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Chemisches und Veterinäruntersuchungsamt (CVUA) Freiburg

Abt. 7 Diagnostik – Bienengesundheit / Bienengesundheitsdienst (BGD)

vom 25. März bis 27. März 2025

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N: Nachwuchswissenschaftler*in

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Hauptvortrag

Neue Methoden, um fliegende Bienen mit hoher Auflösung zu filmen

Neue Methoden, um fliegende Bienen mit hoher Auflösung zu filmen

Andrew Straw

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Bienen lernen leicht, hunderte oder tausende von Metern zwischen Stock und Blumen zu navigieren. Zusammen mit anderen Neurobiologen interessiert uns die Frage, wie ein so kleines Tier mit einem so winzigen Gehirn eine solche Aufgabe leisten kann. Ich werde unsere kürzlich entwickelten Methoden besprechen, darunter eine Multicopter-Drohne, der Bienen über Hunderte von Metern hinweg folgen kann. Wissenschaftlich gesehen hoffe ich, diese Methoden nutzen zu können, um die Navigation der Bienen besser zu verstehen. Diese Methoden könnten auch für andere Zwecke nützlich sein, z. B. um Nester invasiver asiatischer Hornissen zu finden.

Keywords: n.n.

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Session 1: Ökologie, Wildbienen, Bestäubung

V1.1 BeesUp – Entwicklung einer KI-basierten Erkennungsfunktion für Wildbienen

BeesUp – development of an AI-based recognition function for wild bees

Henri Greil, Marco Seeland

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The joint project BeesUp of the Julius Kühn Institute, the Martin Luther University Halle-Wittenberg and the Technical University Ilmenau is divided into three project areas: ecology, genetics and data science. In addition to the investigation of support measures for wild bees in urban areas, the investigation of nesting bees and population genetic studies, the focus is on the development of technical solutions. For example, a multi-camera array has been developed to monitor the activities of ground-nesting wild bee aggregations.

In close interdisciplinary cooperation, the Data-intensive Systems and Visualization Group (dAI.SY) of the TU Ilmenau and the Institute for Bee Protection of the JKI are developing a recognition function for wild bees. Taxonomic details have to be clarified, tens of thousands of photos have to be taken, compiled and annotated. Based on the successful plant recognition application "Flora incognita", the models will be further developed for the specific requirements of wild bee recognition. Suitable state-of-the-art architectures of convolutional neural networks (CNNs) have been selected for training the automatic image-based identification tool. First results show that the uneven distribution and variance of the images in the photo collection significantly affect the quality of the identification. By optimizing the model architecture and hyperparameters, the recognition rate of 237 wild bee species was increased to 87% in 2022. In 95% of the test cases, the correct wild bee species is among the first three results. The analysis of misidentifications showed that about 90% of the incorrect identifications occurred within the same genus and were often due to insufficient scale, perspective and visual similarity. The correct identification rate at the genus level was 96.8%. Furthermore, it proved beneficial to train different classification targets within the same model. The identification app is available from 2025.

Keywords: wild bee, artificial intelligence, species identification, bee recognition application

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zurück zum Programm

V1.2 BeeVision – Monitoring von Bestäuberpopulationen mit künstlicher Intelligenz

BeeVision – a new AI-based approach to pollinator monitoring

Leland Gehlen, Michael Glück, Jonas Funk, Colin Gebler, Dr. Regina Pohle-Fröhlich, Dr. Andreas Wagner, Dr. Kirsten Traynor

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The loss of biodiversity worldwide is an alarming problem facing society today. Especially in the last couple of decades, insect populations have plummeted. Insects however are often keystone species that play several important roles in almost every terrestrial ecosystem as decomposers and as the main food source for many mammals and birds. Furthermore, they are the most important pollinators and elemental to our food production, as 75% of the leading agricultural crops depend on pollination. Sadly, data on insect distribution and abundance is incomplete as today's monitoring methods are time-consuming and often invasive.

BeeVision is a new non-invasive insect monitoring method that works with dynamic vision sensors (DVS). In contrast to conventional cameras, DVS have the advantage that they only record changes in brightness and thus require less storage space than conventional cameras while still recording high levels of detail. DVS cameras are able to detect insect flight trajectories and even singular wing beats. Because of the stereo setup of the DVS cameras, the speed and size of the insects can also be determined. With the help of neural networks and image-based learning, individual insect trajectories can be isolated and separated from background movements e.g. plants, cars or people. Insect flight patterns, speed and size vary and so with enough data, an AI neural network can be trained to identify and cluster isolated flight trajectories to specific pollinator groups (honey bees, bumble bees, other wild bees, hover flies, butterflies and other pollinators). The goal of BeeVision is to create an autonomous pollinator monitoring system that can track insect abundance and diversity. BeeVision is funded by the Wildcard funding program of the Carl-Zeiss-Foundation.

Keywords: pollinator, monitoring, DVS, artificial intelligence

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V1.3 Zu heiß zum Summen? Temperatureffekte auf die Blütenbesuchsrate von Bienen und anderen Bestäuben

Too hot to forage? Temperature effects on the flower visitation rate of bees and other pollinators

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Flower-rich green areas in urban environments are an increasingly important habitat for pollinators. However, cities come with numerous stress factors and due to their dense construction trap heat in paved surfaces. In densely built-up areas, temperatures thus rise much faster than in natural areas, creating urban heat islands. Currently it is unclear how heat-retaining ground coverings and the associated temperature increases affect the attractiveness of pollinator-friendly plantings.

In our study, we investigated pollinator visitation rates at two different locations at the State School of Horticulture in Hohenheim, where a defined area of the meadow was covered with roofing tar paper, while the surrounding unpaved green space served as a control. We placed 15 flowerpots in an identical grid per surface type, each planted with one of three different pollinator-friendly perennial species. Each flowerpot was observed regularly for five-minute windows between July and September 2024 while its position was switched between the surface types every other week. We also documented the number of flowers, the global irradiance and the current wind speed, while the temperature was recorded at three different heights and directly at each flowerpot. In total, we were able to perform 1,579 individual flowerpot observations in which we documented over 3,000 pollinators.

Our preliminary results show that wild bees (including *Bombus*; N=933; p= 0.022) and wild pollinators (N=1,344; p= 0.024) preferentially visit perennials on the tar paper compared to plants on the meadow grass. This was not true for honey bees, which did not differ in their flower visitation rates (N=1,676; p=0.411). The continuation of the experiment in 2025 and a more detailed analysis of the recorded temperature data will provide even more precise insights into the effects of heat islands and the resulting temperature increases on plant attractiveness.

Keywords: foraging behavior, climate change, urban heat island, pollinator observation

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V1.4 Flugfangfallen zur Erfassung von Bienen in blühenden Baumkronen

FIT for purpose? Using flight interception traps for sampling bees in flowering tree canopies

André Krahner, Anke C. Dietzsch

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Tree canopies can provide floral resources for bees in abundance. However, bee researchers often do not consider these resources, because they are difficult to access. Observer-based sampling methods, such as hand-netting along transects, are often used for collecting bees near-ground. Flight interception traps (FIT) can be used for collecting bees from elevated resources like flowering canopies. FIT are not attraction-based and may thus have the potential to assess activity density. We investigated the suitability of two FIT, i.e. window traps and aerial Malaise traps, for sampling bees from flowering canopies. We sampled in canopies of four different tree taxa (*Salix*, *Malus*, *Robinia*, *Tilia*) in Braunschweig between March and June 2024. Moreover, we compared samples from pan traps with FIT in flower strips in July 2024. In total, we collected 596 bee individuals from 9 genera, including 298 honeybees. Communities sampled in tree canopies showed marked differences in the ratio of honeybee to wild bee individuals. In *Salix* trees (March; n = 8 traps), the proportion of honeybees in the sample was lower than in *Tilia* trees (June; n = 8, Binomial GLMM & Tukey test, odds ratio = 0.087, SE = 0.057, p = 0.001) and especially than in *Robinia* trees (May; n = 5, odds ratio = 0.007, SE = 0.010, p = 0.001). The top collector units of both the window traps and the aerial Malaise traps collected not a single bee individual. Samples from pan trap triplets contained more bee individuals than window traps after 2 days of trap exposure (Poisson GLMM & Tukey test, $n_1 = n_2 = 5$ traps, ratio = 26.000, SE = 13.248; df = 14, $p < 0.001$). This was also true when exposure of window traps was extended to 5 days (ratio = 1.651, SE = 0.264, p = 0.019). We discuss our findings in a wider context, based on a systematic literature search in the Web of Science for studies using interception traps for sampling bees and other hymenopterans, and review methodological options for using FIT traps to sample bees.

Keywords: flight interception traps (FIT), flowering canopies, bee sampling, biodiversity monitoring

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zurück zum Programm

V1.5 Verknüpfung von Landschaftsmustern und wiederholten Messungen der Bienendiversität für ein standardisiertes Langzeitmonitoring in Agrarlandschaften

Interlinking landscape pattern and repeated measurements of bee diversity for standardized long-time monitoring in agricultural ecosystems

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In 2017, the "Krefeld-study „pointed to a drastic decline of *Pterygota* biomass by up to 75% in Germany. To improve the long-term monitoring of changes in biodiversity within agricultural landscapes, the collaborative initiative MonVIA was launched in 2019. To establish a robust baseline for trend analysis respective to agricultural land use changes, we developed and approach to interlink biodiversity with standardized landscape data, which is presented here.

Data were collected at 54 determined monitoring spots in northeastern Germany, spread across 5 federal states. To determine bee diversity, we used a standardized pan-trap approach (April-May 2023, 154 pan traps sets). The sampled biomass was submitted to DNA-metabarcoding for taxonomic analysis. The BLAST search generated 1288 matches from 6956 *Apiforme*s in total. The consensus resulted in 88 taxon matches at family, 82 at genera and 48 at species level.

For each of the 54 sites, the specific landscape pattern was characterized by analyzing the relative share of landscape type polygons in a buffer area of 1,77km ^{2}, surrounding the centroids of the pan traps. Input variables for the cluster analysis to define eight respective landscape types included data of regional climate, geomorphology, land cover, landscape structure as well as the intensity of agricultural use. Complementary landscape information from the German HNV-Farmland monitoring was used to characterize the monitoring sites. Depending on the site, specific landscape pattern comprised 4-7 landscape types. Predominant type was 'Medium-scale intensive arable farming', followed by 'Large-scale intensive arable farming' presented in 32% and 26% of the monitored area respective. Results indicate that bee diversity is richest in more fragmented agricultural landscape types, but this needs to be confirmed in through repeated measurements conducted as part of MonViA in the subsequent years.

Keywords: Biodiversity, Pollinators, Agro-ecology, Agricultural landscapes, Monitoring

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V1.6 Bee Well: Eine Untersuchung der Naturverbundenheit einer bienen- und bestäuberfreundlichen Gemeinschaft und wie die Beziehung zu Bienen das Wohlbefinden verbessern kann

Bee Well: An investigation into the nature connectedness of a bee and pollinator friendly community and how relating to bees can improve well-being

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It is well known that contact with nature is associated with positive outcomes in physiological and psychological health. However, studies have shown it is the quality rather than quantity of nature contact experiences that determine well-being effects. Using experimental intervention and self-report surveys, the aim of this study was to empirically investigate the effect of compassion and emotional regulation on the relationship between nature connectedness and well-being. Bees, their habitats and ecosystems provide a diverse resource for strengthening nature relationships in both rural and urban environments. The plethora of stimuli is ideally suited for subjective interpretation, allowing individuals to interpret their own unique meaning making experiences. Hence, bees are an excellent gateway species for building nature-connectedness by developing compassion for wildlife in contextually flexible interactions.

In a small pilot (n = 6) participants were randomly assigned to control and intervention groups. The intervention was grounded in the five-pathways framework and used direct, active exposure to explore and understand the needs of wild and honey bees to strengthen the participants relatedness to nature. There was a significant increase in well-being compared to control (η^2 = .74*) and over duration (η^2 = .71*). In a non-experiment, 418 participants (204 English, 214 German) from 100+ cities, Stuttgart (n = 84) and Washington, DC (n = 10) accessed an online survey. Altogether 270 participants completed the survey (109 English, 161 German) aged from 18 to 90+ (Mean = 54, SD = 19.92). Their responses revealed that self-compassion (*b* = .10*) and self-compassion with emotional regulation (*b* = .03*) fully mediated the effect of nature connectedness on well-being. These results support the three-system model as a descriptive process of the functional emotional response to nature connectedness.

Keywords: beekeeping, nature connectedness, well-being, five pathways

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V1.7 Bäuerliche Bienenhaltung: Mithilfe eines Gruppenberatungsinstruments („Farmer Bee Schools“) wird aktive Bienenhaltung Teil der biologischen Landwirtschaft und ändert die Landschaftswahrnehmung auf Bauernhöfen

‘Farmer Bee Schools’: A Modified Group Consulting Tool that integrates Active Farm Beekeeping into German Organic Agriculture and changes Landscape Perception on Farms

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In recent years Europe has been facing a growing separation and labour division between farmers and beekeepers, although agriculture and apiculture remain directly interrelated. In order to re-establish an emotional connection between farmers and the insect world, we applied the concept of Farmer Field Schools to the training of farmers in ‘Farmer Bee Schools’ (FBS) in ‘BienenHaltenHof’ (BÖL-funded 11/2022-12/2024). In regular meetings over three years on host farms, 12 participants in two regional groups learned about apiculture and discussed the integration of insects into their operation. Accompanying research analyzed under which conditions active beekeeping is feasible on modern organic farms and if FBS participation influences farm management decisions. Methods included repeated semi- structured interviews with participants, questionnaires for FBS facilitators and participatory observation. Results show that all participants successfully established healthy honeybee colonies despite some particular challenges (social, personal and organizational framework conditions). Through the eye- opening contact with the bees, supported by laboratory tests of honey and wax, floral resources on farms were assessed more and more realistically. FBS participation changed the perception of the landscape as well as other unexpected domains (social dimension of farm life). The implementation of biodiversity measures and related plans for the future increased during the project duration. We discuss farm beekeeping as a distinguished form of beekeeping, located in between professional apiculture and amateur beekeeping and framework conditions for successful FBS participation as a tool to invigorate efforts for biodiversity friendly farming.

