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17th COLOSS eConference

Presented via Zoom

Full Proceedings

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DETAILED SCHEDULE**Thursday, 14 October 2021**

Time (US CDT)	Time (GMT+2)	Session Specifics
08:00	15:00	Introduction to the COLOSS eConference & Zoom & Protocols
08:15	15:15	Core Projects Monitoring, BEEBOOK, B-RAP State-of-the-Art Presentations & Questions/Answers
09:00	16:00	Break
09:15	16:15	Task Forces Varroa, Apitox, Nutrition State-of-the-Art Presentations & Questions/Answers
10:00	17:00	Break
10:15	17:15	2021 COLOSS Excellence Award Winner Presentations
10:30	17:30	Rapid Presentations Group ! (Monitoring, BEEBOOK, B-RAP, Varroa, Apitox, Nutrition themes)
11:30	18:30	End of COLOSS eConference Day 1

Friday, 15 October 2021

Time (US CDT)	Time (GMT+2)	Session Name
08:00	15:00	Introduction to the COLOSS eConference Day 2 & Zoom protocols
08:05	15:05	<p style="text-align: center;">Task Forces Breeding, Small Hive Beetle, Velutina State-of-the-Art & Update Presentations Questions & Answers</p>
08:50	15:50	Break
09:30	16:30	General Assembly
10:30	17:30	Break
10:45	17:45	<p style="text-align: center;">Rapid Presentations Group 2 (3-mins. ea.) (Breeding, Small Hive Beetle, Velutina, Survivors, Viruses Themes)</p>
11:15	18:15	COLOSS eConference Closing Remarks
11:20	18:20	Social Event – Apiculture Pub Quiz (Trivia Night)
13:00	20:00	End of COLOSS eConference Day 2

17th COLOSS eConference

TOPICS

- International online meeting of COLOSS to provide an update on the network's achievements and future directions, including online meetings for COLOSS Core Projects and Task Forces
- Annual General Assembly Meeting

WHEN

- | | |
|-------------------|---|
| 14 October | <ul style="list-style-type: none">• Core Project & Task Force 'State-of-the-Art' presentations |
| 15 October | <ul style="list-style-type: none">• Core Project & Task Force 'Update' presentations• General Assembly Meeting• Rapid Presentations by conference participants• Virtual mixer – Trivia Night |

WHERE

Zoom – Links to specific sessions will be distributed to registered participants

Auburn University Panopto – web-links to view all Workshop 1 recordings and all submitted Rapid Presentations will be provided during the conference and will be available for viewing for 2 weeks only.

ORGANIZER CONTACTS

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Jan Maehl COLOSS jan.maehl@coloss.org	

REGISTRATION

- Registration fee is 30 USD (~25 Euros equivalent), and can be paid at: <https://aub.ie/colossconference>

RAPID PRESENTATIONS

- All submitted Rapid Presentations must be accompanied by an abstracts.
- Rapid Presentations should be less < 3 min long and include one status slide.
- A submission should be submitted only once by the lead/presenting author.
- Student submissions will compete for the prestigious COLOSS Student Award.

TRAVEL & ACCOMMODATIONS

This is a virtual event. Please enjoy from the comforts of your home or office.

A note from the president

Dear colleagues,

On behalf of the organizing team, I would like to welcome you to the 17th COLOSS.

This is our 2nd e-conference, reflecting the urgent need for online networking in the times of COVID-19. Even though any online meeting cannot replace physical meetings, this will be a way forward for our global networking. Indeed, if people cannot join physical meetings due to apparent health concerns or lack of financials means, online solutions will offer a feasible approach. I therefore regard the experiences gained from one or two e-conferences as really valuable to strengthen our future global networking.

I am therefore delighted to say that we have an all-time high of registered participants for our conference, thereby reflecting a general acceptance of online events.

I would like to sincerely thank all of the people who made this meeting possible. In particular, it would have been impossible without the exceptionally organized efforts of Geoffrey Williams and the Auburn team.

Appreciation is also addressed to all contributors. Please be so kind and consider potential future activities, in particular your active participation in our Core Projects and/or Task Forces as well as joint experiments and fund raising.

Financial support for this meeting is kindly granted by the Ricola Foundation *Nature and Culture*, Veto Pharma, the Eva Crane Trust, IBRA and the Vinetum Foundation.

I am looking forward to fruitful online discussions with all of you, and hope you will enjoy this conference. I am delighted to see many new faces from all over our COLOSS globe!

Yours sincerely,

A handwritten signature in blue ink, appearing to read 'Peter Neumann', with a long horizontal flourish extending to the right.

Peter Neumann, President, COLOSS Association

Submitted Rapid Presentations

Note: Some Rapid Presentations associated with these abstracts will be played 'live' during the COLOSS Conference. All Rapid Presentations will be available on Auburn Panopto for two weeks after the event. The web-link will be distributed during the conference.

Authors (arranged by first name of first author; *indicates student)	Abstract title
*Aleksandra Splitt	COLOSS Excellence Award Winner 2021: Data Stewardship Makes a Bee-line for International Scientific Projects
Alessandra Giacomelli, Umberto Vesco, Vanni Floris, Massimiliano Gotti	Results of the UNAAPI Italian national loss and management survey - Second year (2019/2020)
*Amara Jean Orth, Carol Fassbinder-Orth	Secret Sounds of Bees: The Use of Hidden Markov Models to Predict Colony Health
*Amara J. Orth, Emma H. Curran, Eric J. Haas, Andrew C. Kraemer, Audrey M. Anderson, Nicholas J. Mason, Carol A. Fassbinder-Orth	Land use influences the composition and antimicrobial effects of propolis
Anastasios Galanos, Philippos Vardakas, Martin Reczko, Vaggelis Harokopos, Pantelis Hatzis, Efthimios M. C. Skoulakis, Georgos A. Pavlopoulos, Solenn Patalano	Bee foraging preferences, microbiota and pathogens revealed by direct shotgun metagenomics of honey
*Aneta Boksova	The Method of Determining Pesticide Repellent
Anna Gajda, Ewa Mazur, Bartłomiej Molasy	Health status of bee colonies kept in traditional hives on trees at Klodzka Valley Forest. A case study
Anna Papach, Alexis Beaupaire, Orlando Yanez, Meret Huwiler, Geoffrey Williams, Peter Neumann	The promiscuous mating system of small hive beetles (<i>Aethina tumida</i>)
Anthony Nearman, Dennis vanEngelsdorp	The Age of Bees - 50-year Meta-Analysis suggests worker honey bees have declined in age, which correlates to high colony loss rates in the U.S.
Aygun Schiesser, Selcil Karahisar Turan	Microplastics and Honeybees
Dalal M. Aljedani	Controlling Varroa mites infesting honey bees (<i>Apis mellifera</i> L.) using some essential oils and amitraz under colony conditions

Authors (arranged by first name of first author; *indicates student)	Abstract title
*Dan Aurell, Selina Bruckner, Clint Wall, Geoffrey Williams	Limited varroa control from oxalic acid treatment of splits established using queen cells
Daniel Schlaepfi, Nor Chejanovsky, Orlando Yanez, Peter Neumann	Honey bee viruses commonly infect labyrinth spiders, <i>Agelena labyrinthica</i> and may impact cocoon-building
Dirk Louis P. Schorkopf	A constructive criticism and reflection over the use of the terms "toxicity" and "sublethal effects" in bee research
Eduardas Budrys, Vesta Skrodenyte-Arbaciauskiene, Svetlana Orlovskyte, Laima Blazyte-Cereskiene	Dark European Honeybee (<i>Apis mellifera mellifera</i>) in Lithuania: supplementary data on haplotype diversity in Europe
Eric Dubois, Frank Schurr, Marie-Pierre Riviere, Marie-Pierre Chauzat	Would you participate at the next proficiency test on the quantification of honey bee viruses?
Esther E. du Rand, Zoe Langlands, Karl Crailsheim, Abdullahi A. Yusuf, Christian W. W. Pirk	Trophallactic nutrition of the small hive beetle - what is shared by their honeybee hosts?
Evangelia Kagiali, Philippos Vardakas, Aggelina Kanellopoulou, Goras Georgios, Fani Hatjina, Solenn Patalano	Four years monitoring of loss rates of honey bee colonies in Greece
Gherardo Bogo, Manuela Giovanetti, Piotr Medrzycki, Emanuele Carpana, Antonio Nanetti, Marino Quaranta, Laura Bortolotti	BeeNet: bees and biodiversity in environmental monitoring
Giannopoulou Eleni, Stamatakis George, Haristos Leonidas, Samiotaki Martina, Hatjina Fani, Patalano Solenn	Rapid detoxification of both honeybee and hive from natural oxalic and formic acaricide treatments but long-term exposure of the non-systemic amitraz
Ivan Pavlovic, Milan Stevanovic	Monitoring of important parasitic insects of bees in Serbia
Ivan Toplak, Laura Simenc, Metka Pislak Ocepek, Danilo Bevk	Determination of genetically identical strains of four honeybee viruses in bumblebee positive samples
J. Elijah Powell, Sean P. Leonard, Zac Carver, Nancy A. Moran	Probiotics composed of native gut bacteria lessen pathogen susceptibility of bees after antibiotic treatment

Authors (arranged by first name of first author; *indicates student)	Abstract title
Janez Presern, Goran Aleksovski, Sreten Andonov, Jernej Bubnic, Bjorn Dahle, Filip Jaman, Magdalena Jovanovska, Uros Kur, Brane Kozinc, Marin Kovacic, Ajda Moskriz, Borce Pavlov, Zlatko Puskadija, Aleksandar Uzunov	BeeConSel - progress report on multinational project dedicated to mating control
*Janko Bozic, Tina Porenta, Blanka Ravnjak	Beekeepers' competences to co-manage and promote biodiversity
Jasmijn Hillaert, Tim Adriaens	Supporting nest detection of yellow-legged hornet (<i>Vespa velutina</i>) by generating a hornet activity map
Jose Serrano, Meral Kekecoglu, Pilar De la Rua, Pedro Di-az Molins, Emine Sonmez, Onur Bayram Kavak, Yildirim Plastik, Raffaele Dall'Olio	Development and validation of a nutritional liquid feeding for honey bees throughout the European climate. Project BEEFEED
Julia Renz, Peter Rosenkranz, Carolin Rein	Stage specific damages of honey bee brood after varroa treatment with lithium chloride
K. Wagoner, J. G. Millar, J. Keller, J. Bello, P. Waiker, C. Schal, M. Spivak, O. Rueppell	Hygiene-eliciting brood semiochemicals as a tool for assaying honey bee colony varroa resistance
Kirsten Traynor	The Risk of Fungicides
*Krisztina Christmon, Dennis vanEngelsdorp	Factors contributing to <i>Varroa destructor's</i> remarkable size variability in U.S. managed honey bee populations
*Laura Simenc, Tanja Knific, Ivan Toplak	Tracing the origin of small hive beetles infesting Eastern and Western honey bee colonies in China
Lesya Tymochko, Galina Savchuk, Volodymyr Karavan, Ostap Palamar, Irina Panchuk	Effects of different carbohydrate diets on histological structure of the midgut and fat body mass of honey bees (<i>Apis mellifera</i> L.) under low-temperature conditions
Liendo MC, Romina Russo, Muntaabski I, Niz JMSalvador R, Scannapieco AC	Bacterially Expressed dsRNA induces <i>Varroa destructor</i> gene knockdown by honey bee-mediated oral administration

Authors (arranged by first name of first author; *indicates student)	Abstract title
Maja Ivana Smodis Skerl, Jorge Rivera-Gomis, Ivana Tlak Gajger, Jernej Bubnic, Gabriela Talakic, Giovanni Formato, Alessandra Baggio, Franco Mutinelli, Wim Tollenaers, Dries Laget, Valeria Malagnini, Livia Zanotelli, Marco Pietropaoli	Efficacy and Toxicity of VarroMed® on varroa mite in different geographical areas
Marin Kovacic, Aleksandar Uzunov, Noureddine Adjlane, Valerija Benko, Leonidas Charistos, Raffaele Dall'Olio, Giovanni Formato, Fabrizio Freda, Fani Hatjina, Dilek Kabakci, Valeria Malagnini, Asaf Otmi, Marco Pietropaoli, Zlatko Puskadija, Victoria Soroker, Ivana Tlak Gajger, Rahsan Ivgin Tunca, Claudio Villar, Ralph Buchler	Queen caging with emphasis on honey production during summer - preliminary results
*Marius Blumenschein, Christian Lippert, Peter Rosenkranz	Varroa treatment strategies in apiaries of professional beekeepers: A survey and an analysis of treatment efficacies
Mate Mackei, Csilla Sebok, Julia Voroshazi, Patrik Traj, Fruzsina Mackei, Barnabas Olah, Zsuzsanna Neogrady, Gabor Matis	One part of the mosaic: the effects of acetamiprid on the redox state of brain and flight muscles in honey bees
*Myriam Widmann, Lotta Fabricius Kristiansen, Patricia Aldea Sanchez, Alireza Arab, Alexis Ballis, Robert Brodschneider, Valters Brusbardis, Luis Cadahia, Norman Carreck, Panuwan Chantawannakul, Jean-Daniel Charriere, Robert Chlebo, Bram Cornelisen, Jiri Danihlik, Ellen Danneels, Ana Dieguez-Anton, Tobias Dittman, Constantin Dobrescu, Mariia Fedoriak, Johann Fischer, Alison Gray, Ales Gregorc, Fani Hatjina, Hlaing Min Oo, Melanie Kirby, Preben Kristiansen, Maritta Martikkala, Ewa Mazur, Wu Ming-Cheng, Franco Mutinelli, Asli Ozkirim, Aivar Raudmets, Rod Scarlett, Flemming Vejsnl's, Anthony Williams, Geoffrey Williams, Linde Morawetz	B-RAP survey reveals differences in beekeeper communication preferences at a continental level

