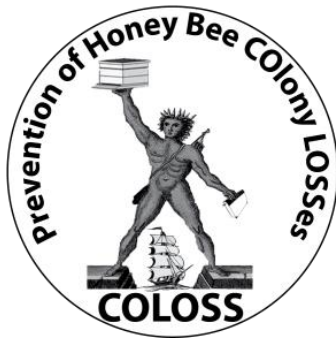


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10th COLOSS Conference

**Murcia, Spain
6-8th September, 2014**

HOTEL SILKEN 7 CORONAS, Murcia, Spain

Registration

- A registration fee of **40.- €** is required, and should be paid on site Friday (6 September) between 18.30-19.00 or Saturday (7 September) between 8.00-8.30.

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Agenda

Saturday, 6 September

Time	Session 1 – COLOSS Executive Committee Meeting 1 (for Executive Committee members)	Room
19:00-20:30	First meeting of the COLOSS Executive Committee	Mar Menor

Sunday, 7 September

Time	Session 2 – COLOSS General Assembly Meeting 1 (for all COLOSS members)	Room
08:00-09:00	Sign-in	Lorca
09:00-11.00	General Assembly discussions	
11:00-11:15	Break , with drinks & snacks	Terrace
Session 3 – COLOSS Core Project & Task Force Updates		
11:15-11.30	BEEBOOK	Lorca
11.30-11.45	Monitoring	
11.45-12.00	Beekeeping	
12.00-12.15	ApiTox	
12.15-12.30	CSI Pollen	
12.30-12.45	Sustainable Bee Breeding	
12.45-13.00	Varroa control	
13:00-14:30	Lunch & poster set-up	Romea & Gaya, resp.
Session 4 – Honey Bee Health		
14:30-14.45	Chauzet <i>et al.</i> , EPILOBEE, a European epidemiological study on honeybee colony losses	Lorca
14.45-15.00	Libor & Crailsheim , What do you feed your colonies?	
15.00-15.15	Pirk <i>et al.</i> , Honeybee colony losses in South Africa	
15.15-15.30	Ruiz-Martínez , Synergistic strategy to control varroa in Tenerife	
15.30-15:45	Short break	Lorca
Session 5 – Core Project Discussion Groups		
15.45-18.00	Monitoring	Salzillo
	BEEBOOK	Mar Menor
	Beekeeping	Rio Segura
	Spare	Lorca
Session 6 – Posters & Social Dinner		
19.00-20.30	Poster session with apéro	Gaya
20:30-	Social dinner (at own expense)	Terrace

Agenda

Monday, 8 September 2013

Time	Session 7 – Task Force Discussion Groups 1	Room (people)
09.00-11.00	Monitoring	Salzillo
	ApiTox	Mar Menor
	RNSBB	Rio Segura
	Spare	Lorca
11.00-11.30	Break , with drinks & snacks	Terrace
Session 8 – Task Force Discussion Groups 2		
11.30-13.30	ApiTox	Mar Menor
	RNSBB	Rio Segura
	Varroa control	Lorca
	CSI Pollen	Salzillo
13.30-14.30	Lunch	Romea
Session 9 – COLOSS General Assembly Meeting 2		
14.30-15.30	Updates from Core Projects & Task Force discussions	Lorca
15.30-16.30	Final GA discussions & Farewell	Lorca
Session 10 – Executive Committee Meeting 2 (for Executive Committee members)		
16.30-18.00	Closing meeting of the COLOSS Executive Committee	Mar Menor

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PLENARY ABSTRACTS

Preliminary results from EPILOBEE, a European epidemiological study on honeybee colony losses

On behalf of the consortium Marie-Pierre Chauzat^{1, 2}, Marion Laurent², Antoine Jacques¹, Cécile Saugeon², Pascal Hendrikx¹, Magali Ribiere-Chabert²

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²*Honeybee Pathology Unit, ANSES, European Union and National Reference Laboratory for honeybee health, Sophia Antipolis, France*

For the first time, an active epidemiological surveillance programme on honeybee colony (*Apis mellifera* L.) mortality (EPILOBEE) has been implemented in Europe. Each member state has developed a surveillance protocol based on guidelines produced by the EURL to harmonize the surveillance procedures. Three visits were performed by bee inspectors: before winter 2012, after winter and during the beekeeping season in 2013. Farming practices and clinical manifestations of the main infectious and parasitic diseases were recorded through a detailed questionnaire. Between autumn 2012 and summer 2013, 31,832 colonies located in 3,284 apiaries were fully visited by 1,354 bee inspectors in 17 member states (total of 8,572 visits of apiaries).