Keywords: agriculture, farm beekeeping, farm biodiversity

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V1.8 *Helianthus annuus* Hybride mit bestäuberfreundlichen Merkmalen prägen beeinflussen die Entwicklung von *Bombus terrestris* Kolonien

Helianthus annuus hybrids with pollinator favourable traits shape *Bombus terrestris* colony development

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Floral resource diversity, abundance, and nutritional quality is a critical aspect for pollinator health and development in agricultural dominated landscapes. Sunflower, *Helianthus annuus*, is a globally important oilseed crop which is increasing in cultivation and appeal as a bee-friendly crop providing abundant floral resources, especially in late summer. Sunflower pollen as a nutritional source is often criticized by its low protein quality and induction of negative development and survival parameters for *Bombus* and *Apis* species. However, most of pollen studies are limited to cage feeding assays and exclude field realistic conditions as well as a natural floral nectar source. The aim of this semi-field study was to investigate the impact of sunflower hybrids varying in nectar and pollen production on the development of *Bombus terrestris* colonies. Colonies that had access to sunflower hybrids with superior nectar producing traits were able to develop more quickly and were significantly heavier two to three weeks after sunflower exposure compared to pollen producing and commercial hybrids (control). Subsequent pollen and nectar analysis further revealed pollen and nectar quality, rather than quantity, might be a contributing factor to the observed differences. Such that, superior nectar producing hybrids had a pronounced difference in the pollen protein quality and a further time dependent enhancement of sugar content in nectar. These findings offer a novel glimpse into how sunflower hybrids bred for favourable nectar producing traits, might offer significant advances to developing pollinator communities and the further importance of sunflower hybrid selection for cultivation.

Keywords: *Bombus terrestris*, floral resources, *Helianthus annuus*, colony development, nutrition

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V1.9 Pilotstudie zum Transfer von Per- und polyfluorierten Substanzen (PFAS) in Bienen und Bienenprodukte

Pilot study on the transfer of per- and polyfluroroalkyl substances (PFAS) in bees and bee products

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Per- and polyfluoroalkyl substances (PFAS) are a group of anthropogenic persistent environmental pollutants that have been detected worldwide in various environmental and agricultural compartments, including honey. However, knowledge regarding the potential effects of PFAS on bees and the quality/safety of bee products is currently limited. Moreover, possible contamination pathways and accumulation patterns of PFAS in bees and/or bee products are currently unknown. Especially in regions where PFAS contamination is high, the transfer from the soil to plants (e.g. nectar, pollen, guttation water) and subsequently to bees and bee products, could have an impact on food safety. Therefore, the aim of this study was to investigate the transfer of PFAS from spiked sugar syrup into bees and consequently, into bee products, such as honey or wax.

Honey bees (approximately 200 g) were kept in mating boxes and fed with different concentrations of PFAS-spiked syrup (10 µg/kg (n=4) and 100 µg/kg (n=4)). Sugar syrup without PFAS (n=2) was used as control. Fortification of the sugar syrup was done by adding 19 selected PFAS of different classes and chain-lengths. After 7 days, samples of beeswax (7±2 g per 100 g bees) and stored sugar water (51±16 g per 100 g bees) were collected from the mating boxes and stored for PFAS analysis. Bees were then fed with PFAS-free sugar syrup for 72 hours, ensuring that no traces of PFAS-contaminated syrup remained in their digestive systems. At the end of the experiment, bees were anaesthetized with carbon dioxide and stored for PFAS analysis. Quantification of the selected PFAS in bees and bee products was done using Liquid Chromatography - Tandem Mass spectrometry (LC-MS/MS).

The study results will expand our knowledge on the chemical fate of 19 structurally diverse PFAS in bee hives and help to investigate the impact of PFAS in bee products with respect to food safety and consumer protection.

Keywords: PFAS, contaminants, transfer, bee products, food safety

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zurück zum Programm

V1.10 Einblicke in das Management der Asiatischen Hornisse in Baden-Württemberg: Nestvorkommen und Hürden der Bekämpfungsstrategien

Insights into the management of the Asian hornet in Baden-Württemberg: nest occurrences and hurdles in control strategies

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The Asian hornet (*Vespa velutina nigrithorax)* is expanding its range, with south-west Germany strongly colonized. To manage the incursion of this invasive pest, a central coordination office was established at the State Institute for Bee Research (LAB) at the University of Hohenheim. We validate sightings from Baden-Württemberg, coordinate nest removal, provide educational programs for beekeepers and other stakeholders, and conduct research.

In 2024, the number of verified individual Asian hornets reported rose to over 3,300; additionally 1,450 *V. velutina* nests were located. The proportion of false and accurate hornet sightings changed throughout the season, with substantially more sightings at the end of the year.

In order to disrupt the reproductive cycle of the Asian hornet, it is imperative to eliminate nests early, before the new gynes depart. However, the majority of reported nests were found in late fall, with 311 reported in October and 506 in November. Successful nest removal was higher for primary nests, with 447 out of 468 removed, as they were more accessible. The removal of secondary nests was less successful, with less than half removed.

Despite massive efforts to destroy Asian hornet nests, the invasive species continues to spread throughout Germany. Current removal measures are neither sophisticated nor cost-effective. Consequently, we need more research to explore economically viable methods of eliminating nests. Additionally, methods for safeguarding honey bee colonies should be investigated. The utilisation of "selective traps" has been proposed; however, these devices are non-selective, capturing non-target species, including the protected European hornet (*Vespa crabro*) and other social wasps. These traps must be improved to enhance their specificity to avoid unintended bycatches.

Keywords: Vespa velutina, Asian hornet, nest removal, Baden-Württemberg

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zurück zum Programm

Session 2: Physiologie & Verhalten

V2.1 'Vitalbiene' – Auswirkungen innovativer Bienenhaltung auf Leistung und Vitalität von *Apis mellifera L*

'Vitalbiene' – Effects of innovative beekeeping on performance and vitality of *Apis mellifera L*

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Keine Veröffentlichung des Abstracts!

Keywords: Varroa destructor ; summer brood interruption ; honey bee health; population dynamics

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V2.2 Wie man die Pigmentierung von Honigbienen verändern kann: CRISPR/Cas9-induzierter Knockout von Genen für die Pigmentsynthese und die Auswirkungen auf die Physiologie

How to change honey bee pigmentation: Knocking out pigment synthesis-related genes using CRISPR/Cas9 and the effects on physiology

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The cuticle represents the outermost layer of an insect. It is a hardened structure of chitin and other compounds like sclerotins, creating a tight barrier towards the environment. Different pigments are incorporated into the cuticle during development. They define the color of the insect. The functions of pigmentation are diverse, including adaptation to temperature, humidity, UV and pathogens, and sometimes contributing to sexual selection. While pigmentation has been studied in some insects including the fruit fly *Drosophila*, knowledge about pigment synthesis in honey bees (*Apis mellifera*) is scarce. We investigated the function of *ebony* and *tan*, two genes required in pigment synthesis, in the Western honey bee (*Apis mellifera carnica*) using CRISPR/Cas9-mediated gene knockout. While *ebony* drives the production of yellow sclerotin from dopamine, *tan* catalyzes the reverse reaction. Lab-reared honey bee mutants differed significantly from wild type controls in their cuticle pigmentation. Mutation of *ebony* led to very dark bees with pale hair and dark wing venation. Knocking out *tan* made the emerging bees brighter and more yellowish than wild type bees, while the pigmentation difference was less prominent compared to that of *ebony* mutants. RNA-sequencing indicates differential gene expression in head, thorax and abdomen between mutants and wild types. Differentially expressed genes were related to diverse pathways and functional gene clusters, for instance, metabolism. Our study demonstrates that *ebony* and *tan* are important genes driving honey bee pigmentation and cuticle-related transcriptomic processes.

Keywords: Apis mellifera carnica, CRISPR/Cas9, pigmentation, ebony, tan

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V2.3 Die Rolle von Acetylcholin in der Entwicklung der Honigbiene: Von der Ernährung über die Rezeption bis zum Signal

The role of acetylcholine in honeybee development: From nutrition through reception to signaling

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Honeybees raise their brood using specialized glandular secretions known as brood food, representing a sophisticated form of social care in insects. Acetylcholine (ACh) in brood food serves as a crucial signaling molecule during larval development, with disruptions to the cholinergic system causing developmental delays. The concentration of ACh varies significantly between castes and across different larval stages. The molecular mechanisms governed by ACh and the precise location of its receptors in developing larvae remain unknown, limiting our understanding of this essential developmental signaling system. Here we show seasonal variations in brood food ACh content, caste-specific differences in ACh concentrations, and successfully map ACh receptor subunit distribution using HCR-FISH in honeybee larvae.

We found that ACh levels in brood food peak in April at three times the concentration observed in August, independent of abiotic factors. Brood food fed to queens and drones contains distinct ACh concentrations compared to worker-destined food. Our HCR-FISH analysis of late-stage embryos reveals clustering of the α1-subunit near the esophagus. These seasonal fluctuations may influence winter bee development, while caste-specific differences could contribute to caste dimorphism, and the receptor localization suggests specific developmental targeting. In our next steps, we will investigate ACh's influence on juvenile hormone production through qPCR analysis, as this hormone pathway is crucial for development, caste determination, and winter bee formation.

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V2.4 Verständnis der altersabhängigen Interaktion zwischen Darm und Gehirn von Honigbienenarbeiterinnen durch die Integration von Multi-Omics-Ansätzen

Comprehension of the age-dependent gut and brain interaction of honey bee workers by integration of multi omics approaches

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In honey bee colonies, division of labour is a key feature, with age-related behavioural transitions being closely associated with molecular changes in the brain, gut, and microbiota. Despite evidence of both microbiome and brain changes, most studies focus solely on either aspect or a single method of investigation, limiting our understanding of their interconnected roles in development and task differentiation. This study investigated the molecular changes in the gut and brain in honey bee workers of different ages using (meta-)proteomics and metabolomics, to better understand their contribution to behavioural modulation. (Meta-)proteomics and metabolomics of the gut and brain provided insights into the global structural and functional dynamics of the microbiota, as well as the functional and metabolic alterations in the host gut and brain, and their interactions. Our results indicate the transport of amino acids between the gut and brain, potentially influencing functional pathways and behavioural phenotypes. We found a correlation between concentrations of tryptophan and its metabolic products between honey bee brain and gut. This highlights the gut-brain axis as a key internal communication for different host mechanisms in honey bees. Furthermore, we show that microbial community richness peaks after three days, coinciding with short-chain fatty acid concentrations. Our proteomic results from both the host and the microbiota reveal that altered metabolic and functional pathway abundances may be due to energy expenditure, task differentiation, and age of onset of foraging. Overall, our findings are the first to comprehensively describe the global (meta-)proteomic and metabolomic changes in the honeybee gut and brain throughout a worker's life. This provides new insights toward developing potential biomarkers for evaluation of different functional changes related to various environmental stressors.

Keywords: honey bee, host-microbiome interactions, metabolomics, metaproteomics

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Session 3: Genetik & Zucht

V3.1 Genomische Diversität der deutschen Honigbienenpopulation

Genetic diversity of the German honeybee population

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The German honeybee population is heavily influenced by breeding and commercial trade. While the different landscapes in Germany and individual preferences of beekeepers demand diversification, breeding for high productivity and easy management is likely to diminish genetic diversity. We report on a genomic monitoring of the whole honeybee population within Germany.

Via mail we received 2009 samples of workers coupled with questionnaires from volunteering beekeepers. Genotyping was done on the HONEYBEE_2021 SNP chip which comprises 70‘814 markers. The samples from the survey were combined with ~3000 samples of 20 subspecies from Europe, the Middle East, and Africa. Samples and markers were filtered to build a new tool for estimating ancestry from evolutionary lineage. Most German bees show a high degree of *Apis mellifera carnica* subspecies identity, which was also the major genetic background of Buckfast bees though here levels of admixture were also the highest.

To identify regional differences or smaller breeding lines, the German samples were clustered on their own. The clusters were compared to the data from questionnaires. This revealed small degrees of local diversification within Germany, and genetic independence of individual breeding lines, e.g. the so-called Sklenar bee.

A key number for genetic diversity is the effective population size. The mating biology of honeybees is challenging to current methods, and we conducted simulation studies to put results into perspective. Our estimate of the effective population size for the German honeybee population lies above 3000 which suggests a rather large genetic diversity overall. Individual estimates for *Apis mellifera carnica* and the Buckfast bees were also high, although the populations are admixed.

The results suggest that the German honeybee population has a healthy level of genetic diversity.

Keywords: Zuchtlinien, Unterarten ,Monitoring, SNP-Chip, effektive Populationsgröße

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zurück zum Programm

V3.2 Über die Veränderung von Heritabilität und genetischer Korrelation über die Zeit

On the change of heritability and genetic correlations over time

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The heritability of a trait within a species is contingent upon the specific breeding population at a given point in time. Precise quantification of heritability is imperative for the accurate estimation of breeding values and the prediction of breeding efficiency. Accurate knowledge of genetic correlations between traits support the efficiency of simultaneous selection. Heritability and genetic correlations change as allele distributions in a population shift over time.

Reliable estimation of genetic parameters requires sufficient depth and breadth of breeding data. Broader datasets improve robustness, while shorter time frames highlight sensitivity to heritability changes, creating a trade-off: temporal breadth facilitates robust parameter estimation, while temporal narrowness captures variations in heritability over shorter time periods."

In this study, we analyse heritabilities and genetic correlations of key traits in the A.m.carnica breeding population, including honey yield, gentleness, calmness, swarming inertia, pin test results, and Varroa infestation development, over time. This population possesses a substantial history of breeding data spanning several decades (Hoppe et al. (2020), Insects 11:768), allowing fine-tuned estimation of genetic parameters, enabling a comprehensive study of their temporal variation in relation to specific parameters, such as the time span of the data and the year.

In summary, a biphasic pattern can be identified in the development of heritability for all traits. During an acceleration phase, heritability increases, while a saturation phase is characterised by a decrease. Traits with low heritability reach saturation at a later stage.

This analysis provides a deeper understanding of the properties of the Carnica breeding population and its progress, while also supporting the formulation of guidelines for other breeding populations lacking extensive data resources.

Keywords: heritability, genetic correlation, breeding progress, Apis mellifera carnica

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V3.3 Vorabgenotypisierung von Königinnen und Drohnen für die künstliche Besamung bei gezielter Anpaarung

Pre-genotyping of honey bee queens and drones prior to artificial insemination for controlled breeding

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Artificial insemination is an established and powerful approach that allows targeted mating of queens with selected drones in breeding programs. Altough inbreeding is not intended in a classic breeding program, siblingmating is required for investigation of diploid drones. Consequently, the genetic background of drones and queens need to be known a priori to maximize the efficiency of diploid drone production.

Diploid drones result from a homozygous allele combination at the gene *complementary sex determiner (csd)* and have zero fitness. These drones are recognized and eliminated by worker bees in the early larval stage. Here we show a reliable method to produce a high proportion of diploid drones through siblingmating with the contolled match of homozygous *csd* alleles at.