Authors (arranged by first name of first author; *indicates student)	Abstract title
*Nganso T. Beatrice, Kannan Mani, Nurit Eliash, Rafaeli Ada, Soroker Victoria	Role of a Niemann-Pick type C2 transcript (Vd40090) in Varroa-honey bee interaction
*Rachel Ward, Mary Coffey, Kevin Kavanagh	Summer and winter honeybees, a proteomic comparison
Richard J. Hall, Hayley Pragert, Bernard J. Phiri, Qing-Hai Fan, Xiang Li, Wlodek L. Stanislawek, Claire M. McDonald, Hye Jeong Ha, Wendy McDonald & Michael Taylor	Apicultural practice and disease prevalence in <i>Apis mellifera</i> , New Zealand: a longitudinal study
*Saliha Salem Attia, Nora Chahbar, Salaheddine Doumandji, Fouzia Abed, Irfan Kandemir	Genetic diversity of Algerian honey bees <i>Apis mellifera intermissa</i> and <i>Apis mellifera sahariensis</i> based on COI-COII RFLP and sequencing
*Selina Bruckner, Lars Straub, Peter Neumann, Geoffrey Williams	Combined stressor effects of varroa mites and neonicotinoid insecticides on drones
Stephen Smith, Keith Browne, Oisin Duffy, Chiara Binetti, Eoin Mac Loughlin, Grace McCormack	Wild honeybees in Ireland
*Sual Tatlısulu, Erkey Ozgor	Creation of Cyprus propolis ID and investigation of the antibacterial effect of Cyprus propolis on nosocomial bacteria
Suman Mishra, Sneha Kumari and Kamal Jaiswal	<i>Galleria mellonella</i> : Biology of a pest and useful model host
Volodymyr Karavan, Diana Kachmaryk, Liudmyla Yazlovytska, Irina Panchuk	Combined effects of nutritional and temperature stress on protein carbonyl groups level in <i>Apis mellifera</i>
William D. J. Kirk	The International Bee Research Association (IBRA)
*Yoko L. Dupont, Mette G. Balslev, Per Kryger	Seasonal dynamics in pollen diet of honey bees in landscapes in Denmark
Zachary Y. Huang, J. Zhang, L. Yin, S. M. Salim	Honey bee swimming behavior: adaptive and impaired by pesticide

ABSTRACTS

1. COLOSS Excellence Award Winner 2021: Data Stewardship Makes a Bee-line for International Scientific Projects

Aleksandra Splitt^{1,2}

¹Institute of Nature Conservation, Polish Academy of Sciences, Al. Krakow, Poland, ²The National Institute of Horticultural Research, Apiculture Division in Pulawy, Skierniewice, Poland

The profession of a Data Steward has emerged parallelly to the development of the open science movement and is in line with a novel approach to how research outputs should be deployed by research institutions. Data Stewardship solves key issues related to the collection, processing, storage and sharing of scientific data - for example from COLOSS bee-related projects. Modern research starts with a Data Management Plan which requires solid knowledge of a Data Steward. The managed data should meet FAIR principles and be easily Findable, Accessible, Interoperable and Reusable. One of the main COLOSS goals is to coordinate international research, including the development of standard research practices. It is of great importance to establish standard data management and dissemination practices for COLOSS members in order to enable further global analyses and search for their joint additional value on the basis of equal data access and openness. With these assumptions fulfilled, research data increase in credibility, reproducibility and general usefulness. COLOSS Excellence Award 2021 sponsored by the Ricola Foundation - Nature & Culture was granted for the project "Data Stewardship Makes a Bee-line for International Scientific Projects", which is aimed at gaining a Data Steward qualification. The completion of the course of Data Stewardship will allow me not only to implement the highest standards into my bee-connected research, but also to bring new quality to the projects of my current and future coworkers from the COLOSS association. It will provide me with opportunities for scientific networking on a global level, gaining research visibility and increasing reputation. Eventually, this will give me the most effective tool to present bee research findings to the public.

2. Results of the UNAAPI Italian national loss and management survey Second year (2019/2020)

Alessandra Giacomelli¹, Umberto Vesco¹, Massimiliano Gotti¹

¹Italian National Union Beekeeper Association - UNAAPI

During the second year of the Italian national loss and management survey organized by Unaapi, a total of 28,780 hives were monitored in 10 Italian regions. The study analyzed the beekeeping techniques implemented, the colonies losses and the environmental conditions, during the period 1st April 2019- 31 March 2020. The involved beekeepers aimed at producing primarily honey (97.6%), followed by nucs (36.6%) and pollen (26.8%). The average honey production per colony settled at 19.3kg. The average annual expenses per hive for supplemental feeding was 11.7 euros, with a range between 3-38.7 euros per hive. 69.5% of beekeepers felt satisfied with the commercial products available to support the nutrition of colonies. Regarding the environmental conditions, the months in which the families suffered most from the lack of nectar were August (70%) and July (30%). Similarly, pollen deficiency was identified in August (60.7%) and July (33.9%). Sugar syrup was the most widely administered food (76.9% of beekeepers) and 78.4% was the total percentage of colonies fed. About 5l (5.011l) of syrup were used per colony, with an average number of administrations equal to 4.4. Proteic nutrition was given by 7.3% of beekeepers to 46% of their bee families. Colonies were also fed with hive products: honey was given by

26.8% of beekeepers and pollen was supplied by 2.4%, on almost a third of their colonies (30%). Early support feeding to prepare colonies for a high stress period was carried out by 32.9% of beekeepers, for an average period of 2.2 weeks (range: 1-8 weeks of feeding). Regarding colony losses during the active 2019 season, 3.7% of the colonies were lost whilst during the 2019/2020 winter season an estimated 5% of families were lost. Beekeepers reported “Varroa” (41.5%), “Weak colonies in the fall” (37.8%), “Queen failure” (36.6%) as reasons for colony losses in the winter period. The principal causes of colony losses during the active season were reported as “Varroa” (36.6%), “Queen failure” (35.4%) and “Starvation” (30.5%). 45.1% of the beekeepers reported issues related to pesticide treatments. In 83,8% case of these suspected colony losses, the apiaries presented agricultural crops within 2km² as corn (45.9%), vineyards (29.7%) and other cereals (24.3%). Regarding the Varroa destructor control, 80.5% of the beekeepers administered acaricides and several adopted Integrated Pest Management for the varroa control. 51.2% of the beekeepers used the queen caging technique in summer time, 32,9% realized the drone brood removal and the partial worker brood removal (32,7%). Beekeepers also monitored the varroa infestation, thanks to the counting mite fall (36.6%), the observation of phoretic varroa (34.1%) and its observation in drone brood (30.5%).

3. Secret Sounds of Bees: The Use of Hidden Markov Models to Predict Colony Health

Amara Jean Orth¹, Carol Fassbinder-Orth¹

¹Creighton University

Vibroacoustics are sounds and vibrations that are emitted by bees in response to stimuli and may be essential to understand more about honey bee behavior and health. Hidden Markov Models (HMMs) are sound processing algorithms that have been used to study bioacoustic signals in a wide variety of organisms. HMMs process signals in a statistical manner, and can characterize highly complex and variable signals. In this project, honey bee bioacoustic signals were collected using piezo microphones attached to the top bars of 25 colonies over the course of two months in Iowa, USA. HMMs were utilized for bioacoustic signal processing of honey bee sounds through a Matlab Hidden Markov Model Toolkit (MatlabHTK). Several colony states were used for training: queenright, queenless, virgin queen, robbers, wax moths, and rapid population decline. The model successfully recognized colony state approximately 88% of the time. These results indicate that bioacoustic signals and HMMs can accurately predict honey bee colony health states and may be a useful tool in beehive health monitoring.

4. Land use influences the composition and antimicrobial effects of propolis

Amara J. Orth¹, Emma H. Curran¹, Eric J. Haas¹, Andrew C. Kraemer¹, Audrey M. Anderson¹, Nicholas J. Mason¹, Carol A. Fassbinder-Orth¹

¹Creighton University

Honey bee propolis is a complex, resinous mixture created by bees using plant sources such as leaves, flowers, and bud exudates. This study analyzes how cropland surrounding apiaries affects the chemical composition and antimicrobial effects of propolis. The chemical composition and compound abundance of the propolis samples were analyzed using Gas Chromatography-Mass Spectrometry (GC-MS) and the antimicrobial effects were analyzed using the 50% minimum inhibitory concentration (MIC₅₀) assay against four relevant bee pathogens, *Serratia marcescens*, *Paenibacillus larvae*, *Lysinibacillus sphaericus*, and *Klebsiella pneumoniae*. Propolis composition varied significantly with apiary, and cropland coverage predicted mean sum abundance of compounds. The apiary with the highest cropland coverage had

significantly higher MIC50 values for *S. marcescens* and *K. pneumonia* compared to other apiaries. These results indicate that agricultural land use surrounding honey bee apiaries decreases the chemical quality and antimicrobial effects of propolis, which may have implications for the impacts of land use on hive immunity to potential pathogens.

5. Bee foraging preferences, microbiota and pathogens revealed by direct shotgun metagenomics of honey

Anastasios Galanos^(1,2), Philippos Vardakas^(1,3), Martin Reczko¹, Vaggelis Harokopos¹, Pantelis Hatzis¹, Efthimios M. C. Skoulakis¹, Georgos A. Pavlopoulos¹, Solenn Patalano¹

¹Institute for Fundamental Biomedical Research (IFBR), ²Department of Biology, Division of Animal and Human Physiology, National and Kapodistrian University of Athens, ³ Apiculture Laboratory, Faculty of Crop Science, Agricultural University of Athens, Greece

Honeybees (*Apis mellifera*) continue to succumb to human and environmental pressures despite their crucial role in providing essential ecosystem services. Owing to their foraging and honey production activities, honeybees form complex relationships with species across all domains, such as plants, viruses, bacteria (symbiotic and pathogenic), and other hive pests, making honey a valuable biomonitoring tool for assessing their ecological niche. Thus, the application of honey shotgun metagenomics (SM) has paved the way for a detailed description of the species honeybees interact with, in order to better assess the multiple factors governing their health. Here, we describe the implementation of optimized honey DNA extraction methodology coupled to direct shotgun metagenomics (Direct-SM) analysis, and to a computationally optimised and validated pipeline for taxonomic classification of species detected in honey. By comparing honey collected across 3 harvesting seasons in a stable apiary, we show that Direct-SM can describe the variability of sampled plant species, revealing honeybee behavioural adaptation. In addition, we reveal that Direct-SM can non-invasively capture the diversity of species comprising the core and non-core bacterial communities of the gut microbiome. Finally, we show that this methodology is applicable for the monitoring of pathogens and particularly for the biomonitoring varroa infestation. These results suggest that Direct-SM can accurately and comprehensively describe honeybee ecological niches and can be deployed to assess bee health in the field.

6. The Method of Determining Pesticide Repellent

Aneta Boksova^(1,2)

¹Czech University of Life Sciences Prague, ²Czech Bee Research Institute in Dol

Since the mid-20th century, there has been a steady rise in intensification of agricultural production in Europe. The efficiency of food and industrial raw material production has increased significantly by substituting human labour with mechanization and by utilizing large quantities of synthetic fertilizers and plant protection products. However, the intensification of agriculture ultimately causes more and more visible damage to all components within the environment. An important indication of such unfavourable changes are the deteriorating breeding conditions for the most important and useful insects - honey bees. Since it is impossible to eliminate the use of pesticides in conventional farming, it is essential to seek out ways in which the honey bee can, in the agricultural landscape, be protected from the adverse consequences of their application. One possibility is to use plant protection repellency products. However, the characteristics of pesticides in the Czech Republic and abroad do not include information regarding their repellency for bees. In agricultural practice, it would be most appropriate to choose the preparations

with the highest repellent effect on bees in order to minimize their contact with the chemicals. This prevention would not only reduce the frequency of bee poisoning but would also reduce the incidence of dangerous residues in pollen and honey, which directly impact humans. The method of direct attracting was chosen as a way of determining the degree of repellency of individual commercial preparations, but also their active ingredients, as well as the additional substances or different tank mixes. In honey diluted 1: 1 (honey: water), which had a similar consistency and composition as oilseed rape flower nectar, a preparation was pipetted in a quantity that would actually fall on 1 flower, which means 1 cm². The amount of the preparation was converted into a concentration that reflects the usual amount of nectar in flowers. A honey solution similar to rapeseed nectar was chosen as a control mechanism. A 50% honey solution and solutions with preparations were each separately poured into 2 ml Eppendorf microtubes. The prepared solutions were offered to bees in outdoor conditions, on yellow holders, at a height of 1.5 meters and about 20 meters from the apiary. The microtubes were simultaneously opened to allow bees to drink solutions. When the first microtube of each series was emptied, the volume of solution in the remaining tubes was immediately recorded. The aim of the research was to verify the repellency of insecticides, herbicides and fungicides for bees. Repellency was also established for combinations of pesticides and some of their components (active substances, solvents).

