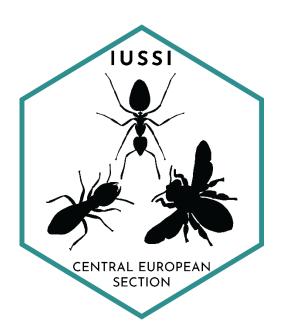
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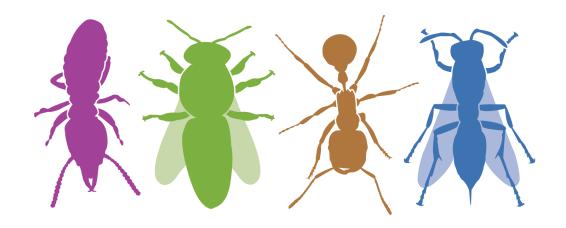


# 7th Central European Section Meeting of the International Union for the Study of Social Insects

17-20.09.2023 Cluj-Napoca



Programme and book of abstracts



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#### **Programme**

#### SUNDAY, 17<sup>TH</sup> OF SEPTEMBER – Emanuel de Martonne st. 1, Casa Universitarilor

17:00 – Arrival and registration of participants

19:00 - Welcome cocktail

**MONDAY, 18<sup>TH</sup> OF SEPTEMBER** – Zoology Building of the Faculty of Biology and Geology, Clinicilor st. 5-7

8:00–8:45 Registration of participants

8:45-9:00 Conference opening

#### Plenary talk

9:00 From the past to the present: 100,000,000 years of ant history.

#### Alexander Radchenko

#### Section chair: Simon Tragust

10:00 Social insect aging from a Cardiocondyla-biased perspective.

Luisa María Jaimes Niño

10:15 Effects of colony fusions on evolutionary optimal colony size in a termite species. *Stephan Lohmar* 

10:30 Convergent evolution of helping behaviour in a neotropical Army ant.

Juan José Lagos-Oviedo

#### Coffee Break

#### Section chair: Tomer J Czaczkes

- 11:15 Ecological drivers of sociality in *Xyleborinus saxesenii*, a widely distributed ambrosia beetle. *Antoine Melet*
- 11:30 Wood-pastures promote environmental and ecological heterogeneity on a small spatial scale: the "ecosystem complex" approach. *Adam Lőrincz*
- 11:45 Environmentally driven behavioural trait variation may promote species coexistence in ants. *Florian Menzel*
- 12:00 Effect of different wood-pasture habitat types on ant functional traits.

István Elek Maák

#### Lunch

#### Plenary talk

14:00 Ants under parasitic siege: from specialist to generalist fungi. *Enikő Csata* 

#### Section chair: Heike Feldhaar

15:00 Social immunity behaviors in ant-nematode interactions. *Florian Strahodinsky* 

15:15 A new puppet-master and its puppet? Behavioural manipulation in the host-parasite system between the fly *Strongygaster globula* and its ant host. *Simon Tragust* 

15:30 Host-parasite interaction cause morphological changes in a solitary bee and its endoparasite. *Silvio Erler* 

#### **Coffee Break**

#### Section chair: Ioan Tăușan

- 16:15 Combined stressors in the agriculture Investigating more effects than honey bee colony development. *Karoline Wüppenhorst*
- 16:30 Development of ant colonies in an urban-rural gradient. Gema Trigos Peral
- 16:45 Communal nest-founding in Euglossa cybelia. Jonas Henske
- 17:00 Genetic divergence and aggressiveness within a supercolony of the invasive ant *Linepithema humile. Iago Sanmartín-Villar*

#### 17:30-19:00 - Poster session with drinks

19:00 - Dinner

**TUESDAY, 19<sup>TH</sup> OF SEPTEMBER** – Zoology Building of the Faculty of Biology and Geology, Clinicilor st. 5-7

#### Plenary talk

9:00 Why can social insect queens live so long? Insights from termites. *Judith Korb* 

#### Section chair: Gema Trigos Peral

- 10:00 What makes a termite queen? Recurring transcriptomic signatures in termites with totipotent workers. *Silu Lin*
- 10:15 Effect of unbalanced diets and individual amino acids on the longevity/fecundity trade-off in a clonal ant. *Lina Pedraza*

#### **Coffee Break**

#### Section chair: István Maák Elek

- 11:00 Measuring insect flow with AnimalTA's new feature. Violette Chiara
- 11:15 Can ants learn to be better at recognition? *Melanie Bey*
- 11:30 Ants combine object affordance with latent learning to make efficient foraging decisions. *Laure-Anne Poissonnier*
- 11:45 Spatial fidelity and uniform exploration in the foraging behavior of a giant predatory ant. *Priscila Elena Hanisch*
- 12:00 Comparative choice assays allow simple and high-sensitivity quantification of ant feeding preference. *Tomer J Czaczkes*

#### Lunch

#### Section chair: Michaela Hönigsberger

- 15:00 Problem-solving through individual cognition in invasive social insects. *Srikrishna Narasimhan*
- 15:15 Histone deacetylase 3 silencing activates transposable elements activity in the dry wood termite *Cryptotermes secundus* queens. *Louis Allan Okwaro*

15:30 The expression of elongases and desaturases shed light on the CHC plasticity of honey bees (*Apis mellifera*). *Daniel Sebastian Rodríguez León* 

#### Coffee break

16:15–18:00 – General Assembly of the CE section of IUSSI

18:30 – Farewell dinner

#### POSTER SESSION

#### Section chair: Violette Chiara

- 1. Investigating bumblebee foraging decisions with robotic flowers. Mélissa Armand
- 2. Evolutionary drivers of rescue and wound care behaviour in the termite hunting ant species *Megaponera analis*. *Vibhuti Bhat*
- 3. Genetic structure and mating system of the ant *Temnothorax makora*.

#### Emil Eichelbrönner

4. The diseased honey bee colony – distribution of a brood pathogen.

#### Sandra Ehrenberg

5. Sanitary grooming in ants is induced by microbe-specific cues.

#### Michaela Hönigsberger

- 6. Biophysical and functional consequences of cuticular hydrocarbon variation in ants. *Selina Huthmacher*
- 7. Real-time visualization of nest disinfection & its effect on pathogen control in ants. *Lukas Lindorfer*
- 8. Effects of sinkhole habitats on the functional and behavioural traits of *Myrmica ruginodis*. *Bonita Ratkai*
- 9. Information provision and use in ant pheromone trails.