This requires samples for the extraction of DNA without sacrificing the individuals. Notably injuries or irregularities in a honey bee queen can result in her replacement by the worker bees. A non-destructive method of sampling queens involves nymph skin remaining in the queen cell. The middle legs was used to determine the genetics of the drones. For both drones and queens *csd* was amplified by PCR and subsequently sequenced.

We show the sucessful implementation and the differention of the *csd* alleles. According to their genetic background of *csd* the drones were assigned to their sisters. The sperm of the drones was collected as a mixture for each *csd* allele group and the queens were inseminated. Although the queens were inseminated with several drones, they were only inseminated with a single haplotype in terms of *csd*.

Pre-genotyping therefore makes it possible to combine the advantages of mutiple and single drone insemination, and to perform targeted mating. This method is an efficiant and reliable way to produce diploid drones in a more targeted way and it might contribute to an improved selection for various traits in a breeding programme, if the genetic background of the individuals is known in advanced.

Keywords: Apis mellifera, pre-genotyping, artificial insemination, non-destructive sampling, csd

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V3.4 Die Bewahrung der Heterozygotie in der thelytoken Kap-Honigbiene *Apis mellifera capensis*

The preservation of heterozygosity in the thelytokous Cape Honeybee *Apis mellifera capensis*

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The Cape Honeybee *Apis mellifera capensis* is a subspecies of the Western Honeybee, native to the fynbos biome in South Africa. Many Cape Honeybee workers are able to produce female offspring from unfertilized eggs via thelytoky with central fusion, a form of parthenogenesis where diploidy is restored by the fusion of meiotic products. While this mode of reproduction may be adaptive, e.g. to re-queen the colony from worker-laid eggs, it is predicted to result in a loss of heterozygosity (LOH) and therefore in a reduction of genetic variation and fitness. Despite this detrimental effect, a permanently thelytokous, parasitic lineage of Cape bees is persisting for over 30 years, while large fractions of its genome retained heterozygosity. Yet it remains unclear whether this is a consequence of reduced recombination or strong selection against homozygotes and what long-term effects there are.

Using whole genome sequence data of families as well as developmental and population time-lines of South African Honeybees from the native population and the parasitic lineage, we estimate recombination rates in queens and workers, the level of selection against LOH during development and the long-term consequences of thelytokous reproduction in the parasitic lineage.

The Cape Honeybee represents an ideal study system to investigate mechanisms of preserving heterozygosity despite asexual reproduction and inbreeding and serves as a model for other thelytokous organisms.

Keywords: Cape Honeybee, parthenogenesis, recombination, selection, genetic diversity

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Session 4: Bienenschutz & und Pflanzenschutz

V4.1 Einfluss der Pollenqualität auf die Widerstandsfähigkeit der Honigbiene (*Apis mellifera*) gegenüber Pflanzenschutzmittel

Influence of pollen quality on honey bee (*Apis mellifera*) resilience against pesticides

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Food quality plays a decisive role in the stress resistance and resilience of honey bees. However, in intensive agriculture, a monofloral food supply often dominates, leading to an unbalanced diet for bees. This study assessed the potential of different pollen diets (*Brassica napus, Helianthus annuus, Phacelia*) with varying nutritional quality to reduce pesticide toxicity in honey bees. Purity of plant sources of the bee-collected pollen samples was analyzed palynologically. Furthermore, pollen samples were characterized for their composition of amino acids, secondary metabolites, sugar derivatives and fatty acids. We conducted parallel cage studies at two labs, exposing nurse bees to four different pesticide treatments: 1) a field relevant dose or 2) a max. permitted dose of fungicides (Boscalid + Pyraclostrobin), 3) an insecticide (Dimethoate) as a positive control and 4) no pesticide as a negative control. For each pesticide treatment there were five pollen options: single pollen type feeding, a mixture of all pollen types and a no pollen control. We recorded daily food consumption and survival.

Our results show that the three pollen types differ in amino acid composition, occurrence of secondary metabolites and fatty acids. Longevity varied significantly across pesticide treatments and pollen diets, with the sunflower diet and the no-pollen diet resulting in the highest mortality ($p < 0.001$, $N = 240$). Increased mortality also occurred when feeding the max. permitted fungicide concentrations, as expected, this was also the case with the positive control dimethoate. Furthermore, pollen consumption differs between treatments and diets ($p < 0.05$, $N = 192$). These findings indicate that specific pollen diets can reduce the lethal effects of fungicide exposure on honey bees. The nutritional variance of the different pollen types may explain their protective effects.

Keywords: fungicide exposure, artificial pollen diet, sublethal effects, survival, Apis mellifera

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V4.2 Pestizidexposition von Honigbienen (Apis mellifera) in Apfelplantagen beim Einsatz von kupferhaltigen Fungiziden

Real-world pesticide exposure of honey bees (Apis mellifera) in apple orchards with copper fungicide application

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This study investigated the effects of a copper (Cu) fungicide on honey bee (Apis mellifera) colonies placed in apple orchards. Field experiments were conducted at three sites with varying exposure to Cu fungicides and other pesticides. As the sites are located in a region of intensive agriculture, levels of Cu were high even prior to Cu application. Orchard treatment with Cu increased Cu resides in apple and dandelion blossoms, but due to high starting levels the change was small ($p > 0.05$, $N = 21$). To better understand bee foraging preferences and exposure risk, we separated the bee collected pollen visually by color into seven different groups. Each pollen color was identified to the plant source. Cu residues were found across all of the sorted pollen types, indicating that bees face exposure from a wide range of forage sources, though exposure varied significantly between pollen sources ($p = 0.0016$, $N = 19$). Cu levels in honey crops (0.91 mg Cu/kg, $N = 25$) and stored honey (0.18 mg Cu/kg, $N = 15$) remained low, suggesting that bees possess physiological mechanisms to filter or detoxify Cu, potentially reducing its transfer into honey. Pesticide residue analysis of the trapped pollen ($N = 22$) demonstrated site-specific variations in contamination, with some samples showing residues of pesticide substances that were not sprayed within the orchards themselves. This could be an indicator of the wide foraging radius of honey bees, which can lead to unexpected pesticide exposure at external sites that is then carried back to the colony. These findings underscore the complexity of real-world Cu and pesticides exposure in agricultural landscapes. While Cu levels in honey were low, the widespread presence of Cu in bee-collected matrices calls for further research into its long-term impacts on colony health.

Keywords: copper fungicide, synthetical pesticides, exposure risk, filter mechanism, detoxification, *Apis mellifera*

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V4.3 Jenseits des chemischen-synthetischen Pflanzenschutz: Bewertung von Tankmischungen aus dem Ökolandbau, hinsichtlich der Sicherheit für Honigbienen (*Apis mellifera*)

Beyond Chemical Synthetic Pesticides: Evaluating biopesticide tank mixtures for honey bee safety (*Apis mellifera*)

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Biopesticides have become an increasingly important market in recent years as they are presumed to be safer due to their naturally occurrence in the environment. Biopesticides are not only natural substances that act in a similar way to chemical synthetic pesticides, but a large proportion of biopesticides are products that contain microorganisms as active ingredients.

During the approval process of plant protection products (PPPs), it is necessary to assess their effects to honey bees. In contrast to chemical synthetic PPPs, microorganisms take longer to exert their effects and can be pathogenic and infectious. Therefore, the available guidelines developed for testing the effects of chemical PPPs on bees are not helpful for microorganism based products without appropriate modifications.

This study aims to understand the effects of biopesticide tank mixtures (TM) on bees. TM are commonly used as they are more economical and reduce resistances. It is known from conventional farming that mixtures of PPPs can increase the toxicity to honey bees. However, studies on the toxicity of TM for bees in organic farming are largely lacking. Therefore, we investigated different products commonly used in ecological apple orchard farming. For example NeemAzal-T/S (10.6 g/L azadirachtin) and DiPel DF (1.17 x 10^{13} cfu/kg *Bacillus thuringiensis subsp. kurstaki* ATBS-351).

The tests followed the existing OECD 213 and OECD 239 guidelines, with adaptions to account for the presence of microorganisms in the products (e.g. autoclaved control, extended test duration).

In the laboratory, the oral exposure to the TM of NeemAzal-T/S and DiPel DF showed no statistically significant effects on adult honey bees compared to the controls and individual products. On the other hand, it showed strong effects on honey bee larvae. Thus, further higher tier studies should be conducted to assess potential effects on honey bee brood under more field realistic conditions.

Keywords: biopesticides, tank mixtures, honeybee larvae, microorganisms, apple orchard

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V4.4 Osmia Arten unterscheiden sich in ihrer Sensitivität gegenüber Pestiziden und Fungizid erhöht die Mortalität von Insektiziden

Osmia species differ in pesticide sensitivity and fungicide increases lethality of insecticides

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Pesticides are considered be an important driver of anthropogenic induced loss of bee species. Although sensitivity to pesticides varies between species, studies and risk assessments are primarily focused on managed *Apis mellifera* and, to a lesser extent, on other generalist bee species. However, the suitability of *A. mellifera* as a model species has been questioned due to its substantial ecological differences to other bees, particularly solitary and oligolectic species, which might vary in how these bees are affected by pesticides. Moreover, the toxicology of pesticides is mostly assessed as individual compounds, although pesticides are often applied in mixtures and interactions between pesticides can occur.

As part of the EU project WildPosh, we conducted a lab study to analyse the sensitivity of the oligolectic *Osmia brevicornis* compared to the polylectic *Osmia bicornis.* The acute oral and contact toxicity of the insecticides acetamiprid and cypermethrin, the fungicide tebuconazole, and their combinations were analysed to assess species sensitivity and potential pesticide interactions.

Our results show that *O. brevicornis* is more sensitive than *O. bicornis* when contact exposed to pesticides (bootstrap: $p < 0.01$ in 3/7 treatments), suggesting that a robust body morphology with a thicker cuticle may be a critical determinant for contact sensitivity. Also, synergism in impact between cypermethrin and tebuconazole on the survival of both species and a trend toward synergy between acetamiprid and tebuconazole were observed, suggesting that tebuconazole might inhibit detoxification processes of insecticides in *Osmia.*

The results highlight the need for a broader range of test species and consideration of pesticide interactions in risk assessment as tebuconazole might be safe to bees on its own but it has the potential to increase the toxicity of insecticides.

Keywords: Osmia, Pesticide toxicity, Synergistic interaction

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V4.5 Eine Krise der männlichen Unfruchtbarkeit als Ursache für den Rückgang der Hummeln?

Male infertility crisis underlying wild bumble bee declines?

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In eusocial Hymenoptera, males typically do not engage in colony labor but play a crucial role in colony fitness. Given the well-documented evidence of male infertility in various species, including humans, among others due to environmental pollution, similar effects may also hold true in bumble bees. However, data on the impact of agrochemical exposure on male Hymenoptera remain limited. Here, we show for the first time that exposure to a glyphosate-based herbicide (GBH; Roundup®) can significantly reducing male fertility in bumble bees, *Bombus impatiens*. In the laboratory, following standard OECD protocols, we challenged individual males chronically for 10 days with a field realistic concentration (7.6 mg a.i./L) of Roundup®. Survival was assessed daily and total living sperm at two different time points - day three and 10. Intriguingly, Roundup® exposure significantly enhanced longevity (Kaplan Meier, $\mathcal{X}^2 = 9$, $P = 0.002$), possibly due to an underlying hormetic effect. Concurrently, a time-dependent significant negative effect of exposure on the total living sperm was revealed (GLM, $P = 0.01$). Males exposed to Roundup® for 10 days ($N = 22$) revealed a 34% reduction in total living sperm compared to control males ($N = 19$) of the same age. When considered alongside past research on neonicotinoids, the present findings strongly indicate that male insect infertility resulting from agrochemical exposure may indeed be a widespread, yet vastly overlooked, phenomenon. The impact of environmental pollution on male fertility thus emerges as a key driver of the observed declines in wild bumble bee as well as other social and solitary insect populations. Therefore, these findings underscore the urgent need for decisive mitigation efforts, including revised and more robust toxicological risk assessments.

Keywords: Bombus impatiens, herbicide, spermatozoa, sub-lethal, toxicology

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V4.6 Wann gilt ein Pestizid als „geringes Risiko“? Eine simulationsbasierte Evaluation von EFSA‘s Äquivalenztest und alternativer Methoden

When is a pesticide 'low risk'? A simulation-based evaluation of EFSA's equivalence test and alternative methods

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Pesticide risk assessments have traditionally used difference testing, where failure to detect significant negative pesticide effects is interpreted as safety. However, this approach risks overlooking harmful effects in underpowered studies. To address this, the European Food Safety Authority (EFSA) advised a shift from difference testing to equivalence testing in their latest guidance document for pesticide risk assessments for bees. Under the new paradigm, a pesticide is classified as ‘low risk’ when a null hypothesis of relevant negative pesticide effects can be rejected. Specifically, low risk is concluded if the reduction in honeybee colony size in the pesticide group relative to the control group is significantly smaller than 10%, the threshold defined as the specific protection goal. However, concerns have been raised that the new guidance would make it too difficult to demonstrate a pesticide’s safety.

Here, we evaluate EFSA’s equivalence testing approach through simulations based on data from a field study. We compare this approach both to difference testing and to an alternative equivalence test proposed by Hotopp et al. (2024, IEAM 20:1496-1503) that calculates treatment effects as the difference between the pesticide group mean and the lower bound of the 90% confidence interval of the control group. We explore how the probability of classifying a pesticide as low risk varies with testing approach, true effect size, sample size, fixed and random effects structures, and colony allocation process (random allocation vs. anticlustering randomization based on initial colony size).

Keywords: Equivalence testing, risk assessment, pesticides, Apis mellifera, plant protection products

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zurück zum Programm

Session 5: Bienenpathologie

V5.1 Erste Ergebnisse der *Varroa destructor*-Befallsbestimmung in Bienenvölkern mittels automatisierter Bildauswertung

An Automated AI-Based Diagnostic Method for Assessing *Varroa destructor* Infestation in Honeybee Colonies

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Varroosis, classified as a Category C, D, and E disease under EU Regulation 2018/1882, is a primary cause of winter losses in beekeeping. The parasite, *Varroa destructor*, directly weakens affected colonies as a brood parasite and serves as a vector for various viruses. Traditional diagnostic methods include wash tests, the "powdered sugar method" and counting mites dropping onto a bottom board. This study aims to validate an automated, AI-based method for determining within-colony mite fall.