7. Health status of bee colonies kept in traditional hives on trees at Klodzka Valley Forest. A case study

Anna Gajda¹, Ewa Mazur¹, Bartłomiej Molasy¹

¹Warsaw University of Life Sciences, Institute of Veterinary Medicine, Department of Pathology and Veterinary Diagnostics, Laboratory of Bee Diseases

In recent years traditional beekeeping becomes increasingly popular. In Poland there is many projects that fund this kind of activity, so more and more traditional hives with “feral” bees appear in Polish forests. They are usually not tended to, as the popular belief states that bees will manage on their own. In this case study we examined 16 colonies found in traditional hives in Klodzka Valley Forest. All of those colonies were found dead. In this forest there were no other colonies that could be found (alive or dead). The samples were collected in May 2021. We collected everything that could be salvaged from said colonies, meaning dead bees, hive debris and combs, in some cases with brood. *Varroa destructor* was found in 81,25 % of samples, from which 46,15% examined colonies had high to very high infestation levels, however, due to a poor quality of some samples mite counts could be underrepresented. *Nosema ceranae* was found in 81,25% of samples, from which 56,8% had very high spore counts. The samples were also examined to find signs of hunger in colonies, which we observed in 15 out of 16 cases (93,75%). All dead colonies were examined for the presence of the most common bee viruses, but none were found, which was probably due to a very poor quality of the samples (they stayed dead in the hives for months prior to collection). In 3 cases we found birds nests and a marten, that could have influenced the fitness of colonies before their death. This data set, even though small, shows, that bees cannot be left unattended, mainly because in Poland there is no mite resistance amongst bee populations, nosemosis is a very common disease and, should food sources get scarce (which happens quite often in Central Europe, mostly in second part of summer) the bees will not survive without help from a beekeeper (concerning both, treatment and feeding).

8. The promiscuous mating system of small hive beetles (*Aethina tumida*)

Anna Papach^(1,3), Alexis Beaufrepaire^(1,3), Orlando Yanez^(1,3), Meret Huwiler¹, Geoffrey Williams², Peter Neumann^(1,3)

¹Institute of Bee Health, Vetsuisse Faculty, University of Bern, Bern, Switzerland., ²Department of Entomology & Plant Pathology, Auburn University, Auburn, AL, USA., ³Agroscope, Swiss Bee Research Centre, Bern, Switzerland.

Multiple mating in animals is common despite costs associated with sex. Among the benefits proposed to favor multiple mating, increased genetic diversity may be especially valuable for invasive species to overcome bottlenecks and to foster adaptation to selection pressures in the novel ranges. Small hive beetles (SHB), *Aethina tumida* (Coleoptera: Nitidulidae), are parasites of bee nests endemic to sub-Saharan Africa and have become a widespread invasive species. However, their mating system is still poorly understood. Here we investigated the mating system of these parasites and looked at the paternity skew among the offspring. In an invasive population in the USA, a combination of laboratory experiments and DNA parentage offspring analysis with microsatellite markers for field-caught SHBs were used to estimate number of mating by both sexes and paternity skew in the offspring. Our data clearly show for the first time that SHB have a promiscuous mating system with both males and females commonly mating with multiple partners. The analyzed females had mated with up to eight males and paternities were skewed towards certain males. Moreover, males showed higher levels of multiple mating with 7-13 females starting oviposition. Such promiscuous mating system may be one possible factor contributing to the invasion success of this species.

9. The Age of Bees - 50-year Meta-Analysis suggests worker honey bees have declined in age, which correlates to high colony loss rates in the U.S.

Anthony Nearman¹, Dennis vanEngelsdorp²

¹University of Maryland, ²University of Maryland

The high loss rates of honey bee colonies drive research for solutions aimed to mitigate these losses. While honey bees are often viewed as a superorganism, experiments that measure the response to stressors of caged individuals allow for inference in a controlled setting. In an initial experiment, we show that caged honey bees provisioned with various types of water have greater median lifespans than those that are not. While researching the history of water provisioning in cage studies, we observed that the median lifespan of caged honey bees has been declining in the US since the 1970's, from an average of 37.8 days to 17.3 days. In response to this, we again turned to historical record and found a relationship between this trend and a decline in the amount of honey produced per colony over the same time period. We then wanted to further understand the relationship between individual bee lifespan and colony success. Using an established honey bee population model, we simulated the decrease in historic worker lifespans, which revealed declines in downstream measures of colony population, overall honey production, and colony lifespan. Last, the colony loss rates obtained from population modelling allowed us to further model what might happen in a beekeeping operation where lost colonies are replaced. The end result were loss rates reflective of what beekeeper's experience today, which suggests the average lifespan of individual bees plays a role in the loss of entire colonies.

10. Microplastics and Honeybees

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Microplastics are mostly human-made environmental pollutants that seriously threaten ecosystems. Plastic particles smaller than five millimeters are called microplastics. Some microplastics used in industry and cosmetics are deliberately produced small. Others are formed from larger plastics, such as plastic covers, bags, greenhouse plastics or textiles that break down over time. Although there is not enough information on this subject yet, studies have increased in the last 10 years. Most studies are on marine ecosystems and most of the in vitro studies are done with aquatic invertebrates. The microplastic hazard in terrestrial ecosystems has been studied in less than 10 years and only a small part of these studies is related to pollinators. Although there are only a few studies on microplastics and honey bees, these studies reveal very important results. The aim of our study is to demonstrate the effect of microplastic particles on bees with cage experiments. Cage experiments, which provide a lot of data, especially nutrition regime and life span, have also provided many results regarding the effects of microplastics. Single comb with capped worker brood close to emergence were obtained from a honey bee colony from an apiary near Hacettepe University Beytepe Campus. The comb placed in an incubator and kept at $34 \pm 1^\circ\text{C}$ and $55 \pm 5\%$ relative humidity (RH). Emerging adult worker bees were collected and pooled on the following 3 day. 50 bees were placed in each cage. Results show that bees can easily consume a large amount of microplastic particles, which are abundant in terrestrial ecosystems, and this situation may endanger the health of pollinators and also humans through bee products. Honeybees are already under many threats, adding the negative effect of microplastics will be a more worrying situation regarding bee health.

11. Controlling *Varroa* mites infesting honey bees (*Apis mellifera* L.) using some essential oils and amitraz under colony conditions

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Varroa mite (*Varroa destructor*) is the major challenge for beekeeping worldwide, this pest is caused damage in the terms of honey yield and mortality of adult honey bees. Therefore, this study tested some essential oils and chemicals (amitraz), for the control integrated on *Varroa destructor* and effected of *Apis mellifera* L. Five essential oils (garlic oil, Peppermint oil, Cinnamon oil, thyme oil, Lavender oil) of plant natural products, and chemical pesticide (amitraz) were evaluated against varroa mite infested in the honeybee colonies, and the fallen mites were monitored through the sticky card placed at the bottom of the hive. Data was recorded after 1,3, 7, 15, 22 days of treatment, under colony conditions. The natural approaches employed in the control and spread of bee illnesses have been demonstrated to be effective. Garlic oil and thyme oil were found to be particularly efficient against *Varroa* Mites and honey bees (*Apis mellifera* L.), Garlic oil outperformed all other treatments in terms of reducing the number of varroa mites after treatment, with a significant difference of (Mean \pm Std. Error) (9.330 ± 2.392) throughout the study, with increased *Varroa* mortality in all treatments compared to natural *Varroa* mortality during the pretreatment period. Also, the daily dead bees were counted during the study period, it is clear that there were insignificant differences between honey bee colonies with different treatments in the daily dead worker bees treated with certain oils and chemicals. In addition, all the treatments were safe for worker

bees at the applied dose. The natural approaches employed in the control and spread of bee illnesses have been demonstrated to be effective, and that essential oils can improve the health of bee families.

12. Limited Varroa control from oxalic acid treatment of splits established using queen cells

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Damage from the ectoparasitic *Varroa destructor* mite is a leading cause of honey bee (*Apis mellifera*) colony losses. *V. destructor* parasitizes both adult bees and brood during its dispersal and reproductive phase, respectively. When the brood cells are sealed by a wax capping, *V. destructor* is protected from chemical treatments. Periods with limited numbers of capped brood cells can occur naturally or be artificially induced, for example by establishing new colonies (splits) using queen cells. This method results in a partial brood break and has been promoted as an optimal treatment opportunity. Such brood breaks are an opportunity to use lower-risk Varroacides like oxalic acid. However, the efficacy of oxalic acid treatments at this opportunity is unclear. In the spring of 2021 in Alabama, we established 40 splits with approximately three frames of brood plus resource frames and adhering bees. The next day, we assessed the *V. destructor* infestation level of each split and introduced a queen cell to each. Twenty days after the introduction of the queen cell, half of the colonies were given a one-gram oxalic acid vapor treatment, while the others were left untreated. Six weeks later, the *V. destructor* infestation level was again assessed. Our results showed that a single oxalic acid vapor treatment resulted only in a limited reduction of *V. destructor* infestation. To further assess the potential of artificial brood breaks in splits as a treatment opportunity, we plan to test several lower-risk Varroacides including oxalic acid in spring of 2022. This could provide the opportunity for an effective *V. destructor* treatment during a treatment window for all beekeeping operations which start splits with queen cells.

13. Honey bee viruses commonly infect labyrinth spiders, *Agelena labyrinthica* and may impact cocoon-building

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Virus host-shifts are of major concern for human and animal health. However, host-range, transmission routes and the possible impact of these viruses on alternative hosts remain poorly understood. Several RNA viruses known from honey bees, *Apis mellifera*, are now considered multi-host species that are frequently transmitted across the arthropod community, but their occurrence and impact on spiders are poorly understood even though these generalist predators frequently consume honey bees thereby possibly promoting foodborne transmission. Here, we show that natural infections of labyrinth spiders *Agelena labyrinthica*, with honey bee viruses are common in the field and may impair cocoon building. Field-collected spiders were screened for viruses common in honey bees: Acute bee paralysis virus (ABPV), Black queen cell virus, Chronic bee paralysis virus, Deformed wing virus type A and B (DWV B), Israeli acute paralysis virus, Lake Sinai virus and Sacbrood virus. Further field-collected spiders were fed with ABPV and DWV B infected adult honey bees or not (controls) in the laboratory. The results suggest that natural infections of *A. labyrinthica* with ABPV, DWV A, DWV B and SBV are likely in the field, supporting the wide host range of RNA viruses. The laboratory data also indicate that high DWV-B titres may impair cocoon

building in female spiders. This is the first report of possible clinical symptoms of honey bee viruses in Araneae. Since labyrinth spiders without cocoons cannot reproduce, host-shifted honey bee viruses might affect the fitness of these important generalist predators, which would constitute a major concern for nature conservation.

14. A constructive criticism and reflection over the use of the terms "toxicity" and "sublethal effects" in bee research

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Here, I will briefly revisit the term "toxicity" in bees and the increasingly used and important term "sublethal effects". I will engage in constructive criticism regarding the use of both terms and - hopefully with the help of the audience - reflect over potential ways on how research may (or may not) profit from a more careful use of both terms when working with the species and subject of their choice.

15. Dark European Honeybee (*Apis mellifera mellifera*) in Lithuania: supplementary data on haplotype diversity in Europe

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Across Europe, the population of dark European honeybee (*Apis mellifera mellifera*) has been significantly affected by introduction and propagation of non-native subspecies of honeybees. In the Baltic countries, including Lithuania, the introduction of bees from abroad is in progress as well. The state of the surviving native Lithuanian *A. m. mellifera* bees was unknown until now. Thus, the aim of our study was to determine the current distribution of *A. m. mellifera* populations in Lithuania. A total of 494 honeybee colonies from traditionally managed apiaries were sampled to investigate their maternal ancestry, using the COI-COII intergenic spacer sequence of mtDNA. The results revealed that 45,3 % of the maternal lines investigated belonged to *A. m. mellifera*. They represented 15 different COI-COII spacer haplotypes. Among the latter, three haplotypes (M4b, M4b', M4g), representing 22.5 % of the studied Lithuanian *A. m. mellifera* colonies, are known in Western Europe as well. The other 12 haplotypes seem to be new to European haplotype checklists. The commonest of them, preliminary named MLT1 and differing from the M4b by a single deletion, represented 47.6 % of the studied *A. m. mellifera* colonies. Other haplotypes were scarcer; three of them were represented by single colonies. This is the first study in Lithuania that provides an insight into the diversity and distribution of haplotypes among the local dark European honeybee populations as well as the supplementary data on the occurrence of *A. m. mellifera* across Europe.

16. Would you participate at the next proficiency test on the quantification of honey bee viruses?

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The European Union Reference Laboratory (EURL) for bee health frequently organises inter-laboratory proficiency testings (ILPT) on methods used for the diagnosis of honey bee diseases. In 2022, a new ILPT will be organised on the quantification of honey bee viruses (ABPV, BQCV, CBPV, DWV, and SBV) by PCR. This ILPT will provide an assessment of the trueness and precision of the participant's data. The Coloss' members interested in such assessment are invited to register at this ILPT (contact eurl.bee@anses.fr) before the end of 2021. They should pay the shipment (according to UN 3373 requirements) of the samples from France to their laboratory. They will receive crushed bees to be tested by their own quantitative PCR methods (including viral RNA purification, revers-transcription and PCR steps). The viral load results (only quantitative results; no qualitative results will be considered) will be returned to the EURL for Bee Health in due date (one month after sample reception) in order to be compared to the other participant results according to the international statistical standard ISO 13358. Participants will receive an ILPT report by July 2022 graphically showing the accuracy of the viral loads quantified by all the participants (anonymised). The ILPT results will then be submitted for publication in an international journal. Each participant will be able to suggest one name as co-author.