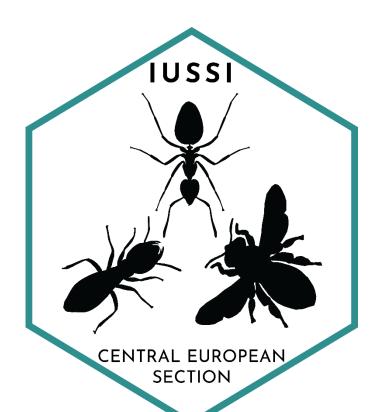
#### Federico Javier Olivera Rodriguez

- 10. Does the macronutrient composition affect the survival of the *Myrmica scabrinodis*, Nylander 1846 (Hymenoptera: Formicidae) ants infected with the ectoparasitic fungus *Rickia wasmannii?* **Ágota Szabó**
- 11. Revealing a taxonomic mystery: *Reticulitermes* sp (Isoptera: Rhinotermitidae) in Romania. *Ioan Tăușan*

#### WEDNESDAY, 20<sup>TH</sup> OF SEPTEMBER

Postworkshop trip to Rimetea (please bring hiking boots and clothes)

# Abstracts of Plenary Talks



### From the past to the present: 100,000,000 years of ant history

#### **Alexander Radchenko**

Schmalhausen Institute of Zoology of National Academy of Sciences of Ukraine, Ukraine



Ants belong to the family Formicidae of the superfamily Formicoidea (Hymenoptera: Aculeata). The oldest fossil ants are known from the Lower Cretaceous, ca. 115 Ma. 77 ant species from 41 extinct genera have been described from Cretaceous deposits. Most of them belong to 5 extinct subfamilies – stem groups. However, the oldest known genus belonging to the crown ants (modern subfamily Formicinae) was described most recently from Burmese amber, ca. 100 Ma (Wu, Radchenko, Engel et al., in press.). Starting from the Cenozoic only crown subfamilies existed, and modern genera appeared in the Early Eocene (ca. 55 Ma). In total, 851 species from 171 extinct and 85 extant genera from about 100 deposits have been described till now.

There are two kinds of fossils – imprints in sediments and inclusions in amber. Imprints are much less informative than amber inclusions. There are 26 major amber ant deposits in the World; they dated from ca. 100 (Cenomanian) to 15 Ma (Miocene). The richest and best studied are Late Eocene European ambers (37.8-33.9 Ma) – Baltic, Danish, Bitterfeld and Rovno: 184 species from 68 genera of 12 subfamilies are recorded in them.

### Ants under parasitic siege: from specialist to generalist fungi

#### Enikő Csata

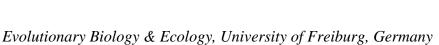
Museum and Institute of Zoology, Polish Academy of Sciences, Poland



Pathogens, parasites and their hosts provide excellent models for studies on coevolution, they are intrinsically linked but their evolutionary interests are different. Not just unitary organisms can have parasites and thus exhibit behavioral modifications due to parasitic effects, but social organisms such as ants can also be targeted. Ants are frequently exploited by parasites, which, in some instances, trigger alterations in their life history, physiology and behavior. The social life of ants implies a high frequency of contact among nestmates that increases the risk of spreading any pathogens among colony members. Some ant parasites became iconic species due to their smartness in manipulating host behavior or because of their aesthetic beauty but the ecology of less charismatic ant pathogens and parasites, unfortunately, such as microscopic fungi remains insufficiently explored, leading to a limited understanding. In this talk, I delve into studies examining ants and their interactions with parasitic fungi across Europe, focusing on different types of relationships involving three myrmecopathogenic fungi and their respective ant hosts.

### Why can social insect queens live so long? Insights from termites

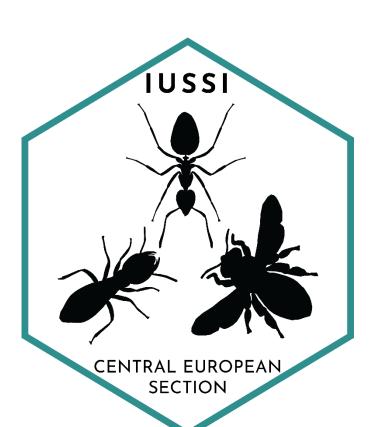
#### **Judith Korb**





Social insects are emerging model organisms to study the evolution of ageing. They are especially suited for this purpose because (i) queens reach extraordinary lifespans for an insect, (ii) these long-lived queens co-occur with short-lived worker nestmates, (iii) queens have apparently overcome the common trade-off between longevity and fecundity, (iv) queens' lifespan seem to change with the degree of sociality, and (v) social insects are amenable to experimental manipulations. I will present results of long-term research in which we aimed to identify ultimate causes and proximate mechanisms underlying the long lifespan of termite queens. Starting with long-term monitoring data, we determined survival patterns and quantified longevity of queens from two species at different ends of the termites' sociality spectrum. We showed how workers mitigate fecundity costs of queens, thus contributing to explain why queens can be longlived and highly fecund. By manipulating food quantity and quality as well as environmental stress, our research points to causes and interactions underlying caste-specific ageing. Complementing these studies with transcriptome analyses, further enabled us to identify associated molecular life history pathways. Finally, our molecular experiments pinpoint to specific alterations in their molecular networks that can contribute to explain why termite queens live so long.

## Abstracts



#### Investigating bumblebee foraging decisions with robotic flowers

Mélissa Armand<sup>1</sup>, Tomer J. Czaczkes<sup>1</sup>, Massimo De Agrò<sup>1</sup>

<sup>1</sup> Animal Comparative Economics Laboratory, Department of Zoology and Evolutionary Biology, University of Regensburg, Germany

When faced with limited resources, such as time or information, both humans and non-human animals often rely on heuristics or "rules of thumb" to make decisions. These cognitive "shortcuts" often lead to satisfactory decisions, but they can also result in cognitive errors and apparent irrational choices. In the context of mutualistic plant-pollinator relationship, we propose that plants may exploit pollinator preferences by leveraging their reliance on heuristics. Our research aims to investigate how bumblebee (*Bombus terrestris*) decision-making is influenced by their use of heuristics when foraging. To investigate this hypothesis, we have developed robotic flowers that allow precise control of color, scent, and nectar rewards. We aim to use these robotic flowers to assess whether bumblebees foraging decisions are influenced by their own cognitive biases. The development of such robotic, fully controllable flowers opens up exciting opportunities to automate complex, long-term behavioral experiments of free-flying insects.