The BeePal Varroa-Detector utilizes "debris diagnosis" at the bottom board combined with AI-driven image recognition. Unlike conventional practices that conduct periodic mite fall assessments for a limited duration, this system analyzes debris continuously. The debris is transported on a conveyor belt into a diagnostic area where it is photographed, stripped, and collected in a drawer. Captured images are evaluated using cloud-based AI, producing scan results displayed through a dedicated software interface. The AI generates personalized infestation forecasts for each colony in combination with additional sensor information to predict future infestation-levels and helps the beekeeper in planning varroa-treatment and verifying treatment success.

Various technical challenges associated with automation have been addressed. Validation was achieved by counting mites in high-resolution scans over a complete month. The AI detection accuracy stands at approximately 95%. Results derived from the AI analysis were compared with findings from a wash test conducted on the same colony, with colony strength initially determined using the Liebefeld estimation method. The mite counts identified through AI detection were significantly higher than those anticipated from the wash method, indicating potential underestimation by traditional techniques.

Further investigation is warranted to explore these findings in depth; the implications of the results will also be discussed.

Keywords: Automated detection, Varroa destructor, artificial intelligence

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V5.2 Lithium Citrat als Behandlungsmittel gegen *Varroa destructor*: Dosisfindung und Ermittlung der Effizienz

Lithium Citrate as *Varroa destructor* control agent: finding the right dose and evaluating its efficacy

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Lithium, bound as salt, is a promising new control agent against the honey bee parasite V*arroa destructor*. In the process of developing lithium salts into a registered veterinary treatment, we wanted to investigate, if lithium citrate (LiCi) is as effective as lithium chloride (LiCl), which has shown high efficacy in prior studies. Our goal was to establish an ideal dosage, determining the minimum treatment with maximum effect.

The trial was undertaken in August 2024, in cooperation with the Varrolis GmbH and the Bavarian State Institute for Wine and Horticulture Veitshöchheim. Thirty honey bee colonies were set up in two locations, Hohenheim (n=15) and Veitshöchheim (n=15). Here we focus on the results from Hohenheim. All queens were caged and released after three weeks, when colonies were broodless, and all colonies were fed four litres of a 1:1 sugar solution. The sugar solution contained dissolved LiCi in four different doses: 12,5 mM, 25 mM, 37,5 mM and 50mM (n=24; n=6 per group). Six colonies were sprayed with oxalic acid as a positive control. During the trial period of six weeks, dead *V. destructor* mites were counted on bottom boards on a regular basis. Ten days after treatment the remaining mites were killed with Flumethrin (Bayvarol®) to determine the efficacy in all groups. The control group showed the lowest efficacy (avg. 74%), similar to the lowest dose of 12,5 mM (avg. 78%), followed by 25 mM (avg. 91%). In the highest doses all colonies were treated with an efficacy above 95% (avg. 97%, min = 97%, max = 98), except one with a 92% efficacy (50 mM), suggesting that the optimum dosage might be around 30 mM. Thus, LiCi is an effective, easy to use and non-harmful varroacide to treat *A. mellifera* colonies against *V. destructor*. A detailed statistical analysis and pooled data from both locations will be presented in the talk.

Keywords: Lithium Citrate, *Varroa destructor*, *Apis mellifera*

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V5.3 Bewertung integrierter Varroa-Behandlungsmethoden: Auswirkungen auf Milbenbefall, Bienengesundheit und Wirksamkeit der Behandlung

Evaluation of four integrated varroa management methods: effects on mite infestation, colony health, and treatment efficacy

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The parasitic mite *Varroa destructor* poses a severe threat to honey bee (*Apis mellifera* L.) colonies worldwide, compromising their health and reducing their survival by feeding on their fat body reserves and transmitting deadly viruses. This study systematically evaluated the effectiveness of four *Varroa destructor* control strategies: 1) chemical free biotechnical control methods, 2) drone comb cutting combined with organic formic acid treatment, 3) an integrated approach combining biotechnical and organic oxalic treatments, and 4) synthetic control using a registered acaricide (flumethrin). The research was conducted using 38 honey bee colonies at three locations in southern Germany. Colony strength, mite infestation levels, treatment effectiveness, and various health parameters—including winter bee survival, fat body content, and virus loads—were assessed.

Results demonstrate that biotechnical methods and formic acid treatments provided the highest *Varroa destructor* reduction (~81%), while flumethrin and oxalic acid combined with induced broodless stages exhibited lower efficacy (~71% and 61%, respectively). Colonies without drone brood removal exhibited significantly higher mite populations. Swarm tendency was lowest in colonies managed with drone brood removal, whereas those without it showed increased swarming behavior. Notably, synthetic treatments negatively impacted fat body reserves and winter bee survival, whereas biotechnical and organic methods had fewer physiological side effects.

This study highlights the importance of integrated management, emphasizing the effectiveness of biotechnical methods for sustainable mite control. However, labor-intensive implementation remains a challenge for large-scale beekeeping operations. These findings provide beekeepers with essential insights into the benefits and limitations of different *Varroa destructor* control methods, enabling informed decision-making for colony health and productivity.

Keywords: *Varroa destructor*, integrated pest management (IPM), honey bee health, biotechnical varroa control

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V5.4 Winterbrutpausen verringern den Fortpflanzungserfolg und die Fekundität von *Varroa destructor*

Winter brood breaks reduce the reproductive success and fecundity of *Varroa destructor*

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The western honey bee (*Apis mellifera*) plays a vital role in global agriculture. Yet colony losses continue to plague beekeepers, especially over the winter. *Varroa destructor* is considered one of the most serious threats, as it parasitizes honey bees, feeding on their fat body and hemolymph, and transmitting multiple viruses, with negative health impacts at both individual and colony levels.

Many studies have demonstrated that *V. destructor* populations follow a predictable seasonal pattern, increasing in summer, peaking in early fall, and declining over winter. During winter, when brood is absent or limited, mites are forced into the dispersal phase on adults for several months, rather than the typical 3-5 days. Despite winter brood breaks, *Varroa* populations successfully recover in the next season. However, little is known about the mechanisms underlying this population recovery after prolonged winter brood breaks. In this preliminary study, we determine how many mites survive long breaks and investigate how mite populations rebuild when the first batch of brood becomes available.

We compared mite reproductive ability in the first batch of brood in colonies with naturally low winter brood populations (n = 4) and previously broodless colonies due to queen caging for 100 days (n = 5). Our preliminary results showed that about half of the mites in both groups entered the first batch of brood available in February for reproduction, while the other half remained in the dispersal phase. While the mites were still able to reproduce after a long winter brood break, their reproductive success and fecundity were significantly reduced. Understanding the factors influencing *V. destructor* population growth during the winter season is critical for developing effective management strategies and may provide valuable insights into why *Varroa* populations build up slowly in the first part of the beekeeping season and how prolonged brood breaks influence mite population dynamics.

Keywords: honey bees; winter brood break; parasite; *Varroa destructor*

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V5.5 Auswirkungen der herkömmlichen und der innovativen Bienenhaltung auf die Pathogenlast von Honigbienen

Effects of conventional and innovative beekeeping on the pathogen load of honeybees

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Varroa destructor poses a major threat to honeybee colonies, acting as a vector for honeybee diseases and contributing to colony collapse. To control mite infestations, beekeepers apply treatments against the mite. Under conventional management, organic acids are used to reduce mite loads, and drone brood is removed during spring and summer. An alternative approach is an “innovative” beekeeping method that retains drone brood, induces a brood interruption by caging the queen in summer for 25 days and a single oxalic acid treatment is carried out when the hive is brood-free to reduce the increased mite infestation.

To compare infestation dynamics, we monitored *Varroa* loads throughout the year using sticky bottom boards. Foragers of both beekeeping methods were sampled at three critical time points in spring, summer and autumn. These samples were analysed for pathogen loads via high-throughput qPCR, targeting 18 honeybee pathogens. We did not find any differences in pathogen loads after overwintering between the two beekeeping methods. In summer, shortly before treatment, innovatively managed honeybees exhibited significantly higher pathogen loads than conventionally managed colonies. However, the innovative summer treatment effectively reduced pathogen loads before overwintering.

In conclusion, high *Varroa* infestation in summer significantly increased pathogen loads in innovatively kept honeybees. Nevertheless, an efficient biotechnical *Varroa* treatment successfully reduced mite and pathogen loads, ensuring colony health during the critical phase of winter bee production.

Keywords: *Varroa destructor,* beekeeping, brood interruption, Varroa treatment, pathogens

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V5.6 Flachbettscanner als Werkzeug zur nicht-destruktiven Brutüberwachung in *Apis mellifera* Kolonien

Repurposing Flatbed Scanners for Non-Destructive Brood Monitoring in *Apis mellifera* Colonies

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The health and productivity of *Apis mellifera* colonies are closely linked to effective monitoring of brood health and pest dynamics. While current monitoring methods, such as manual cell inspections, are still widely used, they have significant limitations. These approaches are labour-intensive and provide only momentary snapshots of colony conditions, as inspected cells are destroyed and cannot be observed over time.

To address these challenges, we developed a cost-effective approach utilising contact image sensor technology (Borlinghaus et al. (2024), *Smart Agricultural Technology 9:*100655). Specifically, we integrated an off-the-shelf flatbed scanner into a brood frame, along with a lightweight control unit. A 3D-printed comb structure was attached to the scanner surface, providing a stable foundation coated with a thin layer of wax to encourage adoption by the bees.

In a three-month pilot study using drone-sized meshes, we captured 1.1 million cell images of cell interiors through transparent cell floors. We observed the oviposition of 511 eggs, 58% of which were removed. Additionally, 30 *Varroa destructor* mites were recorded invading the cells, of which 12 successfully reproduced, 12 were removed by worker bees, and 6 failed to reproduce. One visible case of *Ascosphaera apis* infection was removed nine days after the larva hatched.

Mites were commonly observed stuck in larval food for hours while waiting for cell capping, making them easily detectable shortly after invading the cell. This observation suggests high mite detection rates. By collecting high-quality data over three months in vivo without signs of disturbance, we conclude that contact image sensor technology offers a scalable, minimally disruptive tool for accurately assessing brood health, including various pests and pathogens, as well as breeding traits such as Varroa-sensitive hygiene.

Keywords: Honey bee brood monitoring, Varroa destructor monitoring

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V5.7 Mikrosporidischer Parasit beeinträchtigt die Fitness von *Bombus terrestris* Kolonien

Microsporidian parasite impairs colony fitness of *Bombus terrestris*

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Collaborative brood care by workers and production of male sexuals are key to social insect colony fitness. Host shifted parasites may impact theses key features in novel hosts. Microsporidian endoparasites *Nosema ceranae* (NC) of eastern honeybees. *Apis cerana* may be linked to declines of novel wild bumblebee host populations. However, the possible impact on bumblebee colony fitness is poorly understood. Here, we show that NC infections can negatively affect female and male tokens of *Bombus terrestris* colony fitness. In the laboratory, workers and drones were exposed to the parasite or not. Then, infection rates and loads, as well as lethal and sublethal parameters were assessed. In infected workers, hypopharyngeal gland acini, which are crucial for collaborative brood care, were significantly reduced by 3.54%, compared to controls ($P < 0.001$). Further, exposed drones exhibited a 58% higher susceptibility to infection ($P = 0.018$), and 26.3-fold increased spore titers, compared to exposed workers ($P < 0.001$), suggesting sex specific differences in susceptibility. Moreover, in exposed drones, mortality was significantly increased by 22% ($P = 0.027$) and total living sperm reduced by 20% ($P = 0.029$), compared to controls. Taken together, our results suggest a significant impact of NC infections on collaborative brood care and male sexuals, which are cornerstones of bumble bee colony fitness. Given that our laboratory findings hold true for the field, this is calling for respective targeted mitigation.

Keywords: Bombus, immunology, spillover, sperm, hypopharyngeal glands

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V5.8 Auswirkungen von *Vespa velutina* auf *Apis mellifera* Völker in einer neu besiedelten Region in Hessen, Deutschland

Impact of *Vespa velutina* for colonies of *Apis mellifera* in a recently colonised region in Hessen, Germany

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The Asian hornet, *V. velutina*, was observed for the first time in Hesse in 2019. Since then, this invasive species has spread from south to north. The EIP-Agri project 'Vespa velutina Hessen' addressed the issue to generate data on its impact on bee colonies (*A. mellifera*) during the initial phase of its spread. Distribution data was obtained from the official HLNUG portal (www.hlnug.de). In 2019, two individuals and zero nests were reported. Five years later, in 2024, 1107 individuals and 280 nests were recorded. To determine nest size, 52 nests from 2023 were measured. The monthly mean number of combs per nest from August to December was 3.6; 5.25; 6.5; 6.9 and 8.5. The respective numbers of the sealed brood cells were 376; 1355; 2001; 465 and 371. Sizes of nests were comparable to that reported from the Iberian peninsula. In order to describe the stress level of the bee colonies, *V. velutina* appearing in front of the hives were counted on ten bee yards during 30 minutes for 10 weeks in 2023. Single digit numbers of hornets were observed until the end of November with a one-off maximum of 32 individuals. A total of 87 bees were captured (max: 17 bees per 30 minutes once). The bee colonies did not show any clear reactions. In 2024, the next question was whether the hornets were harming the bee colonies. Six colonies were set up in a *V. velutina* area and compared with control colonies in an almost *V. velutina*-free apiary. The data showed no evidence of an effect of *V. velutina* on colony strength (cross level interaction site x time; p=0.793; lin. mixed effects model, SPSS). It should be noted that reported nests were removed during the observation period in accordance with legal requirements.

Conclusion: *V. velutina* is a predator of Hessian bee colonies. However, at this early stage of colonisation, its impact on colonies was not significant.

Keywords: *Apis mellifera*, *Vespa velutina*, stress, damage, invasive

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Poster

Session 1: Ökologie, Wildbienen, Bestäubung

P1.1 The bare necessities of a ground nesting bee species

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Ground-nesting bees constitute the majority of all wild bee species and provide several crucial ecosystem services. This important functional group is especially endangered by the loss of nesting opportunities due to urbanization, land management intensification and eutrophication of landscapes. Despite their importance, ground-nesting bees are understudied, as most research focuses on cavity nesting bees, which are often easier to handle. Especially knowledge about nesting requirements of ground nesting bees is lacking, with most nest site descriptions being vague and not data-based. Species-specific nest site descriptions are crucial to understand how ground-nesting bees can be promoted.