17. Trophallactic nutrition of the small hive beetle - what is shared by their honeybee hosts?

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Most parasites exploiting trophallaxis in social insects rely almost exclusively on chemical mimicry to avoid aggression by host workers. The honeybee nest parasite *Aethina tumida* (small hive beetle) is unique as it relies on behavioural mimicry to induce its hosts to feed it the carbohydrate-rich contents of their crops. It is unclear whether the beetles are able to induce the honeybee workers to feed them the valuable protein-rich hypopharyngeal glandular secretions reserved for the queen, larvae and other nest mates using this strategy. Pollen is the main protein source for honeybees and is consumed and digested by nurse bees who distribute it to the rest of the colony in the form of hypopharyngeal gland secretions. Using ¹⁴C-phenylalanine as a qualitative marker for protein transfer, we show that small hive beetles successfully induce worker bees to feed them the protein-rich secretions of their hypopharyngeal glands during trophallaxis, and that females are more successful than males in inducing the transfer of these protein-rich secretions.

18. Four years monitoring of loss rates of honey bee colonies in Greece

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The honey bee is one of the most important pollinating organisms and the only one with such a close relationship with humans. In exchange for hive products, beekeepers are focused on colony care, yet successful overwintering remains a challenge in some areas. Previous studies have shown that factors

such as beekeepers' experience, apiary size, frequency of hive migration and queen replacement impact the chances of successful overwintering. The aim of our study is to verify if these factors apply to Greece but also, if the different foraging sources and acaricide treatment methods could be additional factors that can influence winter losses. In Greece, through the past four years (2018-2021), the COLOSS questionnaire for winter losses, has collected data from 877 professional and novice beekeepers through the monitoring of 91.371 hives in total. With an average of 219 beekeepers and 22.842 hives monitored each year, we were able to draw some preliminary descriptive data that illustrate the country's apicultural sector during this period. Concerning winter honey bee survival, we found that the country's average loss between 2018-2021 varies from 16,3% to 26,1%, with an overall average of 20,3%. Specific regions of northern Greece, like Florina, Chalkidiki and Imathia, along with island locations of Rethymno and Dodekanisa, showed relatively higher losses (over 34,31%), compared to the rest of the country. Our analysis also examines the preferred foraging sources and dominant acaricide treatments in these regions, compared to the rest of the country. The impact of this study is expected to prove useful not only in the understanding of the Greek apicultural sector, but also in the evolution of better beekeeping practices, which can further improve the wellbeing of honey bees.

19. BeeNet: bees and biodiversity in environmental monitoring

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BeeNet is a national monitoring network funded by the Ministry of Agricultural, Food and Forestry Policies. The network was active from 2011 to 2014, and originated from the pre-existing Apenet project (2009-2011). In 2019 the project was refinanced with a different target. Originally created with the aim of evaluating the factors that negatively affect the health of bees, now it's used to assess the degree of pollution of agro-ecosystems through chemical and pathophysiological impact indicators of the colonies, and the investigation of wild Apoidea biodiversity according to the degree of agricultural exploitation of the territory. The current monitoring network consists of approximately 300 apiaries (5 colonies each), distributed throughout the Italian territory. Some of these sentinel colonies are also implemented with high-tech detection systems equipped with sensors that apply IoT (Internet of Things) and AI (Artificial Intelligence) technologies. They collect information on internal and external environmental parameters, and on the strength of colonies, finally allowing multi-factor analyses on quantified data. We carry out colony inspections four times a year, in the initial month of each season (March, June, September and November) in all the 300 apiaries. Upon the inspections, we also collect adult workers for the detection of pathogens. In addition, we sample bee bread twice a year (in March and June) in all the colonies of all the locations, to investigate the presence of pesticide residues and the nitrogen content. With regards to wild Apoidea, in 11 regions we identified two monitoring areas, one located within an intensively agricultural ecosystem (Intensive Agroecosystem - IA) and another in a Semi-Natural Ecosystem (SE). In each site, we record once a month both the species of Apoidea present and the honey plants they visit. From the processing of the collected data, we are developing an aggregate health indicator, aimed at assessing the health of agro-ecosystems in relation to environmental conditions and agricultural practices implemented. BeeNet will finally represent a permanent platform, constantly updating information and connecting all stakeholders involved (beekeepers, farmers, local administrators). The new branch approach of BeeNet may be inspirational to other countries, and finally support food production stability and ecosystem health.

20. Rapid detoxification of both honeybee and hive from natural oxalic and formic acaricide treatments but long-term exposure of the non-systemic amitraz

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Varroa destructor is the most important reason of beehive collapse. A variety of acaricides are used by beekeepers in order to monitor the mite populations. However, the beehive productivity seems to also be affected in the long term, which indicates that residual effects of the acaricide may persist in the honey and potentially impact their health. The aim of this study was to monitor the level of intoxication after exposure to three different chemical acaricides (Amitraz, Formic and Oxalic acids), widely used in beekeeping practices, with various ranges of toxicity. Amitraz is a non-systemic acaricide causing paralysis, overexcitation and death in insects. It gets metabolized rapidly into 2,4-dimethylformamidine (DMF) and N-(2,4-dimethylphenyl)-N-methylformamidine (DMPF), and further into 2,4-dimethylaniline (DMA). Formic and oxalic acids are organic acids naturally found within a beehive. Formic is a natural component of foods, ants and the bee venom and is used as an antiseptic in honey, while oxalic acid is common in plants to repel herbivores. It is a natural part of the bee diet and it is present in honey. These acaricides and their residues were monitored 28 and 58 days after the first application of the treatments. Our experiments showed that amitraz metabolites bioaccumulate and persist in the bee system overtime, since there is no metabolic pathway to naturally remove them. Formic or oxalic acids level are both quickly restored, with oxalic levels being restored much faster than the formic. The results of this study may be useful to the beekeeping community for monitoring more efficiently the beehive welfare and productivity as well as the honey quality. Natural treatments seem to be more bee-friendly than the non-systemic ones.

21. Monitoring of important parasitic insects of bees in Serbia

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Insects from the genera Diptera, Coleoptera and Hymenoptera represent a significant health problem of bee colonies. In Serbia, the most widespread parasitic insect is the wasp *Vespula spp.* (Insecta, Hymenoptera). Wasps are usually present in beehives from August until autumn, when entering the hives and taking honey from bees can cause significant damage, especially to weak societies. In addition to stealing honey from the hive, wasps kill bees that feed to feed the young. The second most common parasitic insects are hornets, the most common of which is the European hornet (*Vespa crabro*). Hornets catch bees in flight and take them to their nests, and they often penetrate hives where they are filled with honey and bees, which they take out without any problems. They pose the greatest threat to bees in August and September, when there are many of them. Senotenuiasis is a miasm that occurs in worker bees during the summer. It rarely occurs in drones, which is explained by their low activity and in the queen because she leaves the hive only at the time of mating and at the time of swarming. Before they transform into a puppet, the larvae can turn into another dead bee and eat its tissues. The mature larva leaves the beetles, crawls on the surface of the soil, and then crawls into the ground, where after 7 to 14 days it transforms into an adult fly. The bee louse *Braula coeca* (Insecta, Diptera) is also less common in apiaries

in Serbia and is not a major problem for beekeeping. Sometimes the infestation can be so great that the queen even dies. As the last for now parasitic species of insects that was observed for the first time in 2021, the members of the genus *Meloinae sp.* (Insecta, Coleoptera), also known as blister beetles. Meloidae adults are phytophagous and often destructive pests of a wide variety of ornamental flowers and agricultural crops. The triangular stages of the larvae of these insects are extremely aggressive. In bee communities, these larvae first devour pollen and then move on to eggs and bee larvae. When they devastate a bee colony, they use bees as a means of transportation to the next hive or apiary. By spreading among bee communities, this type of larvae can certainly contribute to the mechanical spread of bee diseases. The presence of a small hive beetle or *Aethina tumida* (Insecta, Coleoptera) has not been established in Serbia. Given its presence in the surrounding countries, one should be extremely careful and control bee colonies in order to detect their presence in time. Monitoring for the presence of harmful insects should become an indispensable part of preventive apitechnical measures. It would detect and control harmful insects that cause significant problems to bee production in time. They are also extremely dangerous to human health, especially wasps, hornets and *Meloinae sp.* whose stings or secretions can cause serious allergic reactions and possible fatal outcomes.

22. Determination of genetically identical strains of four honeybee viruses in bumblebee positive samples

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In recent years there has been growing evidence that certain types of honeybee viruses could be transmitted between different pollinators. Within a voluntary monitoring programme 180 honeybee samples (*Apis mellifera carnica*) were collected from affected apiaries between 2007 and 2018. Also, from August 2017 to August 2018, a total 148 samples of healthy bumblebees (*Bombus lapidarius*, *B. pascuorum*, *B. terrestris*, *B. lucorum*, *B. hortorum*, *B. sylvarum*, *B. humilis*) were collected at four different locations in Slovenia and all samples were tested by using RT-PCR methods for six honeybee viruses. Direct sequencing of a total 158 positive samples (acute bee paralysis virus (ABPV n=33), black queen cell virus (BQCV n=75), sacbrood bee virus (SBV n=25) and Lake Sinai virus (LSV n=25)) was performed from obtained RT-PCR products. The genetic comparison of identified positive samples of bumblebees and detected honeybee field strains of ABPV, BQCV, SBV and LSV has shown from 98.74 to 100% nucleotide identity between both species. This study not only provides evidence that honeybees and bumblebees are infected with genetically identical or closely related viral strains of four endemically present honeybee viruses but also detects a high diversity of circulating strains in bumblebees, similar as was observed among honeybees. Important new genetic data for endemic strains circulating in honeybees and bumblebees in Slovenia are presented.

23. Probiotics composed of native gut bacteria lessen pathogen susceptibility of bees after antibiotic treatment

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Antibiotics have been applied to honey bee (*Apis mellifera*) hives for decades to treat *Paenibacillus larvae*, which causes American Foulbrood disease and kills honey bee larvae. One of the few antibiotics approved in apiculture is tylosin tartrate. This study examined how a realistic hive treatment regimen of tylosin affected the gut microbiota of bees and susceptibility to a bacterial pathogen. Tylosin treatment reduced bacterial species richness, phylogenetic diversity, and reduced the absolute abundances and strain diversity of the beneficial core gut bacteria *Snodgrassella alvi* and *Bifidobacterium spp.* Bees from hives treated with tylosin died more quickly after being fed a bacterial pathogen (*Serratia marcescens*) in the laboratory. We then tested whether a probiotic cocktail of core bee gut species could bolster pathogen resistance. Probiotic exposure increased survival of bees from both control and tylosin-treated hives. Thus, probiotic treatments using cultured bee gut bacteria may ameliorate harmful perturbations of the gut microbiota, caused by antibiotics or other factors.

24. BeeConSel - progress report on multinational project dedicated to mating control

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Conditions that provide control over mating in honey bee selection and breeding programs are difficult to implement. Project BeeConSel is a collaboration of five partners (Norway - NO, Sweden - SE, N. Macedonia - MK, Croatia - HR, Slovenia - SI) with a focus on establishing functional mating control for enhancement of the breeding programs through genetic improvement and conservation of local honey bees. The main goal is to evaluate the success of different tested mating control practices in local environments, and to identify the most effective ones; to design optimal mating control strategy for specific population structure. Following the guidelines and assessment gained the partners designed the field experiment work. Furthermore, a survey focused on mating control importance has been deployed within breeder community in SI, HR and MK, covering beekeepers, breeders and approaches to the mating control and breeding; altogether, 87 answers from queen breeders were collected. The design of economic aspect of the mating control was also initiated. In 2021 extensive field activities have been performed in SI, HR and MK. In SI and HR, two potential locations for mating stations were identified in each country, whereas in MK three locations were set up and one additional method (Horner system) was applied. Nuptial flights observation protocol was developed, and nuptial flights were observed at several locations in all three countries to assess the reliability of the selected locations for establishment of mating stations. The feasibility of using the potential locations will be additionally assessed using molecular identification of paternity. Sampling protocols were developed, and sampling kits were distributed. Molecular protocols and methods were established. Several techniques of DNA extraction, purification, and quality validation were tested. Reading of the selected microsatellite loci will provide insight into patriline composition to

estimate the proportion between the drone producing colonies and potential foreign drone producing colonies present on each mating site. Based on the gained experiences and collected results next-stage experiments will be planned and performed in 2022. The project "Joint Effort for Honeybee Conservation and Selection - BeeConSel" is funded by Iceland, Liechtenstein and Norway through the EEA and Norway Grants Fund for Regional cooperation.