#### Can ants learn to be better at recognition?

#### Melanie Bey<sup>1</sup>, Naomi Alex<sup>1</sup>, Lisa Maczkowicz<sup>1</sup>, Volker Nehring<sup>1</sup>

<sup>1</sup> Department of Evolutionary Biology and Ecology, Institute of Biology I, University of Freiburg, Germany

Eusociality is an evolutionary stable strategy that has allowed many species to prosper over 100 million years. For eusocieties to last, individuals must cooperate with relatives. Nestmate recognition is the sine qua non condition of kin selection that hold members of the society together. In ants, recognition is based on the identification of colony-specific odours (label), which workers compare to a neural template learnt as their olfactory identity. Multiple studies have shown tremendous individual variability in recognition abilities. This variability is partly explained by literature to be programmed temporal polyethism or habituation product. What cannot be explained is often dismissed as experimental noise attributed to colony differences or even idiosyncratic variation. Our research seeks to investigate the role of associative learning in construction of template and how it contributes to the variability in recognition abilities. We used behavioural experiments to understand how learning modulate the nestmate template. We further analysed brain and antennae transcriptomes of ants subjected to different experiences to investigate the proximal mechanism involve in learning. We seek to provide empirical evidence in support of the distributed nestmate recognition model that argue in favour of the associative learning playing a role in recognition.

### Evolutionary drivers of rescue and wound care behaviour in the termite hunting ant species *Megaponera analis*

Vibhuti Bhat<sup>1</sup>, Erik T. Frank<sup>2</sup>

<sup>1</sup> Univeristy of Würzburg, Würzburg, Germany

<sup>2</sup> Department of Animal Ecology and Tropical Biology, University of Würzburg, Würzburg, Germany

Megaponera analis is an ant species specialized on hunting termites of the subfamily Macro-termitinae. This prey- predator combat has led to the evolution of adaptive behaviours in both. Rescuing the injured ants from the foraging site back to their nest and if needed, treating the infected wounds with antimicrobial compounds are two unique behaviours present in M. analis to minimise combat mortality. This specialised hunter is present across sub-Saharan Africa. Rescue and wound care behaviours have so far only been studied in a West African Population (Ivory Coast) of M. analis. Thus, for a comparative study, we sampled M. analis populations in South Africa and Mozambique. Interestingly, M. analis in South Africa faces a more pugnacious prey compared to Mozambique, where the termites are much smaller and less injurious. This interesting difference in prey size further encouraged us to investigate the occurrence of two adaptive behaviours in these closely present populations of M. analis. Overall, this study allows us to better understand the evolutionary drivers responsible for rescue and wound care behaviours to arise in social insects and why it has evolved so rarely.

#### Measuring insect flow with AnimalTA's new feature

#### Violette Chiara<sup>1</sup>

<sup>1</sup> Museum and Institute of Zoology, Polish Academy of Sciences, Warsaw, Poland

AnimalTA is a free, open-source, and user-friendly video tracking program. This program is especially suitable for invertebrate tracking as it can track small targets, even on heterogeneous backgrounds. Moreover, AnimalTA specializes in high sample sizes and proposes tools to treat rapidly numerous videos. The program is tailored to handle high sample sizes and efficiently process numerous videos, making it ideal for researchers working with large datasets.

In this presentation, we aim to introduce a new functionality of the program, which is the possibility of tracking individuals entering or leaving the video. This novel feature opens up new possibilities for studying the movements of social insects in various situations such as bridge crossing, climbing a tree, or leaving and entering their nest. We believe such functionality, combined with the versatility of the original program, might allow us to measure the flow of social insect movements even in a natural context.

The user-friendly interface ensures that researchers with varying levels of technical expertise can easily leverage its power for their studies, establishing AnimalTA as a useful tool in behavioral ecology, ethology, and ecology research.

#### The diseased honey bee colony - distribution of a brood pathogen

#### Sandra Ehrenberg<sup>1</sup>, Lukas Rüttinger<sup>1</sup>, Robert Kammerer<sup>1</sup>, Marc Schäfer<sup>1</sup>

<sup>1</sup> Friedrich-Loeffler Institute, Greifswald, Germany

Honey bees are considered superorganisms due to their eusocial behaviour. They live highly condensed in hives under homeostatic conditions, what makes it easy for pathogens to survive and spread through the colony.

The brood disease American foulbrood (AFB) is caused by the bacterium *Paenibacillus* larvae that is ingested by larvae in its spore form together with the larval food. AFB only causes symptoms in larvae and is spread by adult bees via food. To get a better impression of the pathogen distribution in the hive various hive samples (larvae, adult bees, honey supper and swabs from combs and colony entry) using different diagnostic methods (bacteria cultivation, PCR and lateral flow device=LFD) were investigated for their pathogen load.

In 2022, 20 colonies from AFB-suspicious apiaries were sampled for different hive materials and tested for the presence of *P*. larvae. The AFB causing agent was present in 69 % of the tested colonies.

Generally, *P*. larvae can be detected in all of the tested hive materials with differences in occurrence according to pathogen load. These results give useful information about the distribution of the pathogen in the colony and should be used for further transmission and diagnostic studies in honey bee colonies.

#### Genetic structure and mating system of the ant Temnothorax makora

Emil Eichelbrönner<sup>1</sup>, Laura Neubauer<sup>1</sup>, Keiko Hamaguchi<sup>2</sup>, Kyoichi Kinomura<sup>3</sup>, Marion Cordonnier<sup>1</sup>, Jürgen Heinze<sup>1</sup>

<sup>1</sup> Zoology / Evolutionary Biology, Universität Regensburg, Regensburg, Germany

<sup>2</sup> Forestry and Forest Products Research Institute, Kansai Branch, Fushimi-ku, Kyoto, Japan

<sup>3</sup> Gifu-Aikawa High School, Gifu, Japan

What are the genetic structure and mating system of *Temnothorax makora*? Temnothorax is a large ant genus inhabiting substantial parts of the world. While the biology of North American and European species has intensively been studied, less is known about the 16 Japanese representatives. Given the variability of social structures and life histories in this genus, investigating the reproductive tactics in colonies with multiple queens is of particular interest. We investigated the genetic structure of 15 colonies of facultatively polygynous T. makora from Gifu, Japan, a species in which sibmating has previously been observed in lab nests (Murase et al. 2004, Proc. Arthropod. Embryol. Soc. Jpn. 39, 47-49). The numbers of queens and workers were recorded in all colonies. In behavioural experiments, we investigated if sexuals mate inside their natal nests and if gynes return after mating with alien males. Based on the seven microsatellite markers with the highest variability (among 42 candidates), the genotypes of 12 workers per colony allowed reconstructing the genetic colony structure. This supported the contribution of multiple queens to the brood and indicated an excess of homozygous genotypes. Together with the behavioural observations this corroborates the occurrence of intranidal mating, which is quite unique in this genus.