In our study, we conducted precise measurements of relevant soil parameters at 27 nesting aggregations of the oligolectic mining bee species *Andrena vaga* within the city of Braunschweig and compared them to uncolonized control areas, where no bee nests were found. As reported by other studies, the bees nested in sandy soils, but also loamy sand and sandy loam. We identified the proportion of bare ground, soil (surface) temperature and compaction as well as the water content to be the main factors distinguishing nesting sites from uncolonized control areas. However, the causal relationships of soil characteristics are unclear, with ground-nesting bees potentially increasing proportions of bare ground, soil loosening and water infiltration. Further research is needed to disentangle effects of single parameters. Still, our results allow conclusions, how public places like parks, cemeteries or roadsides can be managed to provide suitable nesting sites for ground-nesting bees.

Keywords: nest site selection, autecology, solitary bee, wild bee, ground nesting bee

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zurück zum Programm

P1.2 Wie beeinflusst die Honigsorte die Eigenschaften von Met?

How does the type of honey affect the properties of mead?

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The production of mead has a long tradition in beekeeping and offers a good opportunity to increase the product diversity of beekeeping businesses. Different types of honey are used for mead production and fruit juices are often added to influence the taste and colour of the mead.

We produced mead with three different types of honey using an identical recipe under the same environmental conditions. At the end of fermentation, the mead was drawn off from the yeast sediment and stabilized by adding 60 mg/L sulphurous acid. The products produced were tasted and evaluated by 46 people (participants in a beekeepers' forum who had not received any special sensory training or instruction) without any further addition of sweetness or acidity in form of honey or acid.

The mead produced from the honey variety „flower honey with lime“ was the most popular, followed by the mead produced from „honeydew honey with melezitose“. The mead made from „honeydew honey“ received the lowest score.

In a further step, the participants were asked to select up to 12 attributes (CATA method). The attribute “fruity” was chosen most frequently for all mead variants. This was followed by the attributes “lemon” and “woody” for mead made from flower honey with lime, “flowery” and “woody” for mead made from honeydew honey and “woody” for mead made from honeydew honey with melezitose, followed by “flowery” and “malty”. The differences between the types of honey are also apparent after processing into mead and can be perceived and described through sensory perception. Despite a melezitose content of 18 percent of the total sugar spectrum, honeydew honey with melezitose was successfully fermented into a tasty mead.

Keywords: diversification, melezitose, citizen science, sensory evaluation

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P1.3 *Vespa velutina* in Bayern – Dokumentation der Besiedlung

Vespa velutina in Bavaria – documentation of the colonization

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In 2022, *Vespa velutina* was detected for the first time in Bavaria. It was a single drone in northern Bavaria. In 2023, five sightings of individuals at different locations were reported and all nest sites were identified and the nests destroyed. In 2024, there were sightings at a total of 38 locations. Of these, seven founding queens were observed and killed early in the year. Over the course of the season, 18 nests were found, 17 of which were removed. *Vespa velutina* workers were reported at a further 13 sites, but the nests were not found. Some nests were examined by counting the adult individuals (workers, queens and drones). The results show a good development of the nests under local conditions depending on the time of nest removal. The nests should be removed before mid-September to interrupt reproduction and prevent the nests from producing queens.

Keywords: Vespa velutina, velutina, asiatische Hornisse

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zurück zum Programm

P1.4 Melezitosehonig: Einfluss auf die Mortalität von Honigbienen und Honigqualität

Melezitose honey: effect on honey bee mortality and honey quality

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In summer 2024, many beekeepers faced the challenge of crystallized honey in the honey combs. Honey with a melezitose content above 10% crystallises rapidly and is difficult to extract. Some beekeepers used a wax melter for harvest, thus exposing the honey to an elevated temperature for a prolonged period of time. We studied the influence of the temperature and duration on honey quality. We measured the enzymatic activities (invertase and diastase) and the hydroxymethylfurfural (HMF) content of two honeys with a melezitose content of 12g/100g and 14g/100g, respectively, before and after heating in a water bath at 90°C for three or six hours. Invertase activity was absent after three hours and diastase activity after six hours. Meanwhile, the HMF content increased from below the limit of quantitation to 17 mg/kg and 32 mg/kg after three hours and to 64 mg/kg and 130 mg/kg after six hours, respectively. We also analysed two honeys obtained from wax melters. Honeycombs were melted for 20 minutes at 98°C in a steam wax melter or for five hours at 90°C in a cappings wax melter. The invertase activity from originally 162 U/kg decreased to 21 U/kg and 34 U/kg in the honeys. In contrast, diastase activity was less affected (decrease in Schade units from 22 to 19 and 16, respectively) and the HMF content remained below 2 mg/kg. The loss of invertase activity shows that exposing honeys to heat, e.g. in a wax melter, can affect its quality.

We also tested the effect of melezitose honey on the mortality of honey bees. Fifty bees in cages were fed with melezitose honeys from a steam and a cappings wax melter. As controls served a sucrose solution and a honeydew honey of the previous year (no melezitose). Our results confirmed that melezitose honeys lead to reduced survival rates when compared to a sucrose solution, similar to the rates of bees fed with honeydew honey. Melezitose and honeydew honeys are not suitable for feeding colonies before the winter, but may be used during the spring and early summer season

Keywords: honey bees, melezitose, invertase, diastase, HMF

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P1.5 Empfehlungen für die Fangdauer – Der Einfluss der Farbschalenexposition auf die erfasste Bienengemeinschaft und den Beifang

Best practice for trapping duration - The impact of pan trap exposure on bee samples and bycatch

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Although pan traps are an established method for sampling bees across a wide range of habitats and geographical regions, uncertainty persists as to how pan-trap characteristics influence sampling results.

We investigated the effect of pan-trap exposure (24h, 48h, 72h), on sampled bee communities and bycatch on agricultural sites in Germany, using fluorescent blue, white and yellow pan traps, and DNA metabarcoding for analysing taxonomic diversity (barcode identification numbers, BINs).

Based on 4963 collected bee individuals, we observed interacting effects of exposure and temperature on the number of collected bee individuals, on the number of sampled bee taxa, and on the number of sampled insect taxa without bees. Irrespectively of temperature, exposure also had an effect on bycatch biomass (sampled biomass without bees).

Sampled bee abundance increased from 24h to 48h exposure (Negative binomial GLMM & Tukey test, df = 63, $p < 0.05$), and at cooler temperatures (T_{median} = 13-15°C), sampled bee taxon richness did so as well (Poisson GLMM & Tukey test, df = 64, $p < 0.01$), while we observed no differences between 48h and 72h exposure ($p \geq 0.215$). In contrast, bycatch biomass increased from 48h to 72h exposure (Tweedie GLMM & Tukey test, df = 61, $p < 0.001$) and, at warmer temperatures (T_{median} = 15-18°C), bycatch taxon richness did so as well (Poisson GLMM, df = 64, $p \leq 0.004$).

Based on our results, we advocate for a pan trap exposure of 48h in order to maximize trap efficacy in terms of sampled bee individuals and sampled bee taxa, while limiting unnecessary bycatch. Especially in studies focussing on bee taxon diversity and limited by time allocated to sampling, 24h exposure may be an alternative option that further reduces bycatch.

The presented study was financed by Julius Kühn Institute funds being part of 'Farmland biodiversity Monitoring' (MonVIA), which is funded by the German Federal Ministry of Food and Agriculture.

Keywords: bee sampling, pan trap exposure, biodiversity monitoring

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P1.6 Auswirkungen von Schmelzverfahren zur Honiggewinnung auf die Honigqualität

Effects of melting processes for honey extraction on honey quality

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Melting devices are often used to extract honey crystallized due to a high melezitose proportion from honey combs. According to the manufacturers of these devices, honey quality is not considerably affected by the melting process. To check this, we compared the quality parameters saccharase activity, diastase activity, and hydroxymethylfurfural (HMF) content in honeydew honeys obtained either by common honey extractors (centrifugation) or melting devices with appropriate non-parametric tests.

For Honey extraction by melting beekeepers used hot air capping wax melters (n = 49) or direct wax melters with a heated grid (n = 9). Both melting methods resulted in significantly reduced saccharase activity compared to centrifuged honeys (n = 105). Here, the direct wax melters produced the highest thermal damage ($p < 0.001$). Significant quality losses could also be observed regarding diastase activity ($p = 0.003$) and HMF content ($p = 0.003$) in honey harvested with melting devices.

Among the honeys extracted with capping wax melters we found considerable effects of thermal insulation of the melting devices. Thermal insulation (n = 22) resulted in lower saccharase activity in the honey compared to not insulated devices (n = 24, $p < 0.001$).

Furthermore, reheating of melted honey by melitherm (n = 18) led to an additional reduction in saccharase activity (without melitherm: n = 49, $p = 0.036$).

To produce honey of acceptable quality using a melting process, non-insulated hot air capping wax melters should be used. With insulated melters, the honey must be drained from the device swiftly to cool down. Direct wax melters were not at all suitable for melting melezitose honeycombs.

The requirements of the German honey ordinance or the German Beekeeper Association (DIB) were met by all or 79% of the melted honey samples, respectively. However, the usually reduced quality of melted honeys should be considered when choosing the best before date.

Keywords: melezitose, capping wax melter, direct wax melter, honey quality

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zurück zum Programm

P1.7 Von der Kristallisation bis zum Ausschmelzen – Auswirkungen temperaturabhängiger Extraktionsverfahren auf melezitosereiche Honige

From crystallisation to liquification – effects of temperature-dependent extraction processes on trisaccharide melezitose rich honeys

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The occurrence of 'cement honey' due to melezitose in honeydew is one of the well-known challenges of beekeeping. Beekeepers often feel excited about what appears to be a large honey harvest, only to find the unextractable 'cement honey' in the combs when it comes time to harvest. Unable to extract the honey with centrifugal force in an extractor, beekeepers lose out on large honey harvests. Out of necessity, many beekeepers try to treat their 'melezitose combs' with heat in order to separate the honey from the wax. What is the impact on the quality of these honeys and do they still meet the required quality parameters of the German honey regulation (HonigV)?

In order to understand the consequences of heat treatments, we collected honeys from beekeepers in Baden-Württemberg, who extracted honeys with such melting processes, and compared the quality parameters with the untreated honeys. A total of 75 honey samples were analyzed, including 55 heated and 20 unheated honeys. Although heating had a detectable effect on HMF content and diastase- and invertase-activity ($p<0.05$), all honeys still complied with the legal requirements according to the German HonigV. The diastase-activity of all heated honeys was above the defined minimum activity of 8 DZ and the HMF content was below the defined maximum value of 40 mg/kg. 23 honeys (42%) of the heated samples showed a reduced invertase-activity below 64 U/kg and thus no longer complied with the DIB guidelines. Due to this reduced invertase-activity, an enzyme that is very heat sensitive, these honeys cannot be marketed under the DIB brand, and only sold in neutral honey jars. Due to their unique properties, honeys differ in their susceptibility to heat damage and therefore no clear correlation could be found between temperature, duration of heating, or a combination of these factors and the severity of heat damage (HMF and enzyme activity) within the heated honeys ($p>0.05$).

Keywords: honeydew, melezitose, honey quality, temperature effects

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P1.8 Verbesserte taxonomische Auflösung von Honig mit DNA-Metabarcoding

Enhanced taxonomical resolution with DNA-Metabarcoding of honey

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Classical microscopical pollen analysis (MPA) enables the identification of pollen origins in honey, typically at the genus or family level. It also allows the detection for honeydew elements. However, DNA metabarcoding offers a more comprehensive approach to assessing biological diversity in honey. This molecular technique allows for the simultaneous detection of multiple taxa within a single sample. The method involves extracting insoluble honey components, isolating DNA, amplifying specific barcode regions, and sequencing them. A DNA barcode is a short gene fragment that varies across taxa and enables species-level identification by comparison with reference libraries. For eukaryotic identification, particularly of plants, the Internal Transcribed Spacer (ITS) regions are well-established markers.

In this study, 55 honey samples were analyzed using the ITS1 and ITS2 barcode regions. More than 200 plant species from 33 families were identified, providing a higher taxonomic resolution than conventional MPA. This improves the assessment of beehive location quality and enhances the evaluation of agricultural measures. Moreover, metabarcoding based on ITS barcodes enables the detection of non-plant eukaryotic DNA in honey, allowing for species-level identification of honeydew elements such as mold hyphae and spores, as well as unicellular algae. Additionally, DNA from other fungal taxa was detected, including yeasts and the causative agent of Chalkbrood disease.

These findings highlight the potential of metabarcoding as a powerful tool for honey biodiversity analysis. However, future research should focus on methodological validation, refining bioinformatics pipelines, and standardizing analytical workflows to enhance reproducibility and accuracy.

Keywords: Honey, biodiversity, botanical origin, ITS, metabarcoding

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P1.9 DNA-Metabarcoding pflanzlicher und tierischer DNA in Honigtauhonigen

DNA metabarcoding of plant and animal DNA in honeydew honeys

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The use of DNA methods to improve honey analysis is heatedly debated in current bee journals, the media and scientific publications. There are already companies that market DNA analysis for blossom honeys that use the plant DNA found in honey to infer the foraging preferences of bees. In honeydew honeys, bees collect the excreta of phloem-sucking hemipterans instead of nectar. Thus, it seems probable that the DNA of these animal honeydew producers could be used to identify the varietal of honeydew honey. In a preliminary study, three spruce honeys, one fir honey and one 'forest honey with fir' were analysed. From the latter, several DNA samples were extracted to determine the repeatability of the method.

DNA was extracted from these honeydew honeys and DNA metabarcoding was carried out for both plant and animal DNA. These DNA spectra were then compared with the results from current standards of honey variety identification, in particular, microscopic analysis of the plant pollen in the sample. In the plant DNA spectrum, the genera *Trifolium*, *Rubus* and *Plantago* each accounted for the largest proportion of reads (together over 50% of the total DNA). The Fagaceae family accounted for a significantly lower proportion when analysing the plant DNA than when counting the pollen. Although plant DNA metabarcoding and pollen analysis qualitatively detected the same plants, they differ substantially in their quantitative weighting. The proportion of the most important honeydew plants, *Picea spp.* and *Abies spp*., did not exceed 0.3% of the total DNA in any of the samples.

In the metabarcoding of animal DNA, both the quantity and the quality of the DNA were significantly increased compared to previous experiments. Also in spruce honeys, the majority of the detected Hemiptera-DNA came from phloem-sucking species living on fir trees. These preliminary results seem promising and additional DNA analysis of a larger suite of honeydew honeys will be conducted.

