virulence determinants. *D. Yue, A. Ashiralieva, K. Hedtke, E. Genersch* (Institute for Bee Research Hohen Neuendorf e.V., Friedrich-Engels-Str. 32, 16540 Hohen Neuendorf, Germany)

American Foulbrood (AFB) is a global and severe bacterial disease of honeybee larvae (Apis mellifera) causing considerable economic losses to beekeepers all over the world. The etiological agent of AFB is Paenibacillus larvae (P. larvae), a spore-forming bacterium. Spores are the only infectious form of this pathogen. First instar larvae become infected by the ingestion of spores through contaminated larval food. To analyse the early steps in the pathogenesis of AFB we developed an in situ-hybridisation protocol for honeybee larvae experimentally infected with P. larvae. Using rRNA targeted, fluorochrome-labelled oligonucleotide probes, fluorescent in situ-hybridisation (FISH) analysis, and Confocal Laser Scanning Microscopy (CLSM) vegetative P. larvae and honeybee larval cells were simultaneously detected via a probe specific for P. larvae 16S rRNA and a probe universally hybridising with eukaryotic 18S rRNA, respectively. Nuclei were stained with TOPRO. FISH analysis of larvae fixed and sectioned on day 1 to day 6 post infection revealed that prior to penetration of the gut epithelium the vegetative bacteria massively proliferate in the larval midgut indicating that P. larvae is able to live on what is present in the larval midgut. These data very well correlated to one of our recent studies on the metabolic profile of P. larvae. There we had shown that P. larvae is not a pure carnivore but is able to metabolise and live on different sugars present in honey and larval food. Hence, we could show that FISH analysis is a useful tool to analyse and understand the hostpathogen-interaction between honeybee larvae and P. larvae.

82. FISH-analysis of the early steps in AFB pathogenesis. D. Yue¹, N. Nordhoff², L. Wieler², E. Genersch¹ (¹ Institute for Bee Research Hohen Neuendorf e. V., Friedrich-Engels-Str. 32, 16540 Hohen Neuendorf, Germany; ² Institut für Mikrobiologie und Tierseuchen, Fachbereich Veterinärmedizin, FU Berlin, Philippstr. 13, 10115 Berlin, Germany)

American Foulbrood (AFB) is a severe bacterial disease of honeybee brood caused by *Paenibacil-lus larvae*, a spore forming bacterium. The bacterial endospores are the only infectious form of this pathogen. Young larvae of honeybees (*Apis mellifera*) become infected by the ingestion of spores through contaminated larval food. Subsequently, the spores germinate in the larval midgut, breach the epithelial barrier and proliferate in the haemocoel, thereby destroying all larval organs and tissues. Using rRNA targeted, fluorochrome-labelled oligonucleotide probes and fluorescent *in situ*-hybridisation (FISH) analysis we analysed these early steps in the

pathogenesis of AFB. In exposure bioassays worker larvae of A. mellifera were experimentally infected with the reference strain of P. larvae, ATCC 9545. In situ-hybridisation analysis was performed with infected larvae from day 1 to day 6 post infection simultaneously using a P. larvae-specific, 16S rRNA targeted oligonucleotide probe and a probe universally hybridising with the eukaryotic 18S rRNA. This strategy allowed visualization of both the vegetative P. larvae cells and the larval cells. The specificity of the P. larvae-specific probe was tested in silico and experimentally. FISH analysis supported that the spores germinate in the midgut lumen. Subsequently, a massive proliferation of the vegetative bacteria could be observed before the midgut epithelium was found to be locally penetrated by bacteria invading the haemocoel.

83. Vertical transmission routes for Deformed Wing Virus (DWV) of honey bees (*Apis mellifera*). *C. Yue, M. Schröder, E. Genersch* (Institute for Bee Research, Friedrich-Engels-Str. 32, 16540 Hohen Neuendorf, Germany)

Deformed Wing Virus (DWV) is a plus-stranded RNA virus pathogenic for both, honey bees and bumble bees. DWV can be detected in all life stages of the honey bee in the absence of visible disease symptoms. Transmission of DWV by Varroa destructor is often associated with clinical symptoms and colony collapse. Recent studies on the localization of DWV in honey bee queens and drones revealed that the reproductive organs of both were strongly positive for viral sequences, suggesting the possibility of vertical transmission routes. In addition, DWV viral sequences were recently demonstrated in sperm, further supporting the hypothesis that DWV may be transmitted vertically through drones. Conflicting results have been reported concerning the detection of DWV-RNA in eggs, bringing into question whether vertical DWV transmission occurs through the queens' eggs. Here we report for the first time evidence for the vertical transmission for DWV. To prove vertical transmission for DWV we individually analysed unfertilised eggs originating from virgin queens and fertilised eggs from the same queens after artificial insemination of these queens with DWV-negative or DWVpositive semen (as detectable by RT-PCR). By in situ-hybridisation we were able to detect viral sequences only in the ovary of an FO-queen which had laid DWV-positive unfertilised eggs and was inseminated with DWV-positive semen. In conclusion, we were able to demonstrate vertical transmission of

DWV from queens and drones to unfertilised as well as fertilised eggs, respectively.

86. First report of Nosema ceranae in colonies of Apis mellifera in Austria. I. Loncaric¹, I. Derakhshifar¹, H. Köglberger¹, R. Moosbeckhofer¹, R. Martín², M. Higes², A. Meana³ (¹ Austrian Agency for Health and Food Safety (AGES), Spargelfeldstraße 191, 1226 Vienna, Austria; ² Centro Apícola Regional, Camino de San Martín s/n, 19180 Marchamalo, Spain; ³ A. Meana, Veterinary School, UCM. 28040 Madrid, Spain)

In past years, severe losses of bee colonies were observed during hibernation in many bee yards in Austria. Also many colonies were very weak in spring. Due to findings of *Nosema ceranae* in many European countries in the last years, it was suspected, that probably this microsporidian, which infects the midgut of honeybees, is already present in Austrian honeybee colonies. Honey bee samples from Austria were inspected microscopically for *Nosema sp.* spores. Eight positive samples from different apiaries were selected and analysed for *N. apis* and *N. ceranae* using protocols of Higes et al. (2006) J. Invertebr. Pathol. 92, 93–95 and Higes et al. (2007) J. Invertebr. Pathol. 94, 211– 217. In 4 out of the 8 samples analysed up to now, the presence of *Nosema ceranae* was detected. Positive samples came from 3 different apiaries in 2 provinces of Austria.

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