### Spatial fidelity and uniform exploration in the foraging behavior of a giant predatory ant

Priscila Elena Hanisch<sup>1</sup>, E. R. Hanisch<sup>1</sup>, V. Blanco<sup>1</sup>, P. L. Tubaro<sup>1</sup>, A. V. Suarez<sup>1</sup>

<sup>1</sup> Biocenter – Zoology III, University of Würzburg, Würzburg, Germany

Social organisms benefit from division of labour and collective behaviours. However, if individuals overlap widely in their efforts, these benefits may not be proportional to the number of individuals that take part in an activity. We examined foraging behaviour and route fidelity in colonies of the ant *Dinoponera* grandis (formerly Dinoponera australis), a large species with relatively few active foragers that lack nestmate recruitment and chemical trailing behaviour. For 12 colonies, we marked individual foragers and mapped their foraging routes to test the hypothesis that each ant specializes in a particular area around the nest and that this route fidelity increases the overall area covered by the colony. For each individual, we recorded the mean direction and duration of each foraging trip, foraging success and maximal distance from the nest. For each colony, we measured the number of workers and the total foraging area. Additionally, we measured Shannon's entropy to describe foraging behaviour structure of the colonies. Overall, we mapped 272 foraging routes from 95 different foragers. The total area used by each colony averaged 66.2 m2. Within colonies, over 68% of foragers exhibited a high degree of route fidelity, with most foragers following different foraging directions. Most colonies had a high Shannon's entropy, suggesting an even exploration of the foraging territory. Our results suggest that D. grandis exhibit route fidelity and high entropy. This strategy likely increases foraging efficiency and search area and may be particularly important for species with relatively few foragers.

#### Communal nest-founding in Euglossa cybelia

#### Jonas Henske<sup>1</sup>, Mauricio Fernandez Otarola<sup>2</sup>, Thomas Eltz<sup>1</sup>

<sup>1</sup> Department of Animal Ecology, Evolution & Biodiversity, Ruhr-Universität Bochum, Bochum, Germany

The neotropical orchid bees are best known for the pollination services they provide to numerous tropical plants, including hundreds of orchid species, thanks to the peculiar behavior in which male bees collect exogenous volatiles. However, our understanding of female behavior is limited. Finding orchid bee nests is challenging, and nest architecture has been described only for a small fraction of the known species. Nesting and social behavior have been studied in even fewer species. Orchid bees are corbiculate bees but differ from their closest relatives, including honeybees, bumblebees, and stingless bees as they do not exhibit obligate eusociality. Although orchid bees have long been considered solitary, recent studies indicate a broad range of social behaviors. Here, we present observations and kinship analyses of the aerial group nesting species Euglossa cybelia revealing communal nest founding behavior of unrelated individuals with no social hierarchies. All females were mated and capable of reproduction. Notably, recently constructed nests only produced male offspring. Our findings enhance the understanding of the nesting biology of Eg. cybelia providing for the first time kinship analyses of a communal euglossine bee species.

<sup>&</sup>lt;sup>2</sup> Escuela de Biología, Universidad de Costa Rica, San Pedro de Montes de Oca, San José, Costa Rica

#### Sanitary grooming in ants is induced by microbe-specific cues

## Michaela Hönigsberger<sup>1</sup>, Barbara Milutinović<sup>1,2</sup>, Thomas Schmitt<sup>3</sup>, Sylvia Cremer<sup>1</sup>

<sup>1</sup> Institute of Science and Technology, Klosterneuburg, Austria

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Ants provide highly efficient sanitary care by grooming off fungal pathogens from the body surface of exposed colony members, even before an infection has established. Ants therefore detect and react to the mere presence of pathogenic spores on otherwise still healthy colony members, likely by a chemical compound revealing either the pathogenic nature of the spores or only its microbial nature, which would equally occur on pathogenic and non-pathogenic fungi. We characterized the chemical profile of spores of the entomopathogenic fungus Metarhizium and identified the fungal membrane compound ergosterol as a prominent compound. We found that application of ergosterol in its pure form, i.e. in the absence of any tactile cues from the spores and without any integration into the full spore chemical bouquet, elicited sanitary caregiving in nestmate workers. Interestingly, only the fungus-derived ergosterol, but not the structurally similar yet not fungus-derived cholesterol induced this response by colony members. Therefore, sanitary grooming by Argentine ants is triggered by a general microbe-specific, but not a pathogen-specific, compound, allowing us to identify ergosterol as a MAMP (microbe-associated molecular pattern) for social immunity.

Stock M, Milutinović B, Hoenigsberger M, Grasse AV, Wiesenhofer F, Kampleitner N, Narasimhan M, Schmitt T & Cremer S (2023) Nat Ecol & Evol 7, 450–460

### Host-parasite interaction cause morphological changes in a solitary bee and its endoparasite

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The grey-backed mining bee Andrena vaga is the host for the Strepsipteran endoparasite Stylops ater, which is known for extreme sexual dimorphism and hypermetamorphosis. For most regions in Europe, were both are occurring; the population structure, genetic diversity and morphological adaptations are unknown. Here, we sampled hosts and parasites of both sexes at several aggregation areas in and around Braunschweig (Germany) to study their biology and distribution. Genetic analysis revealed the absence of local variation within Stylops. Host bees emerged earlier than non-parasitized bees. Individual male and female bees hosted up to four parasites in their bodies. A trend was detected in the Stylops' preference for hosts of their own sex and the position of extrusion from the host abdomen. Morphological adaptations upon parasitation include: ovary reduction and reduced head width for bees infested by male Stylops, host masculinization (indicated in the shape of the metabasitarsus), and intensified tergal hairiness (most strongly near the point of parasite extrusion). Parasites adapted to the hosts' sex and multiple infestation with reduced cephalothoraxes. Future studies are needed to understand the mechanisms behind parasite-induced host manipulation.

### Biophysical and functional consequences of cuticular hydrocarbon variation in ants

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Myrmica ruginodis. These sister species differ significantly in their cuticular hydrocarbon (CHC)-profiles, however both species occur under the same climate regimes and often coexist next to each other. While the CHC-profile of *M. rubra* mostly consist of methyl branched alkanes, the CHCs of *M. ruginodis* are for the most part unsaturated compounds. I will collect these two species from a maritime to a continental climate regime across Europe, to examine if the local climate has an impact on the CHC profile. Further, I will analyze if the ants from different climatic habitats vary in their ability to acclimate to different constant and fluctuating temperatures. For that, I will investigate the chemical composition as well as the physical properties like viscosities and melting ranges of CHC layers of differently acclimated ants. I will also study how acclimation affects the survival of ants under dry conditions and how much water is lost through the CHC layer.