25. Beekeepers' competences to co-manage and promote biodiversity

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Beekeepers are directly involved in ecosystem service pollination. In general, beekeepers support activities related to biodiversity and understand some basics regarding management of natural resources. To be actively involved in management and promotion of biodiversity they need to build up relevant competences. We propose specific knowledge topics to properly understand pollination services including role of melliferous plants and different type of pollinators that can sufficiently support three major fields of activities of beekeepers to take active part in management of pollination ecosystem service. Knowledge topics and fields of activities: 1. Know the fundamentals of biodiversity and role of pollination ecosystem service. 2. Know the life and needs of main species and groups of pollinators. 3. Knowledge of wild and cultivated melliferous plant species, their phenology, needs for successful growth and their usefulness. 4. Knowledge of invasive plants and their negative impacts on biodiversity including pollinators. 5. Know the activities in agriculture and landscape management that have a positive impact on biodiversity. 6. Be able to assess the biodiversity of plants in an area, using public databases on landscape and vegetation. 7. Know the instruments, including financial resources, for management of biodiversity on local, regional, and global scale. 8. Capable of organizing and providing activities to spread and diversify melliferous plants accordance to the above knowledge. 9. Capable of organizing and providing activities to increase nesting sites for wild pollinators. 10. Capable of active involvement in public activities for promotion and management of biodiversity. The proposal is built up inside Interreg IT-SI project BEEDIVERSITY. We are developing education activities along with practical workshops for the beekeepers. That can empower beekeepers to take active part in building so called "green architecture" as a part of EU common agriculture policy (CAP). Building up competences for co-management and promotion of biodiversity have already been proposed for Slovenian beekeeping sector interventions inside of strategic plan for agriculture 2021-2027.

26. Supporting nest detection of yellow-legged hornet (*Vespa velutina*) by generating a hornet activity map

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In the region of Flanders (North Belgium), the first nests of *V. velutina* were reported in 2016 and the species has been spreading since. Surveillance for hornets and their nests was organized using a citizen science approach through the platform www.vespawatch.be. Whenever an observation is reported on vespawatch.be, a coordinated network of volunteers initiates nest detection using the wick bait station method. To make best use of volunteer time it is necessary to have an up-to-date overview of active outbreaks of *V. velutina* i.e., areas with hornet activity indicating the presence of an undetected nest nearby. To this end we created an interactive online map (<https://inbo->

vespawatch.shinyapps.io/VespaWatch_app/) using Leaflet, the open-source JavaScript library for R and Github. Using all validated reports of nests and individual hornets within one season, a map indicating hornet activity was created by removing nests that have already been managed together with observed hornets that could originate from these nests. A hornet is assumed to originate from a nest if it was observed prior to nest removal and if it occurred at a distance of less than 2 km from a nest, corresponding to the maximum home range of hornets around their nest. Furthermore, a heatmap is applied on the remaining observations, with intensity based on observation density, to quickly identify hot spots of hornet activity where nest detection has to be initiated. The usefulness of this approach will be tested during the management season of 2021.

27. Development and validation of a nutritional liquid feeding for honey bees throughout the European climate. Project BEEFEED

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Honey bees are key to the ecosystems, agricultural crops and orchards for the pollination service they provide. Recently, they are facing an increasingly hostile environment due to climate change, pesticides and other stressors, that can potentially lead to nutritional deficits. For managed colonies, in addition to natural resources, support feeding has recently become a pivotal beekeeping technique, given the need to ensure strong populations throughout the season. The aim of the BEEFEED project is to develop a tailor-made liquid feed with crude protein, sugar and vitamins that meets the nutritional needs of honey bees, and promotes an optimal development of colonies of different subspecies located in their own climate environment. The provision of the liquid nutritional product will also be tested for its use in hives located in a wide range of climates (not crystallizing in cold climates). The acceptance and efficacy of the product will be validated through performance tests carried out by project members in Spain, Italy and Turkey in hives hosting the local subspecies (respectively *Apis mellifera iberiensis*, *A. m. ligustica* and *A. m. anatoliaca*) in apiaries located in regions with different climatic conditions. Additionally, a new automatic feeder for safe syrup supply will be designed and tested.

28. Stage specific damages of honey bee brood after Varroa treatment with lithium chloride

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In view of the fact that high infestation with *Varroa destructor* is still the crucial factor for losses of honeybee colonies and economic damages for commercial beekeeper, the products currently available for the Varroa treatment are insufficient. Lithium chloride (LiCl), the first varroacidal compound with a systemic mode of action, has a promising high efficacy and good tolerability when applied to adult bees, for instance in brood-free colonies (Ziegelmann et. al. 2018). Unfortunately, preliminary studies showed that LiCl harms the development of the honeybee brood, leading to significant brood damages when applied in effective concentrations of 25mM LiCl (short presentation during the 2020 COLOSS meeting). In a further experiment we now analyzed the side effects on larvae of different age. For the experimental setup we conducted a brood assessment in free-flying colonies (Schur et. al. 2003) where larvae of different age were fed over different time periods with a 25 mM LiCl sugar solution. Every other day, the viability of the brood was checked to evaluate the time of brood removal and the brood termination rate

for each experimental group. The results clearly show, that all developmental stages of the honey bee brood are damaged, however the range of these damages depend on the duration of the LiCl feeding. For instance, a continuous feeding over the entire larval development from the egg till the sealing of the brood cell resulted survival rates of only 7% while shorter application periods of 4 and 2 days resulted in significantly higher survival rates of about 25% and 75%, respectively. Furthermore, we perceived that most of the treated brood is removed after the pupation of the larvae within the sealed brood cells. Through opening and inspection of sealed brood cells we could confirm that malformations and damages peaked on day 14 of larval development, i.e., just after metamorphosis. Our results indicate that LiCl disturbs the brood development mainly during the sensitive period of pupation. An application of LiCl in effective concentrations of 25mM in breeding colonies is therefore not recommended. Currently, we focus our research activities on the development of smart applications of LiCl in order to prevent these brood damages.

29. Hygiene-eliciting brood semiochemicals as a tool for assaying honey bee colony *Varroa* resistance

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Despite control efforts, the ectoparasitic mite *Varroa destructor* (*Varroa*) and the pathogens it vectors remain the primary biological threat to the health of the honey bee (*Apis mellifera*). The social immune mechanism *hygienic behavior* is the ability of honey bee workers to detect, uncap, and remove unhealthy brood from the colony. While selective breeding of hygienic behavior represents a promising avenue for improved *Varroa* management, the ability of existing selection methods to confer *Varroa* resistance varies. Current selection methods tend to trade efficacy for efficiency, because those achieving the highest levels of *Varroa* resistance can be time-consuming, and thus expensive and impractical for widespread use. Here, we tested the hypothesis that hygienic response to a mixture of semiochemicals associated with *Varroa*-infested honey bee brood can serve as an improved tool for predicting colony-level *Varroa* resistance. In support of our hypothesis, we demonstrated that a mixture of the compounds (Z)-10-tritriacontene, (Z)-8-hentriacontene, (Z)-8-heptadecene, and (Z)-6-pentadecene triggers hygienic behavior in a two-hour assay, and that high-performing colonies (hygienic response to $\geq 60\%$ of treated cells) have significantly lower *Varroa* infestations, remove significantly more introduced *Varroa*, and are significantly more likely to survive the winter compared to low-performing colonies (hygienic response to $< 60\%$ of treated cells). These findings highlight the significant potential of the UBO assay to contribute to the control of *Varroa* through facilitation of improved breeding and management decisions. As an efficient and effective tool to predict *Varroa* resistance, the UBO assay has the potential to significantly improve honey bee health, and thus strengthen global pollination services and food security.

30. The Risk of Fungicides

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Eusocial *Apis mellifera* colonies depend on queen longevity and brood viability to survive, as the queen is the sole reproductive individual and the maturing brood replenishes the shorter-lived worker bees, keeping the colony intact and productive. Production of many crops relies on both pesticides and bee pollination to improve crop quantity and quality, yet sublethal impacts of this pesticide exposure are often poorly understood. The diversity and use of fungicides have increased in recent decades and are forecasted to intensify. Fungicides are not deemed a direct exposure risk to honey bee health by environmental agencies because this class of pesticides typically exhibits very low toxicity. High concentrations of fungicides in samples collected during pesticide monitoring studies have counterintuitively been correlated with increased incidences of fungal diseases such as *Nosema spp.* and chalkbrood. In feeding trials, continuous sublethal exposure at field-relevant doses has also been linked to an increased rate of queen events and brood loss. This class of pesticide deserves reevaluation to understand its impacts on colony health.

31. Factors contributing to *Varroa destructor*'s remarkable size variability in U.S. managed honey bee populations

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Varroa destructor an ectoparasitic mite of the European honey bee (*Apis mellifera*), and the viruses they transmit, are the most important factors driving high rates of honey bee losses in the United States. In response to concern over the discovery of mites whose size (but not genetics), suggested they were *V. jacobsoni*, we conducted a survey of the varroa mite populations across the U.S. a total sample size of 9,065 collected in 2012-2013 and 2016-2018. We found that only 33 % in 2012-13 and 79 % in 2016-17 of mites collected in the surveyed years fell within the 95% confidence interval (CI) bounds of published *V. destructor* size. We also found a shift in mite size over time from the years of 2012-13 to 2016-18. We explore several hypotheses to explain this shift, including shrinkage from long-term storage, season of collection, colony disease state, and miticidal resistance. Understanding the reason for size change may hold important clues for mite management in managed honey bee populations.

32. The Comparison of Honeybee Viral Loads for Six Honeybee Viruses (ABPV, BQCV, CBPV, DWV, LSV3 and SBV) in Healthy and Clinically Affected Honeybees with TaqMan Quantitative Real-Time RT-PCR Assays

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The viral loads of acute bee paralysis virus (ABPV), black queen cell virus (BQCV), chronic bee paralysis virus (CBPV), deformed wing virus (DWV), Lake Sinai virus 3 (LSV3), and sacbrood bee virus (SBV) were determined in samples with the use of quantitative TaqMan real-time reverse transcription and

polymerase chain reaction (RT-qPCR). A total of 108 samples of healthy adult honeybees from four differently located apiaries and samples of clinically affected honeybees from 89 apiaries were collected throughout Slovenia. Within this study, two new RT-qPCR assays for quantification of LSV3 and SBV were developed and proved to give accurate and reliable results in field samples. Statistically significant differences in viral loads of positive samples were identified between healthy and clinically affected honeybees for ABPV, CBPV, DWV and SBV, while for BQCV and SBV, no statistical differences were observed between both groups. Despite high detected LSV3 prevalence and viral loads around 6.00 log₁₀ viral copies/bee, this lineage probably has a limited impact on the health status of honeybee colonies. The determined viral loads between 3.94 log₁₀ and 13.17 log₁₀ in positive samples for six viruses, collected over ten consecutive months, including winter, present additional information of high viral load variations in healthy honeybee colonies.

33. Effects of different carbohydrate diets on histological structure of the midgut and fat body mass of honey bees (*Apis mellifera* L.) under low-temperature conditions

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The gut and fat body of insects play an essential role in metabolic activities. The midgut contacts with the consumed food, so it is under a direct influence of the eaten carbohydrates. The fat body stores the proteins, fats and carbohydrates, as well as synthesizes the majority of hemolymph proteins. We studied the morphology of the midgut and the fat body mass in honey bees (*Apis mellifera*) kept under different carbohydrate diets at conventional and low temperature regimes. Newly emerged honey bee workers were initially placed in 16 boxes (150-170 individuals per box) and maintained on a diet of a mixture of 25% fructose and 25% glucose at the temperature of 28°C and 80% relative humidity. Then they were maintained on four different carbohydrate diets for 4 days (four boxes per diet): 1) mixture of 25% fructose and 25% glucose (control); 2) 50% fructose; 3) 50% glucose; 4) 50% sucrose. On the 10th day the temperature was switched to 14°C (overwintering temperature) for the two of each the four boxes. A 1% mixture of amino acids was added to each diet to balance the nutrition. After 6 days the midguts were removed, followed by fixation, dissection and staining with Hematoxylin-Eosin. The fat body was extracted from the abdomen of the bees using chloroform. The morphological studies revealed some histological changes of the midgut of bees being kept on different carbohydrate diets both at +28°C and +14°C: the increased holocrine secretion, the pyknosis of the nuclei of columnar epitheliocytes (chromatin lumps are visible), the occasional vacuolation of the cytoplasm, the thickening of the peritrophic membrane. Such morphological features are generally considered evidences of pathogenesis. Also, the rounded and elongated formations among the layers of peritrophic membrane were observed. These formations are considered by us as symbiotic microflora, possibly yeast-like fungi. The most mentioned-above changes of the midgut were observed in bees maintained at 50% fructose and 50% sucrose. However, the midgut structure of the bees maintained on glucose solution (50%) was most similar to the control bees. The mean value of fat body mass of bees being kept at +28°C ranges from 1.38 to 2.50 g I— 103/1 abdomen. The feeding by the 50% glucose and 50% fructose causes the enlargement of the fat body compared to control bees, more significantly when consuming 50% fructose. Depletion of fat body reserves is observed in the individuals being kept at +14°C. This is confirmed by a significant decrease of fat body mass compared to the individuals maintained at +28°C (0.95-1.10 g I— 103/1 abdomen). No significant difference was found between the control and experimental groups. So, the most pronounced changes of the fat body mass and the midgut structure of *A. mellifera* were revealed at the consumption of 50% fructose: at both, the conventional temperature (+28°C) and the temperature stress (+14°C).