Keywords: honeydew, dna metabarcoding, pollen analysis, varietal honey identification

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P1.10 Professional beekeeping project SIB2023-012

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Professional beekeeping plays a vital role in agricultural pollination and crop yield enhancement. This research project aimed to develop science-based innovations to optimize bee health and pollination services through targeted studies on nutrition, pheromones, Varroa management, and greenhouse pollination. The study was structured into five research modules. Module 1 evaluated pollen substitute diets for sustaining brood development, testing seven commercial products alongside a pollen control group, with three products performing relatively well. Module 2 explored the efficacy of synthetic pheromones in greenhouses, where application to 12 colonies showed a trend toward lower bee losses over one month. Module 3 assessed current Varroa control methods, resulting in a comprehensive report to inform beekeepers. Module 4 examined the impact of greenhouse materials and LED lighting on bee navigation and foraging, providing mitigation measures for beekeepers and crop growers. Module 5 presented an in-depth analysis of the pollination service market, identifying strengths, opportunities, weaknesses, and threats. The findings were shared with beekeepers to promote healthier bee colonies and improve pollination efficiency, ultimately benefiting both professional and hobbyist beekeepers while enhancing agricultural sustainability.

Keywords: Pollination, Professional Beekeeping, Pollen Substitutes, Artificial Pheromones, Varroa Mite Control

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Session 2: Physiologie & Verhalten

P2.1 Wie beeinflusst die Honigsorte die Eigenschaften von Met?

How does the type of honey affect the properties of mead?

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The production of mead has a long tradition in beekeeping and offers a good opportunity to increase the product diversity of beekeeping businesses. Different types of honey are used for mead production and fruit juices are often added to influence the taste and colour of the mead.

We produced mead with three different types of honey using an identical recipe under the same environmental conditions. At the end of fermentation, the mead was drawn off from the yeast sediment and stabilized by adding 60 mg/L sulphurous acid. The products produced were tasted and evaluated by 46 people (participants in a beekeepers' forum who had not received any special sensory training or instruction) without any further addition of sweetness or acidity in form of honey or acid.

The mead produced from the honey variety „flower honey with lime“ was the most popular, followed by the mead produced from „honeydew honey with melezitose“. The mead made from „honeydew honey“ received the lowest score.

In a further step, the participants were asked to select up to 12 attributes (CATA method). The attribute “fruity” was chosen most frequently for all mead variants. This was followed by the attributes “lemon” and “woody” for mead made from flower honey with lime, “flowery” and “woody” for mead made from honeydew honey and “woody” for mead made from honeydew honey with melezitose, followed by “flowery” and “malty”. The differences between the types of honey are also apparent after processing into mead and can be perceived and described through sensory perception. Despite a melezitose content of 18 percent of the total sugar spectrum, honeydew honey with melezitose was successfully fermented into a tasty mead.

Keywords: diversification, melezitose, citizen science, sensory evaluation

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zurück zum Programm

P2.2 Entwicklung einer klimaschonenden und ökonomisch resilienten imkerlichen Betriebsweise

Development of a climate-friendly and economically resilient beekeeping practice

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The honey bee *Apis mellifera* is a key pollinator, but climate change, inflation, and rising energy costs threaten beekeeping. We developed a more resilient management approach and compared it to standard practices, focusing on colony health, energy use, labor, material costs, and climate impact.

In 2023–2024, 34 colonies were divided into 'Standard' and 'Resilient' groups. The standard group underwent regular drone brood removal and swarm control. Normal sized split colonies were formed with two brood combs and transported to a new apiary in 3 km distance. *Varroa destructor* was managed using formic acid (60% Nassenheider professional) in summer and oxalic acid in winter.

In the resilient group, drone brood remained, and 5–8 capped brood combs were removed in early summer (end of April/beginning of May) to build strong split colonies which remained at the same apiary. This suppressed swarming and relieved the colonies of a considerable proportion of the *V. destructor* mites, which are mainly found in the capped brood cells. Queens were caged for 25 days in summer, followed by oxalic acid trickling; winter treatment was only applied if mite fall exceeded two per day. Colony development was tracked using the Liebefeld method and we recorded weight change, honey yield, bee longevity, fat body reserves, mite infestation, and management time.

Honey yield and overall colony strength did not differ significantly between groups. However, splits from resilient colonies developed stronger and required less feed. Management time per colony averaged 53 minutes in the resilient group vs. 2 hours 10 minutes in the standard group. Fewer apiary visits also reduced climate impact. Interestingly, the total weight lifted by the beekeeper throughout the year decreased from 1059.7 kg per standard colony to 605.4 kg per resilient colony.

Keywords: resilient beekeeping, honey bee vitality, climate change, *Varroa destructor,* honey yield

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zurück zum Programm

P2.3 Bewertung einer neuartigen modularen 3D-gedruckten Pollenfalle für das Hummelmonitoring: Effizienz und Anwendungspotenzial

Evaluating a Modular 3D-Printed Pollen Trap for Bumble Bee Monitoring: Performance and Application Potential

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Accurate pollen collection is crucial for assessing bumble bee foraging behavior, monitoring colony health, and evaluating environmental risks. This study introduces and evaluates a newly developed modular 3D-printed pollen trap (JKI trap) compared to the USDA 3D-printed pollen trap. Field trials with Bombus terrestris colonies tested the efficiency of both trap designs, using two entrance diameters (6.5 mm and 7.2 mm). Results demonstrated that the JKI trap collected, on average, 65% more pollen than the USDA trap, with no significant influence of entrance diameter. The modular design of the JKI trap allows for species-specific adjustments, enhancing its applicability across different Bombus species and potentially other pollinators. These findings highlight the potential of optimized pollen traps in ecological monitoring, floral resource assessments, and pesticide risk studies.

Keywords: Bumble bee monitoring, pollen trap efficiency, *Bombus terrestris*, ecological monitoring, 3D printing

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P2.4 Der Einfluss von Acetylcholin auf die Hormonproduktion und das Verhalten von *apis mellifera*

The Influence of Acetylcholine on Hormonal Regulation and Behavior in *apis mellifera*

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Interfering with the bee's cholinergic system, for example through pesticides such as neonicotinoids, negatively affects bee development. However, the underlying mechanisms remain unclear. The development of bees is primarily subject to hormonal regulation but if and how hormone homeostasis is affected by the cholinergic system is yet to be shown. The maturation of nurses to foragers is influenced by juvenile hormone in a crucial manner. The aim of this study is to investigate the possible influence of acetylcholine on the internal hormone production. To explore this, the effect of acetylcholine on the sucrose responsiveness will be examined, as studies suggest that the responsiveness to sucrose reflects hormone levels and the respective role of the bee as nurse or forager. Preliminary results indicate that the presence of acetylcholine increases the elicited proboscis extension reflex by approximately 35%, suggesting a positive impact on sugar responsiveness. This implies that bees might be able to detect acetylcholine which requires further investigation. In future experiments, I will examine whether the corpora allata, the hormonal gland responsible for juvenile hormone production, is regulated by cholinergic signaling and whether its activity can be influenced by the oral administration of acetylcholine.

Keywords: sucrose-responsiveness, PER, acetylcholine, juvenile hormone, corpora allata

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zurück zum Programm

Session 3: Genetik & Zucht

keine Beiträge

Session 4: Bienenschutz & Pflanzenschutz

P4.1 Die Suche nach dem Triforce – Mechanismen zur Regulierung der Interaktionen zwischen Bienen, Blüten und Bakterien

Finding the Triforce – Mechanisms regulating interactions of bees, blossoms, and bacteria

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The phylloplane microbiome includes beneficially associated microorganisms and plant pathogens. *Erwinia amylovora*, the causative agent of fire blight disease, is a pathogen that colonises flowers of pome fruit trees, which causes devastating agricultural losses in Europe and the United States. Some beneficial plant-associated bacteria are known to produce contractile phage tail-like particles (CPTPs), named tailocins, which can specifically kill competing bacteria by punctuating the cell membrane and thus disrupting its vital proton motive force. The project ABBAonFire uses an approach based on natural tailocin-producing antagonists (TPAs) with emphasis on non-target organisms and the environment. ABBAonFire aims to search for natural TPAs as a control concept against *E. amylovora* in organic farming. As bees transmit the fire blight pathogen by visiting flowers, entomovectoring also offers an approach to distribute TPAs. However, the introduction of foreign TPAs into the environment harbours the risk of negatively affecting pollinators and their microbiome. The core gut microbiome of bees is highly conserved and fine-tuned. A well-studied mechanism to mediate interspecific interactions in the digestive system of bees are type VI secretion systems (T6SS), a subgroup of CPTPs. However, tailocin-driven interactions of bee gut bacteria, consumed plant-associated bacteria, and pathogens remain unknown. Here, we combine predictions of the PlaBAse web resource, to detect specific tailocin clusters within bacterial genomes and antagonism assays after inducing tailocin expression in bacterial strains of interest. Furthermore, potential off-target effects of candidate TPAs on pollinators have to be excluded by using bioinformatics analyses and standardised lab and (semi-)field assays.

Keywords: tailocins, fire blight, bee gut microbiome, interspecific competition

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P4.2 Analyse von Einflüssen des Klimawandels auf die Imkerei und Entwicklung von Handlungsoptionen für die Imkerschaft

Analysis of influencing factors of the climate change on apiary and development of recommended procedures for beekeeping

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In this project we analyse the influence of aridity and high temperatures caused by the climate change on *Apis mellifera* at the colony and individual level. And we investigate possible solutions for practical beekeeping to counteract potential negative effects.

To investigate the impact of the currently observed heat and drought conditions on the development of *Apis mellifera*, these conditions were simulated in laboratory experiments. Subsequently, various parameters such as body size, weight or ingested amount of sugar water were measured on *Apis mellifera*. The younger the developmental stage of the brood at the time of heat stress, the smaller (one-factor ANOVA; $p = 0{,}001$) and lighter (Kruska-Wallis; $p = 0{,}001$) they were when hatched, and therefore the less sugar water they consumed (Kruska-Wallis; $p = 0{,}001$). The effects of drought and heat on colonies were investigated by exposing colonies to different heat conditions. Among other things, factors as collecting behaviour and pathogen load were recorded. A difference between the individual colonies however could not be found in the foraging behaviour (x^2; $p > 0{,}05$) or the pathogen load. In another laboratory experiment we also investigated if regularly repeated heat stress can impact the life span of *Apis mellifera* workers. And indeed, the group without any heat stressing lived significantly longer (log-Rank; $p = 0{,}005$).

Building on the results so far, further experiments will be conducted, among other things, regarding heat stress during development but with different temperatures or altered heat conditions in populations.

Keywords: climate change, *Apis mellifera*, beekeeping, temperature, humidity

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P4.3 Auswirkungen von Kupfer auf die Entwicklung und das Überleben von in-vitro aufgezogenen Honigbienenlarven (*Apis mellifera*)

Effects of copper on the development and survival of in-vitro reared honey bee larvae (*Apis mellifera*)

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In this study we investigated the effects of different copper (Cu) concentrations on the larval development of honey bees (*Apis mellifera*), including larval feeding time and their survival probability, emergence rate and their post-emergence longevity. To better understand the effect of a single heavy metal pollutant (in this case Cu) on honey bee larval development, we performed an in-vitro study, representing a controlled rearing environment. Four increasing Cu concentrations (0.5, 5, 25, 50 mg Cu/L) were added to the artificial diet of larvae and impacts on development compared with a control diet lacking Cu. The highest Cu concentration in the diet led to a significantly longer feeding time ($p < 0.001$, $N = 904$). As of the 5 mg Cu/L concentration, larval survival decreased significantly with increasing Cu concentration ($p < 0.001$, $N = 960$). The emergence rate of the bees was negatively influenced when larvae were fed 50 mg Cu/L ($p < 0.001$, $N = 960$). In a post-emergence trial, the bees were placed in cages with sucrose syrup and pollen paste and their longevity was monitored to understand if Cu exposure during development had negative long-term impacts on adult bees. Adult longevity decreased significantly with increasing Cu concentrations in the larval diets ($p < 0.001$, $N = 682$), although there was no further Cu exposure post emergence. Overall, these in-vitro studies revealed that Cu had negative impacts on larval development and the effects of this heavy metal exposure during development carried into post emergence adulthood. Further laboratory and field studies investigating the long-term impacts on the individual level and its potential impacts on social behavior at the colony level should be conducted, as copper-based fungicides are intensively used in organic and conventional agriculture.

Keywords: copper toxicity, in-vitro rearing, larval development, post-rearing survival, *Apis mellifera*

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P4.4 ZUFI - Zukunftsfähige Imkerei Bayern – Professionalisierung des Imkereisektors durch betriebswirtschaftliche Entscheidungshilfen für wachstumswillige Betriebe

ZUFI - Sustainable Future of Bavarian Beekeeping Sector – Support and improve economic efficiency and investment-decision-making of beekeepers with growth perspective

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German and bavarian beekeeping sector are characterized by over 96 percent small non-professional apiarists with an overall average of approx. 7 colonies per beekeeper only (EU average is at approx. 21). The Bavarian State Ministry for Food, Agriculture, Forestry and Tourism strongly supports beekeepers willing to expand their operations. Project ZUFI has been setup by the Ministry in order to support decision-making of beekeepers when trying to identify the proper size and specification of required equipment or space or production process for their growing operations. Therefore a database has been designed which collects information on process steps of current good beekeeping practice with the appropriate setup of beekeeping equipment and required operating space. Information has been sourced from experienced beekeping advisors and professional beekeepers as well as using a Citizen Science approach by analyzing surveys on specific beekeeping processes (e.g. honey processing, queen rearing and breeding) offered to the beekeeping community. Using a search algorithm, the database will select recommended equipment and space characteristics for defined production steps, including advantages and disadvantages of certain equipment, safety aspects and required power supply besides the required acqusition cost and depreciation in relation to the expected production output.

Project ZUFI contributes to digitalization of information on beekeeping processes and corresponding ressource requirements and safeguards proper decision making for expanding beekeeping operations

Keywords: sustainability, beekeeping processes, citizen science, digitalization

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P4.5 Untersuchung von Bienenvergiftungen unterstützt durch Satellitenaufnahmen, die mit Webdiensten bereitgestellt werden

Supporting investigation of bee poisoning incidents with satellite imagery provided by web services

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The *Untersuchungsstelle für Bienenvergiftungen* of the Julius Kühn-Institute investigates bee poisoning incidents caused by misuse of plant protection products (PPP) and analyses dead bees and potentially contaminated plants for PPP residues. If a poisoning of *Apis mellifera* occurs, the search of the contaminants origin and sampling of plant material may be complicated even if a specific crop is suspected, as worker bees have a foraging area of many square kilometres. Hence, to support orientation and plant sampling, a web-based framework was developed in the project Sen2Bee for integrating and presenting information of the surrounding of the affected bee colonies. Further, this will enable a more detailed investigation and interpretation of the incident and its circumstances. Up-to-date and retrospective information on crops and their flowering period can be provided by products based on the Sentinel-2 satellites of the Copernicus Programme, which revisits the same location every 5th day. True-colour images from those satellites provide initial orientation in the landscape and we are visualising the flowering of crops, fruits and other sites based on the satellites' spectral bands. Additionally, spatial information of air temperature and further weather elements inside of the assumed flight radius is provided by other web services, which builds on data of a DWD climate model. Potentially, further web services could be added, such as specific information of crop types derived from satellite imagery. A great advantage of the web application will be that both up-to-date and also firstly unnoticed and later detected potential PPP incidents can be better interpreted. Further, identification of locations for plant sampling is facilitated.