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#### Social insect aging from a Cardiocondyla-biased perspective

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While there is a significant interest in understanding why and how reproductives of ants live so long, a lack of demographic data hinders accurately estimating the shape and pace of aging of ant species. Reports of queen lifespan generally rely on data from a single or a few individuals. We revised data from extensively studied field colonies of two species of *Pogonomyrmex* and compared their life trajectories to our model organism *Cardiocondyla obscurior*. The three species exhibit actuarial senescence (an increase in mortality with age), but only very late in life, indicating a non-gradual aging pattern as previously observed in the termite *Cryptotermes secundus*. We propose that this pattern is attributed to a late-life peak of sexual production, as seen in *C. obscurior*. Interestingly, the delay of actuarial senescence, which we termed "continuusparity" as a distinction from iteroparity and semelparity, is also observed in workers of this species. The selection against senescence until late in life is selected in queens, and we suggest that it is relaxed in workers due to their sterility and lack of direct fitness gains, leading to a shape of aging mirroring the one of the queens.

#### Convergent evolution of helping behaviour in a neotropical Army ant

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Group hunting species often evolve strategies to cope with the costs of hunting dangerous prey. Neotropical army ants exemplify collective hunters specialising in arthropods and social insects, but their raiding workers frequently suffer injuries and extremity loss, potentially impacting worker survival. Our study of Eciton burchellii revealed that approximately 7% of workers displayed injuries after a raid, totalling around 140,000 injured workers per colony. Freshly injured workers stand still close to the injury site, bend over their abdomen and open their mandibles. Afterwards, nestmates groom the injured worker at the injury site. Injured workers had reduced movement speed, and nestmates frequently carried them back. Additionally, injured workers whose wound was artificially infected were more likely to die if they did not receive wound care from their nestmates. Therefore, worker wound care was effective in increasing injured workers' survival. This study represents only the second in-depth examination of wound care and helping behaviour in injured ants. Since this behaviour independently evolved in two phylogenetic distant species; Megaponera analis (Ponerinae), and Eciton burchellii (Dorylinae), we suggest that helping behaviour is essential to deal with the costs of hunting pugnacious prey in groups.

### What makes a termite queen? Recurring transcriptomic signatures in termites with totipotent workers

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Social insect castes are prime examples of phenotypic plasticity (i.e., different phenotypes arising from the same genotype). Recently, intensive studies in social Hymenoptera (ants, bees, and wasps) have identified central genes and molecular pathways associated with caste development and caste differences. Some of them have apparently been co-opted from pathways underlying development in solitary insects. Much less is known for termites, which evolved caste phenotypes independently from social Hymenoptera. A recent study identified a module of co-expressed genes in head-prothorax tissue, the Queen Central Module (QCM), that characterizes the queens compared to workers of the termite Cryptotermes secundus (Kalotermitidae). The QCM comprises genes from central pathways underlying development in solitary insects and reflects major queen traits such as reproduction and chemical signaling. We tested whether the QCM is a shared toolkit that gives rise to the queen phenotype in termites more broadly. To this end, we investigated gene expression profiles during development in Zootermopsis angusticollis, a species from another termite Archotermopsidae. We compared transcriptome profiles of early larval instars, functional workers (which correspond to late larval instars) and queens (which develop from workers). We found that QCM genes were significantly enriched among the genes overexpressed in head-prothorax tissue of Z. angusticollis queens but not in abdomens. Furthermore, worker instars become enriched with QCM genes during the development from early larval instars. Our results support the hypothesis of a conserved genetic toolkit that characterizes termite queens and their development. Furthermore, our data imply that tissue-specific expression of key aging-related pathways might have facilitated the evolution of a long lifespan and high fecundity in termite queens.

### Real-time visualization of nest disinfection & its effect on pathogen control in ants

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Ant colonies have evolved rigorous strategies to counteract the risk of epidemics in their densely-populated nests that they often build in pathogen-rich soil. Pathogen exposure of colony members triggers a large repertoire of social behaviors that reduce both the risk of infection for exposed individuals and disease transmission through the colony. Moreover, invasive garden ants also prophylactically disinfect their nest with self-produced antimicrobials such as their formic acid-rich poison. It is not clear, however, whether the ants intensify their efforts under pathogen exposure and how targeted they apply this disinfection measure to contaminants or to their highly disease-susceptible brood. Here we introduce a real-time imaging system to disentangle the spatiotemporal dynamics of poison spraying and its effects on the viability of fungal spores on the nest floor, which consists of a nutrient-rich substrate that permits the growth of fungal spores contaminating the nest. A pH-sensitive dye allows us to pinpoint the location of applied ant poison. Combining high-resolution imaging and advanced analysis software, we obtain spatiotemporally resolved data on poison application and fungal growth patterns. This allows real-time visualization and quantification of the effectiveness of collective nest disinfection in ants.

### Effects of colony fusions on evolutionary optimal colony size in a termite species

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The major evolutionary transition from a group of single organisms to a closely cooperating 'superorganism' involves a gradually increasing linkage of the reproductive success of the individual to that of the colony as a whole, and hence a reduction in conflict within colony individuals. We hypothesize that this reduction in conflict will allow species that are more superorganism-like to form bigger colonies. To test the logic of this hypothesis, we have developed an agent-based simulation corresponding to the life-cycle of a termite species with totipotent workers (considered to have low superorganismality). We simulated the evolution of a reaction-norm trait that determines when workers leave their birth colony as a function of colony size. We considered two distinct scenarios, in which there is either (1) always a single pair of royals in each colony, or (2) the possibility of two colonies fusing. The latter scenario leads to reduced relatedness between workers in a colony and thus to smaller indirect fitness benefits. We will present our modelled predictions, which will later be tested against empirical data from *Cryptotermes secundus* colonies collected in Australia.

### Wood-pastures promote environmental and ecological heterogeneity on a small spatial scale: the "ecosystem complex" approach

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Wood-pastures harbor critical natural and social values and are among the most ancient land use forms of Europe. The crucial conservation value of these silvopastoral systems is generally contributed to their characteristic landscape elements, the solitary trees, which provide microhabitats for a variety of organisms. However, by accommodating four habitat types (grasslands, solitary trees, forests, and forest edges) on a relatively small spatial scale, wood-pastures might host functionally and compositionally distinct communities, thus enhancing the landscape-level biodiversity. To test this, in the presented study we assessed the fine-scale microclimatic conditions of the four different habitat types of three wood-pastures (Hungary and Romania) and investigated the effects of these conditions on ant activity and organization patterns. In addition, niche breadths (quantified by four-dimensional niche hypervolumes), niche overlaps, and interspecific interactions were also considered to assess the importance of interspecific competition in shaping the studied ant communities. The four habitat types showed notable differences in the measured microclimatic parameters, which resulted in different patterns of activity and organization. Our results unveil that by posing various microclimatic and structural conditions, these silvopastoral systems enable the coexistence of four ecologically distinct communities, which makes them ideal targets for biodiversity conservation.