34. Bacterially Expressed dsRNA induces *Varroa destructor* gene knockdown by honey bee-mediated oral administration

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The ectoparasite *Varroa destructor* causes serious losses of *Apis mellifera* colonies and negatively impacts on the beekeeping industry around the world. New control methods have been proposed based on the RNA interference technique. Previous reports showed that parasitized honey bees fed with double-stranded RNA (dsRNA) synthesized in vitro reduce the transcription levels of target genes in Varroa mites. An efficient and inexpensive alternative to produce dsRNA is the use of bacteria capable of achieving high levels of in vivo synthesis. In the present study, dsRNA synthesized in vivo was used to induce gene silencing in *V. destructor* and evaluate their effect on survival of both honey bees and the parasitic Varroa mites. The results evidenced that dsRNA fed to the bees engendered gene silencing in mites, inhibiting expression levels of target genes by approximately 50%. Indeed, a reduction of 50% on Varroa survival was observed when bacterially expressed dsRNAs were administered to mite-parasitized bees. Worker bees that were fed with Varroa-targeted dsRNA by oral route showed no survival differences compared to control bees, fed with sucrose solution. Our results demonstrated that specific dsRNA over-expressed in bacteria is capable of reducing mite survival by bee-mediated oral administration. This study provides an efficient and low-cost method for dsRNA production to control parasites and honey bee diseases.

35. Efficacy and Toxicity of VarroMed® on Varroa mite in different geographical areas

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Partners of different countries (Belgium, Croatia, Italy and Slovenia) of the Varroa control task force (COLOSS) evaluated acaricide efficacy and toxic effects on honey bees of VarroMed® in different climatic conditions. Our results in the tested apiaries showed an efficacy ranging from 71.2 to 89.3% in summer/autumn, and from 71.8 to 95.6% in winter. We observed some toxic effects on bees in one apiary (cold climatic conditions). We confirmed that the treatment could be efficiently applied in broodright as well as in broodless colonies.

36. Queen caging with emphasis on honey production during summer - preliminary results

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Different brood interruption methods combined with chemical treatment may successfully remove most *V. destructor* from the honey bee colonies. Further, the timely brood reduction or break may positively impact nectar intake and is occasionally used by beekeepers during the main spring flow to increase honey production. However, the experience and data on the effect of brood break on honey production during the main summer flow are scarce or contradictory. To try to find an answer, several Varroa Task Force members started an experiment in 2021. The study takes place at 10 apiaries in 7 countries (Algeria, Croatia, Greece, Israel, Italy, Spain and Turkey). At each location, three different groups with at least 5 colonies per group were established. In two groups, queens were confined for 28 days in a small cage 28 (QC1) or 14 (QC2) days before the expected main summer honey harvest. The third group (C) of colonies had free laying queens to serve as a control group. In total 207 colonies are included in this year study. All colonies were evaluated for strength (number of frames with bees and brood) every 14 days after the experiment started. The net weight of extracted honey for each colony was measured. Adult bee infestation was measured at the beginning of the experiment and 42 days after the treatment of the control group. Queen caging groups were treated by OA trickling 28 days from the day of queens' caging. The C group was treated by each partner using usual practices. At the beginning of experiment, there was no difference in colony strength between groups (ANOVA, $F(2, 206) = 0.030$, $p = 0.971$). A number of frames occupied with bees was significantly lower in QC groups comparing to C group 42 days after the caging but not 56 days after caging. Briefly, they started and finished with equal strength. Further, there was no significant difference in adult bee infestation at the beginning of the trial. However, at the last measurement (42 days after the treatment of QC1 and C group and 28 days after the treatment of QC2 group) significant differences are found ($H = 12.825$, $p = 0.002$). Namely, QC1 group had lower infestation comparing to C group. In the analysis of honey production, apiary and group as fixed factors were used in the model. Apiary as a factor had significant effect (GLM, $F(9, 206) = 64.921$, $p < 0.001$) with wide distribution of extracted honey between apiaries (average from 1.45 kg to 29.50 kg/apiary). Significant differences between groups are found ($F(2, 206) = 4.556$, $p = 0.012$), with group QC1 having lower amount of extracted honey comparing to QC2 and C groups. No significant differences were found between QC2 and C group. Preliminary results showed that QC1 group had a significantly lower final infestation, as well as honey production. On the other hand, QC2 group had the same amount of produced honey and final infestation as a control group. Measurements of colony strength before and after wintering period are planned.

37. Varroa treatment strategies in apiaries of professional beekeepers: A survey and an analysis of treatment efficacies

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In cooperation with the German Association of Professional Beekeepers (DBIB) a questionnaire survey was conducted among 67 beekeepers managing between 30 and more than 1.000 beehives per operation. This survey provides for the first time an overview on the various varroa treatment strategies in professional and semi-professional apiaries including use of veterinary drugs, biotechnical methods, diagnostic methods, and the respective costs and required working time. A striking result is that among the 67 participants of the survey no two were identical in their varroa treatment strategy. However, many beekeepers also share some operational commonalities: Oxalic acid and formic acid is used in 94 % and 63 %, respectively, of the professional apiaries of whom about 50 % had an organic certification. This might explain why the acaricides most frequently used are organic acids and that about 70 % of all survey participants included at least one biotechnical treatment method in their concept, preferably the removal of capped drone brood. The question on the economic aspects of Varroa treatment revealed that the costs for varroa treatments can have a significant impact on the profit in commercial beekeeping. We also received clear statements on the dissatisfaction with the veterinary drugs currently registered for the varroa treatment in Germany. In this context, almost 75 % of the participants strongly called for the registration of oxalic acid evaporation in Germany accompanied by a better public financial support for the varroa treatment. In on-site analyses at two commercial beekeeping operations, we analysed the efficacy of different varroa treatments in a total of 279 beehives in from summer to winter 2020. We recorded enormous differences in mite infestation rates and population dynamic of the bee colonies between the two apiaries and between different apiaries of the individual operations. In heavily infested colonies, up to 10 single treatments over the entire season were necessary to prevent colony damages or losses. This underlines that the varroa treatment represents a considerable cost factor in commercial apiaries still. This study also shows that even for experienced commercial beekeepers the varroa problem is still unsolved and that there is an urgent need for effective, tolerable, and easy-to-apply treatment methods. Financial support through the development program “Massnahmen zur Verbesserung der Erzeugungs- und Vermarktungsbedingungen für Bienenzüchterzeugnisse”, implemented by the Federal Ministry for Agriculture, Food and Consumer Protection in Baden-Württemberg.

38. One part of the mosaic: the effects of acetamiprid on the redox state of brain and flight muscles in honey bees

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In the past decades, pesticides received a prominent and inevitable role in modern agriculture. Upon appropriate application, the majority of these chemicals are stated not to cause severe detrimental effects on honeybees; however, various studies provided contrary and often ambivalent results. It is also not yet completely clarified, how neonicotinoid type insecticides (e.g., acetamiprid) affect the overall antioxidant status of pollinator insects. In the present study, we aimed to investigate the effects of acetamiprid on the redox homeostasis of honeybees. Animals have been placed into mesh cages (approx. 300 bees/group) and were kept in a dark incubator room at 34°C with 50% relative humidity. Honeybees were

fed with 50% (w/v) sucrose solution, supplemented with or without acetamiprid in three concentrations (35 mg/l, 17.5 mg/l, 8.75 mg/l feeding solution, respectively). Following 48 hours of exposure, samples were collected and stored at -80°C. Brains and flight muscles were homogenized in M PER lysis buffer supplemented with protease inhibitor. Hydrogen peroxide (H₂O₂) concentration was measured by Amplex Red method, while malondialdehyde (MDA) concentration, superoxide dismutase (SOD), and glucose-6 phosphate dehydrogenase (G6PDH) activities, along with reduced and oxidized glutathione ratios (GSH/GSSG ratio) have been monitored by specific colorimetric tests. Concentration of H₂O₂ was significantly higher following all concentrations of acetamiprid exposure in the brain compared to the controls. In accordance with these findings, MDA concentrations were also significantly elevated in the brain samples of acetamiprid treated bees, while no significant differences were observed in the flight muscles. Regarding SOD activity, no effect was found in the brain, while significant elevation was detected in the flight muscles by the effect of acetamiprid in all applied concentrations. On the other hand, the G6PDH activity intensely declined in the brain of each acetamiprid exposed group, and no alterations were detected in the flight muscles. The GSH/GSSG ratio has been decreased by acetamiprid treatment in every applied concentration in the brain, while no changes were observed in the flight muscles. According to our present data, acetamiprid significantly affected the redox homeostasis causing changes in the H₂O₂ production as well as in the glutathione system. A similar pattern was observed in the concentration of MDA, suggesting intense lipid peroxidation in the brain caused by acetamiprid-induced oxidative stress. Normal function of the pentose phosphate pathway was also affected in the brain, presumably resulting in further changes of the NADPH production and subsequently in the NADPH-dependent antioxidant systems. Taking every detail into consideration, acetamiprid intensely affected the redox homeostasis in the central nervous system of honeybees, which may contribute to its further detrimental effects.

39. B-RAP survey reveals differences in beekeeper communication preferences at a continental level

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Dramatic colony losses of honey bees were first reported in 2003, and generated a strong research focus on the subject that resulted in a constant flow of newly published information on how to maintain honey bee health. Effective communication of relevant scientific findings between bee researchers/beekeeping advisors and beekeepers is essential to enable the beekeeping sector to adapt to existing and newly emerging threats. The COLOSS core group B-RAP (Bridging Research and Practice), therefore, aims to better understand the communication preferences and needs of beekeepers. Recently, B-RAP conducted a global survey that focussed on where beekeepers obtain information on different aspects of beekeeping practice and which sources of information they prefer. For this, an online questionnaire was created, which was available in 25 languages between May 2020 and February 2021. The survey was globally advertised by 35 local coordinators. A total of 11.351 beekeepers from 98 countries answered the online questionnaire. Most answers were obtained from beekeepers in Europe (n = 9730), followed by Asia (n =

780) and North America (n = 400); and the fewest answers came from beekeepers in Latin America (n = 234). Africa and Oceania were excluded from all further analysis due to very small sample sizes (Africa n = 28; Oceania n = 16). Preferences for the type of information source differed significantly between the different continents. In Asia and Latin America, social media and web pages were the most used sources of information and were also significantly more used than in Europe and North America (social media: $\chi^2 = 745.7$, df = 12, $p < 0.001$; web pages: $\chi^2 = 217.4$, df = 12, $p < 0.001$). However, in Europe and North America beekeeping magazines and beekeeping books were significantly more used than on the other two continents (beekeeping magazines: $\chi^2 = 1388.2$, df = 12, $p < 0.001$; beekeeping books: $\chi^2 = 321.8$, df = 12, $p < 0.001$). The continents also differed significantly when comparing the most important social media channels used by beekeepers ($\chi^2 = 847.3$, df = 18, $p < 0.001$). Facebook and YouTube were the most frequently used internet-based sources in North America and Europe. In Asia, the most common source was Facebook, followed by Instagram and WhatsApp, and in Latin America, WhatsApp followed by Facebook. The results of this study showed that preferences in information source type are influenced by location. A deeper understanding of where information is most frequently retrieved by beekeepers allows beekeeping advisors and bee researchers to adapt their communication strategies accordingly. This may result in improved and more effective communication between researchers/advisors and beekeepers on a global scale and thus may facilitate the adoption of new techniques and knowledge in various beekeeping sectors, for improved honey bee management and health.

40. Role of a Niemann-Pick type C2 transcript (Vd40090) in Varroa-honey bee interaction

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Chemical cues are well-known to play a crucial role in Varroa-honey bee interactions, however, the chemosensory machinery that mediate this interaction remains unknown. Recently, via transcriptomic and proteomic analyses, we identified putative soluble carrier and membrane-bound chemosensory proteins. Focusing on the twelve putative odorant carrier proteins identified, we showed, via an exclusion of the chemosensory appendages (forelegs and gnathosoma), that transcripts of five of the twelve soluble carrier proteins were significantly lower, suggesting that they are likely involved in carrying host volatiles. Specifically, three transcripts were found to be foreleg-specific, while the other two transcripts were expressed in both the forelegs and gnathosoma. Furthermore, silencing one highly expressed and foreleg-specific transcript (Vd40090), which encodes a Niemann-Pick disease protein type C2 (NPC2) protein using RNA interference effectively disrupted Varroa host selection, acceptance and feeding as well as significantly impaired the expression of genes. Overall, our results clearly indicate the crucial role of this putative odorant carrier protein in Varroa-honeybee interaction.

41. Summer and winter honeybees, a proteomic comparison

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Apis mellifera workers can be divided into two broad subcastes: summer and diutinus (winter) workers, of which summer workers can be further categorised based on age/role within the hive. Summer and diutinus workers have one fundamentally significant difference, diutinus workers live for ~6 months whereas, summer workers live for ~6 weeks. This work conducted a proteomic comparison between diutinus and summer late hive workers, with the aim to understand the difference between the subcastes of workers. Whole cell lysate proteomic extractions were completed on the head, abdomen and venom sac to provide an overview into the workers proteome. Proteins were analysed via LC MS/MS spectrometry and resulting data was analysed to reveal an increased number of exclusive proteins in summer workers in comparison to diutinus workers. This was especially evident in the proteomes of head samples, summer workers had 433 exclusive proteins while 66 were identified in winter head samples. Summer abdomen and venom sac samples also had a higher number of exclusive proteins compared to diutinus workers. This indicates a conserved proteome of diutinus workers, possibly as a means to reduce cellular stress and increase longevity. Analysis of head and abdominal samples revealed an increase in cuticular protein in summer samples (up to 4836 and 1385 fold change increase respectively), some of which were identified to be exclusive to summer samples. Winter head and abdomen analysis identified an abundance of xenobiotic metabolism proteins (up to 92 and 14.8 fold change increase respectively), of which some were identified as exclusive to winter samples. Summer venom sac samples had a higher abundance of bee venom toxins, phospholipase A2 precursor (694.82 fold change increase) and melittin precursor (658.29 fold change increase), in comparison to winter samples. This suggests that there is an increased concentration of bee venom toxins in summer hive workers in comparison to diutinus workers. The differences in protein group abundance between the samples highlights a substantial variation between summer and diutinus bees. Work conducted here provides a novel insight into the difference between summer and diutinus caste and how pests, diseases and treatments may interact the winter bees in comparison to summer workers. This is particularly important for future Varroa studies as a reduction in cuticular protein expression in diutinus workers could make them more susceptible to parasitisation by the ectoparasite.