Keywords: honey bees, plant protection products, Sentinel-2, Copernicus Programme, bee poisoning incidents

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Session 5: Bienenpathologie

P5.1 Flachbettscanner als Werkzeug zur nicht-destruktiven Brutüberwachung in *Apis mellifera* Kolonien

Repurposing Flatbed Scanners for Non-Destructive Brood Monitoring in *Apis mellifera* Colonies

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The health and productivity of *Apis mellifera* colonies are closely linked to effective monitoring of brood health and pest dynamics. While current monitoring methods, such as manual cell inspections, are still widely used, they have significant limitations. These approaches are labour-intensive and provide only momentary snapshots of colony conditions, as inspected cells are destroyed and cannot be observed over time.

To address these challenges, we developed a cost-effective approach utilising contact image sensor technology (Borlinghaus et al. (2024), *Smart Agricultural Technology 9:*100655). Specifically, we integrated an off-the-shelf flatbed scanner into a brood frame, along with a lightweight control unit. A 3D-printed comb structure was attached to the scanner surface, providing a stable foundation coated with a thin layer of wax to encourage adoption by the bees.

In a three-month pilot study using drone-sized meshes, we captured 1.1 million cell images of cell interiors through transparent cell floors. We observed the oviposition of 511 eggs, 58% of which were removed. Additionally, 30 *Varroa destructor* mites were recorded invading the cells, of which 12 successfully reproduced, 12 were removed by worker bees, and 6 failed to reproduce. One visible case of *Ascosphaera apis* infection wa removed nine days after the larva hatched.

Mites were commonly observed stuck in larval food for hours while waiting for cell capping, making them easily detectable shortly after invading the cell. This observation suggests high mite detection rates. By collecting high-quality data over three months in vivo without signs of disturbance, we conclude that contact image sensor technology offers a scalable, minimally disruptive tool for accurately assessing brood health, including various pests and pathogens, as well as breeding traits such as Varroa-sensitive hygiene.

Keywords: Honey bee brood monitoring, *Varroa destructor* monitoring

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P5.2 Nachweis von BQCV in epitheloiden Zellen befruchteter Honigbieneneier mittels Fluoreszenz *in situ* Hybridisierung.

Detection of BQCV in epithelioid cells of fertilized honey bee eggs using fluorescence *in situ* hybridization.

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Honey bees (*Apis mellifera*) are important pollinators of agricultural crops, and despite reports of increased local colony losses in recent decades, the number of honey bee colonies has increased worldwide. However, the demand for pollination services has increased disproportionately over the same time period, potentially leading to a global pollination crisis. In this context, colony losses due to pathogens and parasites are particularly alarming. Of particular interest are infections with (+)ssRNA viruses such as Deformed Wing Virus (DWV), Acute Bee Paralysis Virus (ABPV) or Black Queen Cell Virus (BQCV). While the pathogenesis of DWV and ABPV infections is intensively studied by international research groups, research efforts on BQCV have been very limited, although outbreaks of BQCV-infections occur frequently in Germany and worldwide. BQCV is particularly dangerous to queen bees, infecting queen larvae and killing them during development. Therefore, BQCV poses a threat to beekeeping, especially to queen breeding, a very profitable area of beekeeping. BQCV has been shown to be transmitted by several routes, including horizontal transmission from infected worker bees and contaminated food sources. It is also linked to *Nosema apis* and *Nosema ceranae* occurrence and to vector-mediated transmission by varroa mites. BQCV was detected by RT-PCR in the ovaries of honey bee queens and in their eggs. However, if the virus is actually replicating in the eggs has not been shown so far. To understand the pathogenesis of BQCV and to elucidate the routes of transmission, we analysed tissue sections of fertilized honey bee eggs using fluorescence *in situ* hybridisation (FISH) of BQCV positive worker bee eggs. Using BQCV specific probes, we identified BQCV RNA in epithelioid cells, proving replication of BQCV in worker bee eggs. The results of our study will help to further understand the pathogenesis of BQCV infections.

Keywords: virus, BQCV, fluorescence *in situ* hybridization, vertical transmission

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P5.3 Entwicklung und Validierung eines Lateral-Flow-Assays zum Nachweis des Schwarze Königinnenzellen-Virus (BQCV)

Development and validation of a lateral-flow-assay for the detection of black queen cell virus (BQCV)

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The Western honey bee *Apis mellifera* is not only an important pollinator, it also contributes to the conservation of biodiversity and is of high economic value due to its honey production. A variety of different viruses can infect the honey bee and pose a major threat to their health. Black queen cell virus (BQCV) is one of the most prevalent honey bee viruses and can cause larval and pupal mortality during honey bee development. This is particularly problematic in queen rearing facilities and breeding programs when queens fail to develop and die as a result of BQCV infection. Detection of BQCV is currently only possible with reverse transcription polymerase chain reaction (RT-PCR), an expensive and time-consuming lab procedure. A lateral-flow-assay for detection of BQCV would simplify the detection and could allow for colony screening before breeding. To develop a lateral-flow-assay (LFA), specific antibodies are required. Therefore, we recombinantly expressed and purified the capsid protein BQCV-VP2 for the production of monoclonal antibodies. The specificity of the obtained antibodies was tested by dot blots and western blots and the best antibodies were used to construct the LFA. The production of monoclonal antibodies and the construction of the LFA were performed by ASKA Biotech GmbH. The finished BQCV-LFA was validated using larvae, pupae and adult honey bees from infection assays and field samples. The infection status of each sample was determined by RT-PCR and compared with the results of the LFA. The LFA has a specificity of 100 %, hence, does not give false-positive signals in negative bee material, and it is able to detect BQCV in larvae, pupae and adult honey bees.

Keywords: BQCV, viruses, detection, lateral-flow-assay

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P5.4 An experimental infection bioassay to analyze SBV tissue tropism in honey bee larvae

An experimental infection bioassay to analyze SBV tissue tropism in honey bee larvae

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Honey bee viruses have attracted considerable interest over the last two decades and it is becoming increasingly clear that viral infections play an important role in the deterioration of bee health. Among the rather neglected viruses in Europe is sacbrood virus (SBV). This neglect could be due to the fact that, although SBV can be quite frequently detected in adult bees in apparently healthy colonies, it rarely causes symptoms and cannot be linked to colony losses. However, the virus can cause significant damage to honeybee larvae, resulting in the appearance of edema and, in severe cases, larval mortality and colony loss. The lack of basic research on this virus also means that the pathogenesis of SBV infection is not yet understood. In order to change this dire situation and bring attention to this neglected virus, we have developed an exposure bioassay for larval infection with SBV as a first step towards in-depth analysis of this virus. Here, we present data on the development, establishment and validation of this SBV exposure bioassay, and use fluorescence in situ-hybridization (FISH) and immunofluorescence (IF) to analyze SBV tissue tropism.

Keywords: virus, pathogen, protein expression, immunofluorescence

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P5.5 MLVA - eine aussagekräftige Methode für epidemiologische Analysen von *Paenibacillus larvae*

MLVA - a meaningful method for epidemiological analyses of *Paenibacillus larvae*

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American Foulbrood is a disease of honey bee larvae that occurs worldwide and is caused by the Gram-positive bacterium *Paenibacillus larvae* (*P. larvae*). Various methods have been used in the past to classify and genotype this bacterium. These methods include repetitive element PCR (repPCR) performed with the Enterobacterial Repetitive Intergenic Consensus (ERIC) primers, Multi Locus Sequence Typing (MLST) based on seven housekeeping genes, multiple locus VNTR analysis (MLVA) and whole genome sequencing (WGS). The various methods differ in their resolution with regard to genomic differences. The highest resolution is achieved with WGS, but this method is also the most expensive and time-consuming. For this reason, more cost-effective methods are being sought that have a similar level of informative value. The well-established methods repPCR and MLST are not suitable for this purpose. The repPCR distinguishes only five ERIC genotypes, and although the MLST analysis is already much better than the repPCR, the MLST database for *P. larvae* still contains only 48 different sequence types. Furthermore, it shows a questionably low genetic variance for the *P. larvae* genotype ERIC II, indicating an unbalanced selection of housekeeping genes among the ERIC genotypes. Compared to MLST, MLVA should enable a much better resolution of genetic variability. MLVA is a PCR-based method that analyzes the number of tandem repeats (VNTR, variable number of tandem repeats) in different regions of the genome. Within the *P. larvae* genomes, we identified 11 sequence regions, suitable for high resolution genotyping, and optimized the protocol for routine use. Analysis of a total of 1274 isolates resulted in 370 MLVA types. The accuracy of the typing was validated with a subset of 180 fully sequenced strains. We show that the resolution of our MLVA scheme is close to that of WGS and that there are no significant differences in genetic variability between *P. larvae* ERIC I and ERIC II.

Keywords: *P. larvae*, molecular typing, MLVA

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zurück zum Programm

P5.6 Unterschiedlicher Gewebetropismus von DWV-Varianten nach oraler Infektion erwachsener Bienen

Differences in tissue tropism of DWV-variants after oral infection of adult bees

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Deformed wing virus (DWV) is an important pathogen of the Western honey bee (*Apis mellifera*). Four master variants have been described so far, of which only DWV-A and DWV-B are frequently detected. Vectorial transmission of DWV-B by DWV-B-infected Varroa mites (*Varroa destructor*) to honey bee pupae causes severe clinical symptoms, including pupal death, emergence of adult bees with crippled wings, or emergence of healthy-looking bees with DWV infection of the brain accompanied by significant reduction in cognitive abilities. We recently showed that an attenuated form of DWV-B, DWV-B_{att}, can be obtained by passaging DWV-B in honey bee pupae. Using injection bioassays that mimic vectorial transmission by the Varroa mite, we have shown that DWV-B_{att} is less virulent than DWV-B when looking at pupal mortality as well as cognitive performance and neurotropism in adult bees. However, whether such differences in virulence also occur when DWV-B or DWV-B_{att} are administered orally to honey bees was not investigated so far. To simulate horizontal transmission of DWV-B or DWV-B_{att}, as it occurs as part of the social immune response during the cannibalism of severely DWV-infected honey bee pupae, we fed both virus isolates to adult honey bees. Again, DWV was detected significantly more often in the heads of bees fed with DWV-B than in the heads of bees fed with DWV-B_{att}. Our results clearly show that differently virulent DWV variants differ in neurotropism regardless of the transmission route.

Keywords: honey bee virus, pathogen, DWV-B, DWV-B_{att}, tissue tropism

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P5.7 STR-Analyse der Populationsgenetik von *Nosema ceranae* in Argentinien und Deutschland.

STR-analysis of the population genetics of *Nosema ceranae* in Argentina and Germany.

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Nosema ceranae is an obligate intracellular microsporidian parasite of honey bees. Several international reports indicate considerable differences in the prevalence, development and distribution of this pathogen, which is explained by various factors such as climatic susceptibility, the degree of host adaptation or the existence of different genetic variants differing in virulence. In Germany, *N. ceranae* showed an increasing prevalence in the honey bee population over the last 20 years, essentially replacing its congener *N. apis*. This has led to increased scientific interest in the population genetics of *N. ceranae* and its potential impact on colony health. Since typically millions of *Nosema* spores are processed per sample, only co-dominant typing methods, such as those based on short tandem repeats (STRs), are suitable to capture the true species diversity. The aim of this study was to establish an STR based genotyping method for *N. ceranae* and investigated the genetic variance within and between *N. ceranae* populations from different origins. STR based genotyping of samples from Argentina and Germany followed by Agglomerative Hierarchical Clustering (AHC) and Principal Coordinate Analysis (PCoA) revealed different genetic variants within the two countries, but also a clear separation of isolates between the two countries. These results indicated for the first time a clear geographical clustering of genetic variants of *N. ceranae*. The STRs developed in this study represent a significant advancement in the study of the genetic diversity of *N. ceranae*. These results help to fill the knowledge gap in the field of bee pathogen microbiology and open new possibilities for the study of the epidemiology and evolution of *N. ceranae*.

Keywords: *Nosema apis*, *Nosema ceranae*, short tandem repeats, genetic diversity

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zurück zum Programm

P5.8 Die Rolle der Flagellen von *Paenibacillus larvae* während der Pathogenese von *Paenibacillus larvae*-Infektionen in Honigbienenlarven

The role of *Paenibacillus larvae* flagella in the pathogenesis of *Paenibacillus larvae* infections of honey bee larvae

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American Foulbrood (AFB) is among the most detrimental honey bee brood diseases threatening the health of bee colonies worldwide. The causative agent of AFB is *Paenibacillus larvae*, a Gram-positive, peritrichously flagellated, spore-forming bacterium that is classified into different genotypes (ERIC I-V). AFB infection commences when the ingested spores of *P. larvae* germinate within the midgut lumen of first instar honey bee larvae. Subsequent to an extensive proliferation of vegetative bacteria within the gut, *P. larvae* invades the midgut epithelium to subsequently infiltrate the hemocoel, ultimately resulting in the death of the larva. During the infection process, the multiple flagella on the bacterial cell surface presumably play a major role for *P. larvae*, since the synthesis of flagella is an energy-expensive process for bacteria. In this study, we demonstrate that the flagella are essential for full pathogenicity using a mutant of *P. larvae* ERIC II deficient in flagella production in an infection assay with honey bee larvae. Furthermore, we analysed sections of infected larvae via fluorescence *in situ* hybridization (FISH). The FISH results indicate that the flagella of *P. larvae* ERIC II play a role in various steps of the infection process, including the colonization of the larval midgut, the attachment of bacteria to the epithelial cells, and the penetration of the intercellular space of the epithelial cell layer. Furthermore, evidence is presented that during the latter step, *P. larvae* ERIC II employs its capacity for swarming behaviour to collectively migrate through the paracellular route. All of these steps have been previously shown to be pivotal in the pathogenesis of *P. larvae*. Consequently, the *P. larvae* flagellum has to be considered an important virulence factor of this deadly bee pathogen.