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#### Effect of different wood-pasture habitat types on ant functional traits

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Wood-pastures accommodate different woody habitat types (solitary trees, forests, and their edges) in a relatively limited spatial scale that creates diverse microclimatic and abiotic conditions, thus enhancing the spatial environmental heterogeneity of the whole system. Moreover, they can provide a glimpse into the rules and underlying patterns of trait adaptation on small spatial scales. In our previous studies on wood pastures, we have found that the ant Temnothorax crassispinus is present in all the woody habitats, so we hypothesized that scattered trees, closed forests and forest edges should differentially affect the functional traits of this species. We collected 30 T. crassispinus nests from every woody habitat type and assessed their functional traits, such as colony and worker size, but also the number of sexuals and larvae. Our results showed that habitat type had differential effects on the functional traits but also on the nesting site preference of the studied colonies. For example, in solitary trees we found more virgin queens and queen pupae, whereas the worker pupae were more abundant on the forest edges. Considering the different nesting sites, the colonies were larger, and more virgin queens were present in the twig-nests, representing more of the forest habitats. However, more worker larvae were present in galls, a nesting site more abundant in solitary trees. Based on our results we can conclude, that the distribution of our functional traits becomes more or less even among the woody habitat types due to the complementary effects of different habitat characteristics, such as the differences in microclimate and the varying availability of nesting sites.

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### Ecological drivers of sociality in *Xyleborinus saxesenii*, a widely distributed ambrosia beetle

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Altruism evolves when indirect fitness benefits are high enough, but ecological factors are also important to explain its evolution. For ambrosia beetles, the nest is the most valuable resource, providing food, a stable environment and protection. Tunneling into wood may be the key innovation leading to the evolution of sociality in ambrosia beetles.

The cooperatively breeding ambrosia beetle *X. saxesenii* is a generalist, it colonizes several species of plants. While able to colonize a broad range of hosts, its performance across these hosts are unknown. In ambrosia beetles it is particularly interesting whether different hosts influence fitness through effects on the beetle's social farming behavior and productivity of their fungal symbionts.

We investigated how the host species impacts the behaviour, life-history and symbiont community of *X. saxesenii*. Our results show that *X. saxesenii* is a true generalist, performing equally in different hosts. We found no evidence for tradeoffs between different life-history traits, suggesting that it uses the same biological strategy regardless of the specific conditions encountered. We posit that such a generalist species has the potential for several speciation events, each one giving birth to a specialized species. Detailed effects on social behaviors and symbiont communities will be discussed too.

### Environmentally driven behavioural trait variation may promote species coexistence in ants

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Community ecology seeks to understand how species co-exist within the same habitat. Trait-based ecology assumes that trait differentiation among species may mediate niche differentiation, thus enabling coexistence. In this context, behaviour constitutes an often overlooked set of traits. However, behavioural traits such as aggression and exploration strongly drive intra- and interspecific competition, especially in ants, where co-existence is often shaped by aggressive encounters.

Here, we studied behavioural variation in three ant species (Formica rufibarbis, Lasius niger and Tetramorium caespitum), which often co-exist in close proximity. We analysed intra- and allospecific aggression, explorative behaviour, and foraging activity under field conditions. These traits were measured for 12 colonies per species, and four times each during several months. We asked how behaviour varies over time and in response to environmental variation. Temperature and the course of the season indeed affected all behavioural traits. However, these effects strongly differed between species; for example, temperature effects on behaviour were stronger in Formica, while seasonal effects were stronger in Lasius. Our results suggest that in our system, there is no stable dominance hierarchy, because aggression varies with temperature and season in a species-specific way. This dynamic response may be crucial to facilitate species co-existence.

#### Problem-solving through individual cognition in invasive social insects

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Biological invasions currently pose one of the major threats to ecology and economics, but they also offer valuable insights into evolutionary processes in a short time because species must adapt to solve new problems in the introduced environment. Problem-solving is usually addressed by behavioural plasticity, but it was proposed that in organisms such as ants, the costs of individual plasticity might outweigh its benefits. We analysed behavioural and neuroanatomical variability in one of the most widespread ant species, the Argentine ant (Linepithema humile), to unravel whether individual behavioural differences might be sufficient to reach colony-level solutions or exposure to problems is the factor shaping individual behaviour and brain. We exposed 173 age-controlled workers from five different colonies to sets of three behavioural tests conducted daily over five days. We tested ants' exploration, neophobia (encounter with an unfamiliar object) and maze-solving under laboratory conditions to analyse the mechanisms underlying problem-solving capability in a novel environment. Repeated behavioural testing allowed us to analyse personality, predictability, repeatability, memory and learning. We analysed the correlation among behavioural variability, cognitive development and micro-glomerular connections of mushroom bodies, the brain areas in charge of processing visual and mechanosensory information. The results we will present are a first step towards unravelling the basis of how introduced social insects cope with new environments, the key to understanding how they succeed to become invaders and how they will respond to the effects of climate change.

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### Histone deacetylase 3 silencing activates transposable elements activity in the dry wood termite *Cryptotermes secundus* queens

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Ageing refers to changes in physiological and molecular properties with age which enhance the likelihood of an organism to die. Transposable elements activity (TE-activity) seems to contribute to caste-specific ageing differences in termites. Long-lived reproductives seem to be better protected against TE-activity compared to short-lived non-reproducing workers. Yet the proximate mechanism that regulates TE-activity remains elusive. Here we used double-stranded small interference RNA to silence a histone deacetylase 3 gene (HDAC3) in young queens of the termite Cryptotermes secundus and analyzed their gene expression profile using fat body tissues transcriptome. Our results revealed 867 differentially expressed genes (DEGs) upregulated in the target and 582 DEGs upregulated in sham control samples. Many upregulated genes were related to TEs. Also, genes involved in cellular defense against TEs, such as Argonaute 2, were upregulated in the target group. Furthermore, 119 genes typically expressed in old queens were among the target upregulated genes. Gene ontology analysis showed the upregulation of gene functions related to histone remodeling, chromatin organization, regulation of autophagy, and positive regulation of ubiquitin proteins. Our results suggest that TE-activity in C. secundus is epigenetically regulated via histone modifications and that HDAC3 could play a key role in regulating queen longevity.

#### Information provision and use in ant pheromone trails

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Ants use pheromones to communicate with each other and to mark trails to food sources. In this study with *Lasius niger*, we investigated three aspects of ant pheromone deposition: (1) whether ants lay more pheromone leading to food after they have made a mistake and followed a pheromone trail that does not lead to food; (2) whether ants follow pheromone trails more when going out to find food or when returning to the colony; and (3) whether ants lay more pheromone closer to food sources that are farther away from the nest.