42. Apicultural practice and disease prevalence in *Apis mellifera*, New Zealand: a longitudinal study

Richard J. Hall¹, Hayley Pragert¹, Bernard J. Phiri¹, Qing-Hai Fan¹, Xiang Li¹, Wlodek L. Stanislawek¹, Claire M. McDonald¹, Hye Jeong Ha¹, Wendy McDonald¹, Michael Taylor¹

¹The Ministry for Primary Industries, New Zealand

In New Zealand, the introduced honey bee (*Apis mellifera*) is a valuable production animal, providing pollination services for horticultural crops and significant export volumes of honey, especially manuka honey. Honey bees in New Zealand are free from a number of significant diseases and pests such as European foulbrood, acarine disease, small hive beetle, Israeli acute paralysis virus and tropilaelaps mites. We sought to determine the health status of honey bees in New Zealand using a longitudinal study that followed 60 beekeepers over 2.5 years, ascertaining disease and pest status in their elected study apiary and interviewing them every spring and autumn. Participant beekeepers accounted for the management of approximately 12% of the beehives registered in New Zealand. Differences in beekeeping practices were observed between the North Island and the South Island. *Nosema ceranae* was found almost

exclusively on the North Island and did not displace *Nosema apis* over the course of the study. *Lotmaria passim* showed a reverse-phase seasonality to nosema, peaking in autumn at near 100% prevalence. The prevalence of *Varroa* in apiaries varied seasonally between 45.0% and 46.7% in spring and between 65.0% and 69.5% in autumn, with most infestation rates below 3 mites per 100 bees. The detection rate of symptomatic American foulbrood disease during our hive inspections was very low, between 0.00% and 0.85% hive-level prevalence dependent on the season. This study sets a foundation for understanding honey bee health in New Zealand.

43. Genetic diversity of Algerian honey bees *Apis mellifera intermissa* and *Apis mellifera sahariensis* based on COI-COII RFLP and sequencing

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To investigate the genetic diversity of two Algerian subspecies, we analyzed the mitochondrial DNA of 92 samples from 18 different locations from the North and South of the country based on the E2-H2 amplification of COI-COII intergenic region. Besides RFLP analysis of mtDNA COI-COII intergenic region by using Dra I, we also sequenced 50 honey bee colonies displaying different restriction enzyme profiles. Our data revealed the presence of four different mtDNA haplotypes at various frequencies (A8, A9, C2 and M4). based on the haplotype diversity we can infer that the Algerian honeybee population mainly consists of two haplotypes namely A8 (30.48%) and A9 (63.41%), the latter being most frequent in native region of Saharan honeybee *Apis mellifera sahariensis* (85.71%) than in rest part of the country (52.72%). Moreover, a low genetic introgression (6.09%) of non-local mtDNA haplotypes (C2 and M4) was also observed, which may be explained by the importation of foreign honeybee queens to Algeria. The sequencing of 50 samples revealed the presence of 15 haplotypes not been described previously (6 sequences of A9, 6 sequences of A8, 2sequences of C2 and 1 sequence of M4), those sequences are available in NCBI. On other hand the phylogenetic tree displays the group of haplotype A8 derived from haplotype A9. Thus, we suggest that the haplotype A8 is more recent than the A9.

44. Combined stressor effects of *Varroa* mites and neonicotinoid insecticides on drones

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Honey bees (*Apis mellifera*) encounter various biotic and abiotic stressors including the parasitic mite *Varroa destructor* and neonicotinoid insecticides. Even so, the potential for these stressors to interact in their effects drones is severely understudied. To address this knowledge gap, we employed a fully crossed experimental design to assess potential interaction effects of neonicotinoids and *V. destructor* on drone mass, survival and sperm quality traits. Known-age cohorts of drones were obtained from colonies that either received pollen patties containing field-relevant concentrations of two neonicotinoids (4.5ppb thiamethoxam and 1.5ppb clothianidin) or which did not. Upon emergence, drones were assessed for *V.*

destructor infestation, and kept in laboratory cages based on treatment group allocation: 1. No neonicotinoid / No *V. destructor*, 2. No neonicotinoid / Yes *V. destructor*, 3. Yes neonicotinoid / No *V. destructor*, and 4. Yes neonicotinoid / Yes *V. destructor*. Once drones reached sexual maturity, sperm quality traits were measured. Our results showed that neonicotinoids increased drone emergence body mass, while *V. destructor* caused a significant reduction. Both stressors combined seemed to interact antagonistically on drone body mass. Our findings also highlight that neonicotinoids and *V. destructor* individually can significantly reduce drone survival, and provide novel evidence for a synergistic interaction between the two stressors. Contrary to our expectations, sperm quality traits were not affected by neonicotinoids and *V. destructor*, either alone or in combination. Nonetheless, reduced drone survival until sexual maturity could severely affect honey bee colony and population health given the importance of drones to mating.

45. Wild honeybees in Ireland

Stephen Smith¹, Keith Browne¹, Oisín Duffy², Chiara Binetti¹, Eoin Mac Loughlin¹, Grace McCormack¹

¹National University of Ireland Galway, ²National Biodiversity Data Centre

After investigating the presence of wild honeybees in Ireland between 2016 and 2019 and determining their genotype and potential survival capacity, the wild honey bee study was launched. In association with the National Biodiversity Data Centre a recording tool was established (<https://records.biodiversityireland.ie/record/wildhoneybeestudy>) where members of the public could report the locations and certain characteristics of wild colonies via an online portal. Characteristics such as cavity type, height, aspect, and location were reported as well as the recorders' confidence in identification. Additionally, whether the colony was accessed by a beekeeper and the estimated and/or known age of occupancy were recorded. After further validation, and where feasible, colonies are sampled for further verification and research. A subset of the recorders agreed to monitor their colony's activity multiple times per year, thereby becoming custodians and part of a wild honey bee community. In addition to >200 wild nests reported between 2016 and 2018 there are a further 390 reports so far between 2019 and 2021, although not all are validated yet. The majority of wild colonies have been genotyped as *Apis mellifera mellifera* via mitochondrial and SNPs/Mitochondrial data which has also indicated that the number of hybrids has increased in certain areas. Colony level (pool-seq) and drone genome sequencing has commenced on wild colonies to help investigate the impacts of introgression due to hybridisation with imported strains and the adaptive strategies of resilient colonies.

46. Creation of Cyprus propolis ID and investigation of the antibacterial effect of Cyprus propolis on nosocomial bacteria

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Propolis is a raw natural product produced by *Apis mellifera* for hive defence and sterilization purposes. Scientists have also investigated propolis in terms of different biological activities such as antimicrobial,

anticancer, anti-inflammatory, anti-fungal, and antiviral effects for many years. These activities depend on the chemical composition of propolis, and the composition is changed according to the botanical origin and sampling site. Therefore, the propolis ID has been created in many places in the world by researchers. Propolis contains more than 300 components, and 22 of them should be taken for the people's health. Many known biological activities of propolis depend on the presence and amount of components. Bacteria, viruses, fungi, and parasites are responsible for nosocomial infections. The major global healthcare problem in the 21st century is serious infections caused by bacteria that have become resistant to commonly used antibiotics. This creates new drug requirements, so new raw material necessities. In this study, propolis samples from Cyprus, where propolis ID was not determined before, were investigated for the new raw material requirements. 13 propolis samples were collected from different locations in Cyprus, and ethanol extraction of Cyprus Propolis (CP) composition was analyzed by GC-MS. Three concentrations (200, 100 and 50 µg/ml) of CP were treated with the three Gram-negative nosocomial bacteria which were *Escherichia coli* (ATCC 25922), *Klebsiella pneumoniae* (ATCC 13883), and *Pseudomonas aeruginosa* (ATCC 27853) for the measure the zone inhibitions by the disc diffusion method. The results showed that propolis samples prevented proliferation on *E. coli* and *P. aeruginosa* while no propolis has an effect on *K. pneumoniae*. This situation may be due to the fact that *K. pneumoniae* is encapsulated. All samples had an effect on *E. coli* but there was no significant difference between them. Although all samples were effective on *P. aeruginosa*, the most effective one was found statistically as CP5. Cis-calamenene, 2,3-dihydro-benzofuran, 2,6 dimethyl hept-5-1-al, 2-pentanone,1-[5-(3-furyl) tetrahydro-2-methyl-2-furyl]-4-methyl-, cyclopentanone,2,3-dimethyl-,cis/trans, Guaiacol,3-allyl- (CAS), Naphthalene,2,6-dimethyl- (CAS) 2,6-dimethylnaphthalene, Phenol,4-ethenyl-2-methoxy-, Î±-calacorene, Î±-curcumene were found in only CP5 sample. The detection of some substances in propolis samples for the first time and the high antibacterial effect of these propolis samples have led to new research on the use of these substances in propolis as a possible antibacterial agent.

47. *Galleria mellonella*: Biology of a pest and useful model host

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¹Babasaheb Bhimrao Ambedkar University (A Central University), Uttar Pradesh India

Galleria mellonella, commonly called as greater wax moth, is a threatening pest of honey bee found in sub-tropical and tropical region around the globe. Adult moth lay eggs in weak colonies of honey bees where the larva of greater wax moth feed on the unsealed cells filled with pollen, stored honey and bee brood. They cause 'galleriasis' and in severe conditions of infestation, absconding of bees has also been reported. Infestation by *Galleria mellonella* is considered as one of the major reasons behind decline of wild and feral honey bee populations. The pest is definitely a nuisance for honey bee as well as bee keepers, but with the changing times, it has emerged as an important model host for in vivo studies of pathogenicity of many viruses, and bacteria. Thus, its use as a model organism has certainly shifted the pressure off mammalian model hosts. Studies have been carried out on *Galleria mellonella* to understand the mechanism of infection, diagnostics and therapies of many infectious diseases. From the 1980s they are widely used in baiting techniques, to study infection models and hence their role as pest and as a virus-transmitting agent has been overshadowed limiting our knowledge. This paper comprehensively reviews a large number of available literature to present a more balanced information on the biology, distribution and the economic perspective on the importance of *Galleria mellonella* in apiculture as well as their newly emerged role as a model host for studies on pathogenicity.

48. Combined effects of nutritional and temperature stress on protein carbonyl groups level in *Apis mellifera*

Volodymyr Karavan¹, Diana Kachmaryk¹, Liudmyla Yazlovytska¹, Irina Panchuk¹

¹Yuriy Fedkovych Chernivtsi National University; Chernivtsi; Ukraine

Nutrition plays an important role in supporting strong and healthy honey bee colonies. However, in some periods of the year are natural resources limited and insufficient for the colony's needs. For this reason, beekeepers normally sustained colonies with additional sources of carbohydrates. A mixture of sugar (sucrose) and water or corn fructose syrup are commonly used to feed honey bee colonies that do not have sufficient stores to survive during winter. However, there is some evidence of negative effects of supplemental these feeding of bees. Winter colony losses caused by a number of interacting stress factors. Temperature influences physiological and biochemical processes and determines the behavior of honey bees. It is known that the influence of stress factors may increase reactive oxygen species (ROS) levels. Proteins are one of the major targets of ROS. As a result of their influence in the side chains of proteins carbonyl (CO) groups are formed. The use of protein CO groups as biomarkers of oxidative stress has some advantages over the measurement of other oxidation products due to the relatively early formation and stability of carbonylated proteins. The aim of our study was to evaluate the protein carbonyl groups content in summer generation worker bees of *Apis mellifera* in laboratory conditions under the prolonged combined effects of low temperature and nutritional stress. The experiment was performed on the apiary of the Chernivtsi National University. One-two-days-old bees were transferred from the frames to the cages (about 300 bees each) and kept during 3 days at +28°C on a diet with mixture of 25% glucose + 25% fructose. After that, the insects were transferred for 5 days to diets with various carbohydrate components: (1) 50% sucrose, (2) 50% glucose, (3) 50% fructose and (4) mixture of 25% glucose + 25% fructose. Then half of the bees were exposed to low-temperature (+ 14°C) stress for 7 days. The level of protein CO groups was measured in tissues of the head, thorax and abdomen of bees. It was shown that 50% glucose diet led to an increase of CO groups' levels in head tissues but a significant decrease in abdominal tissues compared to another diets when the bees were kept at +28°C. Sucrose-fed bees kept at low temperatures showed a significant increase of CO groups' levels in thorax tissues but a significant decrease in head and abdominal tissues compared to the bees, kept at optimal temperature. At the same time glucose + fructose- and fructose-fed bees kept at +14°C was a significant decrease this value only in abdominal tissues compared to the bees, kept at +28°C. Our data show that the prolonged combined nutritional and temperature stress leads to tagma-specific modification in the level of protein CO groups.