Keywords: *Paenibacillus larvae*, flagella, virulence, swarming motility

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zurück zum Programm

P5.9 2,5-Diisopropylpyrazin — ein volatiler Sekundärmetabolit von *Paenibacillus larvae*

2,5-diisopropylpyrazine — a volatile secondary metabolite of *Paenibacillus larvae*

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Paenibacillus larvae, a Gram-positive, spore-forming bacterium, is the etiological agent of American Foulbrood (AFB), a worldwide occurring fatal disease of honey bee larvae. The bacterium is classified into five different genotypes, *P. larvae* ERIC I-V, with ERIC I and II being most frequently isolated from current AFB cases. The genome of *P. larvae* harbours several non-ribosomal peptide synthetase (NRPS) clusters and NRPS/polyketide synthase (PKS) hybrid clusters, which have been previously linked to the non-ribosomal synthesis of various secondary metabolites. These metabolites have been shown to be involved in the interaction of *P. larvae* with microbial competitors, such as bacteria or fungi, to defend its ecological niche, to function as surface-active agents (surfactants), or to sequester iron from the environment.

Within the genomes of *P. larvae* ERIC I and ERIC II, in addition to the NRPS and NRPS/PKS clusters, an additional single NRPS gene has been identified that encodes a cryptic, monomodular enzyme. This minimalist NRPS, designated as StAM (stand-alone monomodular), has recently been assigned to the production of 2,5-diisopropylpyrazine, a volatile compound with a symmetrical structure and hitherto unknown function. Given the hypothesis that this volatile compound could be utilized by *P. larvae* as a quorum sensing molecule, we conducted a study to investigate a potential role of StAM in the multicellular behaviour and virulence of *P. larvae*. To this end, we inactivated the NRPS gene *stam* in *P. larvae* ERIC II and tested the bacterial mutant in swarming and biofilm assays, as well as in an infection assay with honey bee larvae. The results indicate that 2,5-diisopropylpyrazine inhibits swarming motility and extracellular matrix production. However, the deactivation of StAM did not significantly affect the virulence of *P. larvae*, suggesting that 2,5-diisopropylpyrazine may play a more significant role after larval death.

Keywords: *Paenibacillus larvae*, American Foulbrood, secondary metabolites, virulence

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P5.10 Laborvergleichsprüfungen zum Nachweis von *Paenibacillus larvae*: ein Instrument zur Verbesserung der Untersuchungsqualität

Comparative laboratory tests for the detection of *Paenibacillus larvae*: an instrument for improving test quality

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American foulbrood (AFB) is a highly contagious brood disease of honeybees (*Apis mellifera*), which is caused by *Paenibacillus larvae*. In Austria two genotypes of *P. larvae* occur, i.e. ERIC I and ERIC II and for analysis it is crucial to detect both genotypes. For early AFB detection, it is common practice in Austria to take hive samples from food stores or honey. Our department within AGES harbors The Austrian National Reference Laboratory for Bee Health (NRL). Since 2022 this NRL started to offer the possibility to participate in comparative laboratory tests (CLTs) for all laboratories with *P. larvae* analytic services. Since then, three CLTs have been performed. For each CLT, the NRL sent out sample sets containing *P. larvae* (N = 10 to 12, including samples with positive and negative honey or sugar sirup). Qualitative and quantitative results as well as methodological details were reported to the NRL. Unfortunately, three out of five labs in 2022 and four out of seven labs in 2023, partly failed to identify samples containing *P. larvae* spores. This was mostly due to improper ERIC II – genotype results. These laboratories were able to improve their results, especially by changing the culture media used to Columbia Sheep Blood Agar. As a result, none of the seven participating laboratories reported wrong negative results during CLT 2024. Quantitative results of this CLT still showed significant differences between the labs. Likely factors are the type of culture media, different manufacturers of the same culture media and the use of enriched CO_2 atmosphere. Thus, we conclude that the choice of culture media can greatly affect the analysis success. Moreover, it shows the importance for a harmonized methodology of labs in *P. larvae* analytics.

Keywords: ring test, American foulbrood, food stores samples, honey samples, diagnostics

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zurück zum Programm

P5.11 Erste Ergebnisse zur Charakterisierung eines Bakteriozins in *Paenibacillus larvae*

First results on the characterization of a spiteful bacteriocin in *Paenibacillus larvae*

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American Foulbrood (AFB) is one of the most destructive brood diseases affecting honey bees and is caused by *Paenibacillus larvae*, a gram-positive, spore-forming bacterium. The two epidemiologically relevant genotypes ERIC I and ERIC II are widespread and equally prevalent. Despite their similar distribution, infections in honey bee colonies are typically caused by either ERIC I or ERIC II, but rarely by both. We therefore hypothesize that ERIC I and ERIC II compete within the infected host, acting as rivals rather than coexisting. Such targeted elimination within the host of closely related competitor strains while sparing clonemates is known as spiteful behaviour and driven by bacteriocins which belong to a group of microbially active peptides called lantibiotics. Lantibiotics are ribosomally synthesized, post-translationally modified peptides containing unusual amino acids like lanthionine. *In silico* analysis of the genome sequences of *P. larvae* ERIC I and ERIC II revealed gene clusters for the production of bacteriocin in both genotypes. The putative *P. larvae* bacteriocin precursor peptide and the core peptide were shown to be identical for both genotypes. To investigate whether *P. larvae* ERIC I and ERIC II exhibit spiteful behaviour and whether *P. larvae* bacteriocins play a role in this, gene inactivation mutants for these bacteriocins were generated in both genotypes. We will show the first results regarding the characterization of *P. larvae* bacteriocins.

Keywords: *Paenibacillus larvae*, American Foulbrood, bacteriocin, spiteful behavior

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zurück zum Programm

P5.12 Mikrobielle Kollagenase A (*Pl*ColA) - eine sezernierte Metalloprotease und ein Virulenzfaktor von *Paenibacillus larvae*

Microbial Collagenase A (*Pl*ColA) - a secreted metalloprotease and virulence factor of *Paenibacillus larvae*

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American Foulbrood is a fatal disease of the honey bee (*Apis mellifera*) brood caused by *Paenibacillus larvae*, a gram-positive, spore-forming bacterium. An infection begins when *P. larvae* spores, ingested by a first instar honey bee larva, germinate in the midgut. After massive proliferation of the vegetative bacteria in the midgut lumen, they breach the midgut epithelium and invade the larval hemocoel, a process that it causing the death of the larva. The bacteria totally degrade the larval carcass into a ropy mass and sporulate. The ropy mass finally dries out, thus forming the foulbrood scale, composed of billions of spores. For the development of effective treatment strategies a comprehensive understanding of the molecular mechanisms underlying this disease and the identification of bacterial virulence factors are essential.

Since secreted proteases were suspected to be relevant virulence factors of *P. larvae* from the very beginning, we analyzed the protease profiles of culture supernatants of *P. larvae* ERIC I and ERIC II. We identified a protease which showed gelatinolytic activity in degradation assays and zymograms. *In silico* analysis revealed that the genomes of *P. larvae* ERIC I and ERIC II each contain a gene for a microbial collagenase (PlColA), which belongs to the family of M9 metalloproteases and contains the domains required for collagenolytic activity. We confirmed, that the products of the identified *Pl*ColA genes are responsible for the observed gelatinolytic activities, by demonstrating that gene inactivation mutants for *col*A in *P. larvae* ERIC I and ERIC II no longer exhibited gelatinolytic activity in zymograms. Exposure bioassays performed with honey bee larvae and the *Pl*ColA-deficient *P. larvae* ERIC II mutant or the corresponding *P. larvae* ERIC II wildtype revealed that *Pl*ColA as a virulence factor of *P. larvae* ERIC II.

Keywords: *Paenibacillus larvae*, American Foulbrood, virulence factors, proteolytic enzymes

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zurück zum Programm

P5.13 Entwicklung eines Lateral-Flow-Assays zum Nachweis von *Paenibacillus larvae* Sporen

Development of a lateral-flow-assay for the detection of *Paenibacillus larvae* spores

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American Foulbrood (AFB) is an intestinal infection of honey bee larvae (*Apis mellifera*) caused by the gram-positive bacterium *Paenibacillus larvae*. The endospores of *P. larvae* are the only infectious form of the pathogen. These spores are ingested by the larvae through contaminated food, but only young larvae, up to approximately 36 hours after egg hatching, are susceptible to infection. The infection is fatal for the larvae, whose carcasses are totally degraded by *P. larvae* to a ropy mass, which then dries out to a so-called foulbrood scale containing millions of spores. AFB is highly contagious and can result in significant economic and agricultural losses, which is why it is considered a notifiable disease in Germany and many other countries. Diagnosing AFB in the laboratory typically takes up to two weeks, during which valuable time is lost before control and hygiene measures can be implemented to prevent further spread of the disease. In order to achieve a more rapid diagnosis of AFB, we are developing a lateral-flow-assay for the detection of *P. larvae* spores, which could be utilized directly on the colony. The identification of a *P. larvae* specific spore protein, which is part of the exposporium was followed by its cloning, recombinant expression and purification. This protein was then used as the antigen for the production of monoclonal antibodies. The resulting functional antibodies were tested in several assay formats, including western blot, dot blot, sandwich ELISA, and lateral-flow-assay. The results of these tests consistently showed the ability to detect the recombinantly expressed and purified antigen. Preliminary tests with field samples (foulbrood scales) from *P. larvae* infected bee colonies demonstrated successful detection in the lateral-flow-assay.

Keywords: *Paenibacillus larvae*, spore specific protein, lateral flow assay

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zurück zum Programm

P5.14 Nachweis des Infektionszyklus von *Bacillus thuringiensis* in Honigbienenlarven

Detection of the infection cycle of *Bacillus thuringiensis* in honey bee larvael

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Bacillus thuringiensis is used as biological pest control for treatment of Galleria *mellonella* infestation in bee colonies. *B. thuringiensis* kills larvae of *G. mellonella* with insecticidal crystal inclusions, which are produced while sporulation. Germinated vegetative *B. thuringiensis* degrade the cadaver after larval death. After consumption of all available nutrients the vegetative bacteria sporulate again and produce the insecticidal crystal inclusions (Cry toxins) for the next infection cycle. The lethal effect of Cry toxins is usually described as species specific, hence *B. thuringiensis* is often used as a biological insecticide for certain pests, without threatening other insects. *B. thuringiensis* was described as harmless for honey bee larvae in the past. But recent studies have indicated that treatment of bee colonies with *B. thuringiensis* is more noxious for honey bee larvae as expected so far. We confirm in this study that *B. thuringiensis* var. *kurstaki*, isolated out of the pest control product “B401”, is able to complete a whole infection cycle in honey bee larvae. We show that spores of *B. thuringiensis* in larval diet had a significant lethal effect on survival rate of honey bee larvae reared in the laboratory. This lethal effect however was dependent on the timepoint of infection that means younger larvae were more susceptible than older larvae. We demonstrate via fluorescence *in situ* hybridization that spores of *B. thuringiensis* germinated and vegetative bacteria proliferated in both living and dead honey bee larvae. Finally, we can confirm that spores of *B. thuringiensis* were re-isolated out of the cadaver after the degradation of the dead larvae. *B. thuringiensis* is therefore able to complete his infection cycle in honey bee larvae and the use as biological pest control in bee colonies should be reevaluated.

Keywords: *Bacillus thuringiensis,* fluorescence *in situ* hybridization, honey bee larvae, pathogen

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zurück zum Programm

P5.15 Das Deutsche Bienen Monitoring (DeBiMo): Ergebnisse aus der Bienensaison 2023/2024

German bee monitoring: Monitoring results from the bee season 2023/2024

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The German bee monitoring (DeBiMo) is a long-term collaborative monitoring project that has collected data on the health status of honey bee colonies in Germany since 2004. The project was initially launched to investigate factors that contributed to high winter colony losses that occurred in the winter of 2002/2003 throughout Germany. For over twenty years a variety of health indicators such as *Varroa destructor* infestation, pathogen prevalence, and pesticide exposure as well as management practices have been collected in cooperation with beekeepers at approximately 120 apiaries and 1200 colonies distributed throughout the country.

We present an overview of the latest overwintering results from the winter 2023/2024 and the ensuing beekeeping season 2024. *Varroa* mite infestation levels and the pathogen burden carried by colonies in autumn are important parameters that provide significant information on the likelihood of over-wintering success of honey bee colonies. Another aspect is the surveillance of novel invasive threats like *Tropilaelaps*-mites or the small hive beetle (*Aethina tumida).* The non-native Asian hornet (*Vespa velutina nigrithorax*) has caused quite a stir in southern Germany in 2024 due to its rapid geographic spread from southern Europe.

Taken together, these data inform about the current state of honey bee colony health and the challenges and threats beekeepers face in Germany. The constancy of data over 20 years also allows significant insight into the long-term drivers of colony decline, helping to inform policy makers on the risk factors negatively influencing colony health.

Keywords: colony health, monitoring, parasite prevalence

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P5.16 SEA-BEE: Ein Abenteuer von Wissenschaft in die Industrie

SEA-BEE: A voyage from research to industry

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American (AFB) and European foulbrood (EFB) are devastating honey bee brood diseases that can cause high numbers of colony losses when colonies are infected. AFB is caused by the bacterial agent *Paenibacillus larvae* that can be categorized to different ERIC-genotypes. These ERIC-genotypes can differ in virulence. EFB is caused by *Melissococcus plutonius* and is considered less harmful compared to AFB. For the prevention of disease spread and the conduction of suitable and fast control measures, a rapid, reliable and affordable diagnostic tool is needed. Therefore, we developed an antibody based lateral flow assay (LFA) for *M. plutonius* and *P. larvae* detection and also for the distinction of the two main occurring *P. larvae* ERIC-genotypes (ERIC I & ERIC II). For the establishment of the LFA specific monoclonal antibodies (mAbs) detecting *M. plutonius*, *P. larvae* or ERIC II have been generated and characterized for specificity towards field isolates and other bee associated bacteria. The suitability of the generated mAbs was tested in a sandwich ELISA before mAbs were transferred to LFAs. The produced LFAs were tested in the laboratory using bacterial extracts and field samples consisting of larvae from hives that were associated with AFB infections. During that process certain optimizations of the LFA, such as lysis buffer, membrane blocking and antibody stabilization took place.

After the optimizations of the LFA and the sample preparation procedure, a high-quality and sustainable antibody production is needed for maintaining the reproducibility of the assay. Therefore, the SEA-BEE project is dealing with the transfer of the antibody production from hybridoma cells to diatoms and the comparison between the reactivity of the differently produced antibodies. The protein production with diatoms is a sustainable, cheap and reliable tool that will make the LFA affordable for different stakeholders, increase its scope and help to prevent disease spread.

Keywords: Amerikanische Faulbrut, Europäische Faulbrut, Schnelltest, Diagnostik, ERIC-Genotypisierung

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