Our results showed that ants do not lay more pheromone leading to food after they have made a mistake. We also found that there was no difference in the amount of pheromone trails that ants followed when going out to find food or when returning to the colony. However, we did find that ants laid more pheromone closer to food sources that were farther away from the nest.

These results suggest that ants use pheromones to mark trails to food sources, but they do not adjust the amount of pheromone that they lay based on their past experiences. The difference in pheromone deposition between food sources that are close to the nest and those that are farther away may be due to the fact that ants need to mark the trails to food sources that are farther away more strongly in order to attract other ants to those sources.

These findings contribute to our understanding of how ants use pheromones to communicate and to navigate their environment.

### Effect of unbalanced diets and individual amino acids on the longevity/fecundity trade-off in a clonal ant

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Several studies have highlighted the effects of environmental factors, particularly nutrition, on the development of individual traits and caste determination. However, how food affects fecundity and longevity in social insect colonies remains unclear. Clonal ants, such as *Platythyrea punctata*, are a suitable model to study the direct effects of nutrition while avoiding any other confounding factors. In this study, we investigated how different artificial diets influenced the trade-off between longevity and fecundity in P. punctata workers. Contrary to solitary insects, we found that ants confined to a high-amino acids diet lived longer but also laid more eggs than ants raised on a low-amino acids diet. Furthermore, we investigated the effects of excess and deficiency in three essential amino acids known to play crucial roles in longevity and fecundity: methionine, leucine, and tryptophan. Our results revealed that manipulating methionine (either in excess or deficiency) or tryptophan (deficiency) led to decreased longevity compared to the control diet. None of the other treatments had a significant impact on lifespan. However, we observed that increased leucine levels influenced fecundity: ants laid more eggs. These findings contribute to our understanding of the intricate interplay between nutrition, reproduction, and lifespan in social insects.

### Ants combine object affordance with latent learning to make efficient foraging decisions

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After a certain age, as humans we can easily guess the affordance of many objects. An object affordance refers to its' functional properties. For example, a bowl has the affordance of holding water, but a sieve does not. Here, we report that ants learn the affordance of a novel object without this attribute being rewarded or punished, and use the memory of this affordance to avoid predicted, but never experienced, crowding. Ants therefore combine latently-learned information about object affordance with information on nestmate presence. They infer that a restricted feeder cannot allow several ants to feed at once, without experiencing crowding. We provide the evidence that ants can integrate information about space availability at a food source, and the putative source of returning nestmates, to infer whether a food source is overexploited. This ability may allow ant foragers to reduce queuing costs, and improve colony foraging efficiency.

#### Effects of sinkhole habitats on the functional and behavioural traits of Myrmica ruginodis

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Climate change results in the transformation of many habitats. Therefore, areas providing various environmental conditions, like sinkholes, will become increasingly important since they contribute to the survival of species with different environmental preferences. The north-facing slopes of sinkholes are cooler and moister than the surrounding plateaus and previous studies suggest that they can maintain species with different functional traits than the plateaus. However, we have limited knowledge of how such variable habitat conditions affect the different populations of one species which occurs in the sinkholes and plateaus. Thus, we investigated the effects of sinkhole habitats on the functional and behavioural traits of Myrmica ruginodis that can highly influence their fitness. Our results show that sinkholes and plateaus had no significant effect on the functional and behavioural trait patterns. Still, the sinkholes had a significant effect on some functional (e.g., colony size, queen number) and behavioural (e.g., aggressiveness, reaction to prey) traits. Our results provide information about how species are able to adapt to different environmental conditions and highlight the diversifying effect of habitat islands that can be highly important in light of anthropogenic global climate change.

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### The expression of elongases and desaturases shed light on the CHC plasticity of honey bees (*Apis mellifera*)

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Cuticular hydrocarbons (CHCs) protect insects from water loss and mediate interand intraspecific communication. These functions depend on the composition of the CHC layer, which is determined by the diversity and the regulation of genes responsible for the biosynthesis of hydrocarbons. The expression of enzymes in the biosynthetic pathway such as elongases, which elongate the hydrocarbon chain, and desaturases, which introduce double-bonds into the hydrocarbon chain, determines the abundance and richness of compounds in an insect's CHC profile. In the honey bee (Apis mellifera), CHC profiles vary among castes, social roles, and subspecies. However, little is known about the genetic basis for such variation. Here, we examined the correlation of the expression of two elongase and two desaturase-genes to the CHC composition of nurse and forager workers of two highly divergent honey bee subspecies: Apis mellifera carnica (lineage C) and Apis mellifera iberiensis (lineage M). We found a correlation between the CHC composition and the expression of our candidate genes that is consistent with the catalytic function of the respective enzymes. Moreover, we provide evidence for differences in substrate specificity between different elongases and desaturases.

### Genetic divergence and aggressiveness within a supercolony of the invasive ant *Linepithema humile*

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Biological invasions constitute an opportunity to study the evolutionary processes behind species' adaptations. The invasive potential of some species, like the Argentine ant (Linepithema humile), has likely been increasing because they show low intraspecific competition. However, multiple introductions over time or genetic divergence could increase the probability of intraspecific competition, constituting barriers for their dispersal and thus, decreasing invasive success. Here, we studied the genetic and behavioural variability of L. humile workers collected at six locations on the NW coast of the Iberian Peninsula, a possible scenario for multiple introductions and population divergence, due to its high level of maritime traffic and complex coastal geography. We analysed behaviours related to spatial navigation (exploration, wall-following), resources acquisition, and competition (inter and intraspecific aggressiveness) through two relevant seasons for the nest ecology: spring and autumn. Genetic analyses using microsatellites indicated that the nests studied belonged to the most spread supercolony in South Europe. However, we identified the existence of two genetically differentiated clusters in Galiza. Lethal interactions were found between workers from different and similar genetic clusters, but a trend suggests higher agonistic behaviours between the two genetic groups. Genetic differences were positively correlated with the geographical distance, but aggressiveness was not correlated with any of them. Ants from each of the tested nests expressed different behaviours with high plasticity through time. Ants from all nests showed more exploration and aggressiveness, less wall-following and faster detection of food in autumn than in spring, with no intraspecific aggressiveness observed in spring. Our findings suggest competition between nests of the same supercolony and behavioural seasonal variability, supporting the hypothesis of divergent evolutionary processes. The results of our work question the assumed unity of supercolonies of this species and offer insights for understanding the future adaptation of *L. humile* in the introduced areas.