49. The International Bee Research Association (IBRA)

William D. J. Kirk¹

¹Keele University, UK

The International Bee Research Association promotes the value of bees by providing information on bee science and beekeeping worldwide. It publishes two journals (Journal of Apicultural Research and Bee World) and also publishes books on bee science and beekeeping. Please join IBRA to support this valuable work. Our home page is www.ibra.org.uk

50. Seasonal dynamics in pollen diet of honey bees in landscapes in Denmark

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Pollen diet composition affects bee health and colony survival. However, pollen collection by bees is highly influenced by availability of pollen producing flowers in the landscape surrounding the apiary. Furthermore, quantity and species composition of dominant pollen providing plants changes through the season. We investigated the composition of newly collected bee bread in apiaries located in 30 landscapes across Denmark. Citizen scientist beekeepers were instructed in collecting bee bread once every three weeks throughout a field season (up to 10 times per apiary). The botanical composition of pollen was determined to genus or family level by microscopic palynological analysis. Furthermore, we carried out a landscape analysis, quantifying potentially flower-containing areas within a three km radius of the study apiaries. Most landscapes consisted of a mixture of different land use types, including agricultural, urban and natural areas. A total of 99 pollen types were identified from 258 samples of bee bread. However, few pollen types commonly and abundantly occurred in the samples. These were *Pirus/Prunus*, *Brassica*, *Acer*, *Trifolium repens*, *Taraxacum* and *Salix*, which are typically associated with farmland, the dominant landscape type in Denmark. However, considerable variation was found in composition of pollen collected by bees in different landscapes. A seasonal change was observed in the pollen composition, and a hump-shaped distribution of pollen diversity was found, pollen diversity peaked in May.

51. Honey bee swimming behavior: adaptive and impaired by pesticide

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Honey bees have recently been shown to have a unique swimming behavior (PNAS, <https://doi.org/10.1073>) upon dropped onto a water surface. What is not known is whether this swimming behavior is simply a "panic response" or is it adaptive behavior. We dropped honey bee workers to the center of a water bowl (diameter: 21.6 cm) and let the bees swim toward the edge. One piece of paper with a black area was presented to one direction of the bowl. We found a much higher percentage of bees swam toward the black area than expected if they swam randomly (Rayleigh test, $P < 0.01$). We also found that bees that were fed a sublethal level of pesticide failed to show a preference to the dark area (Rayleigh test, $P = 0.36$). We therefore conclude that honey bees, when dropped to a water surface, are behaving in an adaptive fashion: that they swim to a darker area, which presumably represents the closest bank of a small pool of water. Further, this adaptive behavior is changed when bees were pre-fed with a pesticide.

MEETING ATTENDEES

Last Name	First Name	Country
Aljedani	Dalal	Saudi Arabia
Amiri	Esmail	United States
Andonov	Sreten	Macedonia
Antunez	Karina	Uruguay
Anwer	Asmaa	Egypt
Arab	Alireza	Iran
Arjun	Rajini	India
Aurell	Dan	United States
Beaurepaire	Alexis	Switzerland
Benko	Valerija	Croatia
Bienkowska	Malgorzata	Poland
Blacquiere	Tjeerd	Netherlands
Blazyte Cereskiene	Laima	Lithuania
Blumenschein	Marius	Germany
Bocquet	Michel	France
Bogo	Gherardo	Italy
Boksova	Aneta	Czech Republic
Bono	Pat	United States
Bortolotti	Laura	Italy
Bouga	Maria	Greece
Bozic	Janko	Slovenia
Branchiccela	Belen	Uruguay
Brandt	Annely	Germany
Broccard-Bell	Heather	Canada
Brodshneider	Robert	Austria
Bruckner	Selina	United States
Brus	Jan	Czech Republic
Brusbardis	Valters	Latvia
Bubnic	Jernej	Slovenia
Budge	Giles	United Kingdom
Cadahia-Lorenzo	Luis	Norway
Canedo	Andres	Brazil
Carreck	Norman	United Kingdom
Chantawannakul	Panuwan	Thailand
Chapman	Nadine	Australia
Charriere	Jean-Daniel	Switzerland
Chejanovsky	Nor	Israel
Chlebo	Robert	Slovakia
Chong	Genesis	United States
Christmon	Krisztina	United States

Last Name	First Name	Country
Claeys Bouuaert	David	Belgium
Coffey	Mary	Ireland
Collin	Joanna	France
Cornelissen	Bram	Netherlands
Costa	Cecilia	Italy
Costa	Cristina	Portugal
Crailsheim	Karl	Austria
Dahle	Bjorn	Norway
Dainat	Benjamin	Switzerland
Dall Olio	Raffaele	Italy
Dalmon	Anne	France
Danihlik	Jiri	Czech Republic
Danneels	Ellen	Belgium
Dayioglu (Gur)	Miray	Turkey
De Clercq	Jean-Jacques	Belgium
De Jong	David	Brazil
De la Rua	Pilar	Spain
Di Prisco	Gennaro	Italy
Diaz	Rosana	Uruguay
Dietemann	Vincent	Switzerland
Dobrescu	Constantin	Romania
Doorn	Marnix	Chile
Douarre	Vincent	France
Drazic	Marica Maja	Croatia
du Rand	Ezette	South Africa
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Duclos	Jerome	Switzerland
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Elen	Dylan	United Kingdom
Ellis	James	United States
Erez	Tal	Israel
Erler	Silvio	Germany
Evans	Jay	United States
Eyer	Michael	Switzerland
Fabricius Kristiansen	Lotta	Sweden
Falcao	Soraia	Portugal
Fassbinder Orth	Carol	United States
Faulhaber	Marline	Germany
Fauvel	Anne Marie	United States
Fayet	Agnes	Belgium
Filipi	Janja	Croatia
Filipova	Miriam	Slovakia
Flenniken	Michelle	United States

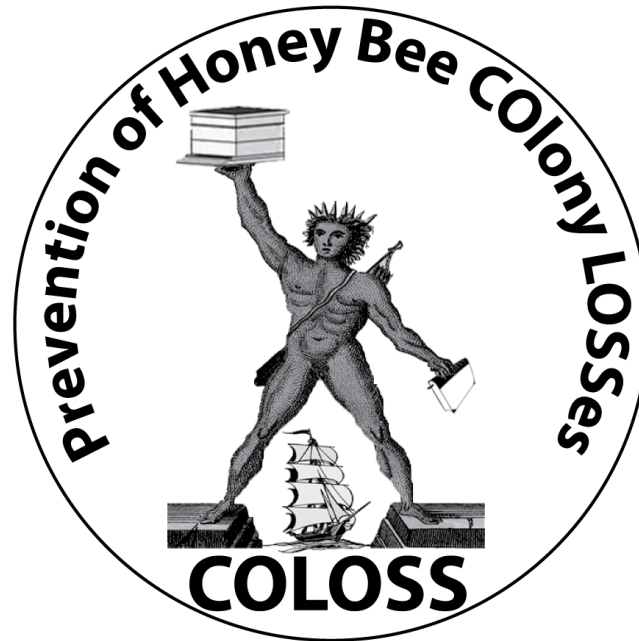
Last Name	First Name	Country
Forsgren	Eva	Sweden
Friedrich	Elsa	Germany
Gabel	Martin	Germany
Gajda	Anna	Poland
Galartza	Egoitz	Spain
Garcia-Perez	Ana L.	Spain
Gessler	Birgit	Germany
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Giannopoulou	Eleni	Greece
Giovenazzo	Pierre	Canada
Granato	Anna	Italy
Gray	Alison	United Kingdom
Gregorc	Ales	Slovenia
Grindrod	Isobel	United Kingdom
Gustavsson	Bjorn	Sweden
Guzman	Ernesto	Canada
Hamidou	Latrech	
Hatjina	Fani	Greece
Hautier	Louis	Belgium
Henriques	Dora	Portugal
Hill	Izzy	United States
Hillaert	Jasmijn	Belgium
Huang	Shao Kang	China
Huang	Zachary	United States
Iredale	Marley	United States
Jack	Cameron	United States
Jenko Rogelj	Mira	Slovenia
kaandeil	Mohamed	Egypt
Kagiali	Evangelia	Greece
Kaku	Naomi	United States
Kamler	Martin	Czech Republic
Kauko	Lassi	Finland
Keehnen	Naomi	Sweden
Kempers	Melanie	Canada
Kennedy	Peter	United Kingdom
Kezic	Nikola	Croatia
Kilpinen	Ole	Denmark
Kimura	Kiyoshi	Japan
Kirby	Melanie	United States
Kirk	William	United Kingdom
Kleckner	Kaylin	United States
Koeglberger	Hemma	Austria
Kovacic	Marin	Croatia

Last Name	First Name	Country
Kozak	Paul	Canada
Kristiansen	Preben	Sweden
Kryger	Per	Denmark
Kuipers	Rachel	United States
Kumari	Sneha	India
Laurino	Daniela	Italy
Levin	Sofia	United States
Lihoreau	Mathieu	France
Lioy	Simone	Italy
Locke	Barbara	Sweden
Lopez-Uribe	Margarita	United States
Lund	Jennifer	United States
Mackei	Mate	Hungary
Maitip	Jakkrawut	Thailand
Malcolm	Eric	United States
Manino	Aulo	Italy
Marinc	Andraz	Slovenia
Marsky	Ulrike	France
Martikkala	Maritta	Finland
Martin	Stephen	United Kingdom
Martins	Cajtia	Italy
Mazur	Ewa	Poland
McCormack	Grace	Ireland
McDonald	Rose	United States
Meana	Aranzazu	Spain
Medrzycki	Piotr	Italy
Meixner	Marina	Germany
Meshner	Colette	Canada
Milbrath	Meghan	United States
Mishra	Suman	India
Moškrič	AJDA	Slovenia
Mondet	Fanny	France
Moore	Barry	United Kingdom
Morawetz	Linde	Austria
Moro	Arrigo	Switzerland
Munoz Gabaldon	Irene	Spain
Mutinelli	Franco	Italy
Nasr	Medhat	Canada
Nave	Anabela	Portugal
Nearman	Anthony	United States
Neumann	Peter	Switzerland

Last Name	First Name	Country
Nganso	Beatrice	Israel
Nordin	Mylee	Canada
Oddie	Melissa	Norway
Olgun	Tugce	Turkey
Opitz	Michael	Austria
Orth	Amara	United States
Ota	Takahiro	Japan
Ozkirim	Asli	Turkey
Pade	Remi	France
Panziera	Delphine	Netherlands
Papach	Anna	Switzerland
Papas	Evangelos	Greece
Parejo	Melanie	Spain
Parekh	Fenali	United States
Parrent	Jeri	United States
Patalano	Solenn	Greece
Pavlovic	Ivan	Serbia
Paxton	Robert	Germany
Piccolomini	Alyssa	United States
Pietropaoli	Marco	Italy
Piot	Niels	Belgium
Pirk	Christian	South Africa
Porporato	Marco	Italy
Powell	Eli	United States
Pragert	Hayley	New Zealand
PRATT	KAREN	United Kingdom
Presern	Janez	Slovenia
Puskadija	Zlatko	Croatia
Reed	Mary	United States
Rein	Carolyn	Germany
Rennich	Karen	United States
Renz	Julia	Germany
Rogenstein	Steve	Germany
Rosenkranz	Peter	Germany
Rowe	William	United States
Rueppell	Olav	United States
Russo	Romina	Argentina
Salem Attia	Saliha	Algeria
Sanders	Jonna A	United States
Sanford	Malcolm	United States
Sawe	Thomas	Tanzania

Last Name	First Name	Country
Scaramella	Nicholas	Sweden
Schaefer	Marc	Germany
Schiesser	Aygun	Turkey
Schlaepfi	Daniel	United Kingdom
Schorkopf	Dirk Louis	Austria
Schreier	Hallel	Israel
Sgolastra	Fabio	Italy
Sheridan	Audrey	United States
Simenc	Laura	Slovenia
Simon Delso	Noa	Belgium
Skerbis	Suzana	Slovenija
Smodis Skerl	Maja Ivana	Slovenia
Soroker	Victoria	Israel
Splitt	Aleksandra	Poland
Spooner-Hart	Robert	Australia
Sprau	Lina	Germany
Stainton	Kirsty	United Kingdom
Standley	Jennifer	United States
Steeger	Thomas	United States
Steinhauer	Nathalie	United States
Steinrigl	Adi	Austria
Stevanovic	Jevrosima	Serbia
Stief	Karsten	Germany
Stoner	Kimberly	United States
Stuhl	Charles	United States
Sundby	Camilla	Norway
Tatlisulu	Sual	Turkey
Teepe	Anneke	Costa Rica
Tehel	Anja	Germany
Tlak Gajger	Ivana	Croatia
Toplak	Ivan	Slovenia
Tosi	Simone	Italy
Traynor	Kirsten	Germany
Treder	Manuel	Germany
Tymochko	Lesya	Ukraine
Underwood	Robyn	United States
Uzunov	Aleksandar	Macedonia
Van Den Bosch	Trudy	Netherlands
Van Dooremalen	Coby	Netherlands
Van Engelsdorp	Dennis	United States
Van Gent-Pelzer	Marga	Netherlands

Last Name	First Name	Country
Vejsnaes	Flemming	Denmark
Visick	Oliver	United Kingdom
Von Knoblauch	Tammo	Germany
Vranicar Novak	Anita	Slovenia
Vu	Amy	United States
Wagoner	Kaira	United States
Ward	Rachel	Ireland
Widmann	Myriam	Austria
Wilde	Jurek	Poland
Williams	Anthony	United Kingdom
Williams	Geoff	United States
Wilson	Mikayla	United States
Yanez	Orlando	Switzerland
Yazlovitska	Liydmyla	Ukraine
Zending	Sean	Netherlands
Zheng	Huoqing	China



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