#### Social immunity behaviors in ant-nematode interactions

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Ant colonies face a multitude of different pathogens frequently, against which they are protected by individual and collective disease defense. In the latter, colony members cooperatively perform sanitary care behaviors and hygiene measures, which allows disease control by "social immunity". It offers effective means to fend off, for example, fungal infections, but the impact in worm-borne disease in less known. By using a complex nematode-bacteria pathogen model, we explore the effects of nematode disease in ant colonies. Our studies show that brood of the black garden ant *Lasius niger* is highly vulnerable to the predatory nematode *Heterorhabditis bacteriophora*, while adults perform protective behaviors including poison spraying and leg scratching and therefore rarely get infected. Moreover, adults provide sanitary care to brood, which prevents most nematode infections in the offspring and greatly increases its survival, as well as prevents disease transmission through the colony.

# Does the macronutrient composition affect the survival of the *Myrmica scabrinodis*, Nylander 1846 (Hymenoptera: Formicidae) ants infected with the ectoparasitic fungus *Rickia wasmannii*?

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Macronutrients play an important role in the proper functioning of organisms, e.g. in the defense mechanisms against pathogens. Previous studies investigated how ants responded to different diets with altered macronutrient composition after being infected with a generalist fungus. In our study, we investigated how artificial diets impact the survival of Myrmica scabrinodis ants infected with the specialist ectoparasitic fungus Rickia wasmannii. We collected infected and healthy ant nests. Old workers were offered three different diets, which varied in their amino acid to carbohydrate ratio. Additionally, a group of ants was subjected to food deprivation. We monitored the survival of the individuals for 56 days. Following this we counted the fungal thalli present on the right side of the infected ants' heads. Our results demonstrate that a diet rich in carbohydrates increased the lifespan of the infected ants. Conversely, as expected, starvation leads to higher mortality rates. In the case of uninfected individuals only food deprivation had a negative effect on their survival, the other three diets did not yield significant effects. Our study indicates that a carbohydrate-rich diet reduces the infection's negative effect.

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### Revealing a taxonomic mystery: *Reticulitermes* sp (Isoptera: Rhinotermitidae) in Romania

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The *Reticulitermes* sp. is distributed in Asia, the Middle East, Europe, and all of North America, playing an important role in natural forest ecosystems but they are also significant pests that feed on wooden structures and buildings. The taxonomic status of several species complex is still in debate. Despite genetic and molecular studies, Eastern Europe taxonomy insights are scarce. Romania is one of these areas where the termite species are understudied. Old data stated for many decades that *Reticulitermes lucifugus* is the only occurring species in Romania. Yet, we challenged those data. In the last years, we collected termite material and carried out genetic analysis using three types of genetic markers (two mitochondrial and one nuclear). Our results show that the Romanian termites belong to a new species, belonging to the *Reticulitermes* sp. Ankara group, which needs further integrative taxonomic analysis.

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### A new puppet-master and its puppet? Behavioural manipulation in the host-parasite system between the fly *Strongygaster globula* and its ant host

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Parasites can alter a wide range of phenotypic traits in their hosts, including morphology, physiology but also behaviour. In host-parasite systems that involve altered host behaviour, parasite manipulation is often invoked. Ants are host to numerous parasites and pathogens that can cause an aberrant host behavioural phenotype. While a few examples are well-characterized, others are barely known. An example of the latter is the association between the fly *Strongygaster* globula and queens of the ant Lasius niger. Like other parasitoid diptera in the family Tachinidae, S. globula lays its eggs in or near Lasius ant queens, presumably right after the young queens leave their natal nest for their mating flight and before the mated queens seal a newly dug nest to start a colony. However, unlike most parasitoid species, S. globula does not kill its host during development, but emerges in spring after queen hibernation, when it pupates and is apparently cared for by the queen until adult emergence. Here we provide a quantitative description of the parasitized host behavioural phenotype and present data of a series of experiments aimed at validating whether the altered host behavioural phenotype is a targeted manipulation of the host by the parasite.

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#### Development of ant colonies in an urban-rural gradient

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The productivity of ant colonies is influenced by various interconnected biotic and abiotic factors, such as queen fertility, food availability, and habitat temperature. In our study, we investigated the potential influence of urban habitats on ant colony development using Lasius niger as the study species. First, we examined differences in gynes' body fat content between individuals from urban and rural locations. Second, we analysed differences in the consumption of carbohydrates, proteins, or fat+proteins between colonies from the two habitat types. Third, we tested the impact of urban and rural environmental temperature on larval development time and worker mortality by simulating the thermal conditions in climatic chambers. Our results show that gynes from rural habitats have higher fat content compared to those from urban habitats, although the quantity of each type of food consumed by ants did not differ between colonies from the two different habitat types. Finally, we observed faster larval development and higher worker mortality in colonies kept in the climatic chamber with higher temperatures (urban environment) than in the cold one (rural environment). Our study suggests potential changes in colony development due to the influence of human-modified environments.

### Comparative choice assays allow simple and high-sensitivity quantification of ant feeding preference

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Getting information about food preferences in ants is critical for many purposes, including the control of pest ants. However, standard approaches rely on group level assays such as 'cafeteria experiments', based on the number of ants feeding at multiple food. In such cases, each individual likely experiences only one type of food.

Here, we demonstrate that by giving ants prior experience with a high-quality food source, they strongly reject slightly bitter food which they would normally accept. We develop a high-throughput dual feeder assay allowing individual ants to compare two liquid foods, and choose between them. This feeder is designed to ensure that ants meet both food sources, with their antennae repeatedly touching the alternative food. We use this assay to test for responses to a variety of potential food additives (quinine, egg protein, nicotine,  $\beta$ -alanine) and common toxicants (Fipronil, Imidacloprid, Spinosad). It turns out that ants don't like quinine or egg protein in their sugar. Neither do we.

This method constitutes a powerful, cheap, and easy to run tool for examining feeding preference. More broadly, we demonstrate that comparative choice is more sensitive for measuring preference than accept/reject assays and should be considered whenever preference needs to be measured.

### Combined stressors in the agriculture – investigating more effects than honey bee colony development

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Agrochemicals (e.g. insecticides or fungicides) are applied to protect plants against pest insects or fungal infections. The use of fungicides in agriculture can affect pollinating insects and soil-living consumers like ants. The quality of plantderived insects' nutrition and quantity thereof may also be impacted by agrochemicals through alterations of plants' microbial communities. Here, a field study was conducted to assess the impact of fungicide exposure and nutritional limitation on the honey bee Apis mellifera and the nectar microbiome of Brassica napus. Changes in nectar yeast community was not associated with fungicide exposure but might resulted from seasonal and regional changes. The exposure lead to limited effects on honey bee population development. Young colonies exposed to combined stressors showed reduced development that was compensated within two month. On the individual level, detoxification measured by GST enzyme activity was significantly reduced during exposure to the fungicide. However, gene expression level of GST-D1 was upregulated at the same time. When setting up honey bee colonies in spring, care should be taken to ensure sufficient food supply enabling resilience to agrochemical exposure.

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