Winter colony mortality rates ranged from 3.5 % to 33.6 % with a south-north geographical pattern. In Greece, Hungary, Italy, Lithuania, Slovakia and Spain over winter colony losses were below 10%. In Germany, France, Latvia Poland and Portugal mortality rates were between 10 and 15%. In Belgium, Denmark, Estonia, Finland, Sweden and the United Kingdom winter mortality rates were above 20%. Overall rates of seasonal colony mortality (during beekeeping season) were lower than winter mortality and ranged from 0.3% to 13.6%.

The production of colony mortality rates and disease prevalence in all the member states demonstrated that the methodology was fully implementable. EPILOBEE project resulted in the compilation of a tremendous amount of data, enabling the filling of a web based database. Future data analyses will unquestionably explore the statistical links between the colony mortality and some risk factors.

What do you feed your colonies?

Anika Libor¹, Karl Crailsheim¹

*¹University of Graz
Department of Zoology*

An adequate supply with protein is essential to ensure larval rearing in honey bee colonies. Hence various artificial protein feeds were analyzed for their nutritive value for honey bees in the last decades. Because feeding with protein supplements or protein substitutes is a common tool to strengthen a honey bee colony or to give a stimulus for brood rearing in times of pollen dearth in many parts of the world. The reasons why and when protein supplements or substitutes are fed can differ due to seasonal needs of honey bee colonies considering the different climatic conditions worldwide.

We are curious about how supplemental feeding of proteins is accomplished internationally. Therefore we want to start a global study to collect data about the protein feeding tradition in different regions of the world. Another step is to assemble a list of homemade protein feeds to show "What is fed to the bees in the world".

Honeybee colony losses in South Africa

Pirk, C.W.W.¹, Human, H.,¹ vanEngelsdorp, D.² and Crewe, R.M.¹

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² Department of Entomology, 3136 Plant Sciences Building, University of Maryland, College Park, MD 20742, USA.

The mean of percentage of losses in South Africa is comparable with reports from elsewhere, although the causes appear to be different. For the first season (2009 survey) the average losses in South Africa were 20.6% and increased to 28.6% in the following season. From the data of both subspecies *Apis mellifera capensis* and *A. m. scutellata*, it was apparent that migratory beekeepers on average lost more colonies than the stationary beekeepers, especially beekeepers pollinating apples/ cherries, onions and sunflowers and those who moved their bees to the eucalypts.

Our study revealed that beekeepers migrating and managing *Apis mellifera scutellata* colonies lost almost double the number of hives that stationary beekeepers did. These losses were attributed by the respondents to the *capensis* problems in the northern parts of the country. The fact that more migratory beekeepers lost colonies to *capensis* invasion supports the previous conclusion that this social parasite relies on human facilitated transmission. Other causes indicated as being responsible for losses were small hive beetles, absconding, Varroa mites and chalkbrood. Of these Varroa and chalkbrood were universal causes of losses whilst the other three causes were of a more African specific origin.

It appears that our honeybee population is affected not only by the same factors as populations elsewhere but also by additional specific African ones. In spite of the fact that the reported losses over both seasons were higher than what is considered to be acceptable internationally, none of our beekeepers perceived the losses as threatening to their businesses or particularly unusual. The high level of losses in South Africa should be seen as a warning about the sustainability of beekeeping and thus as a serious threat to the beekeeping industry in South Africa. This concern will only be effectively dealt with through an annual poll of beekeepers using questionnaires, and a constant re-evaluation of the situation. In a nutshell we have the same problems (pests/diseases/ pathogens) but not the same level of threatened losses. Understanding colony population dynamics at a local level will contribute to an understanding at the global level.

Sinergistic strategy to control Varroa in Tenerife Island

José Antonio Ruiz ¹, Zoa Hernández² and Antonio Bentabol ²

¹ *Beekeeper Associations in Tenerife (APITEN).*

² *Casa de la Miel. Cabildo de Tenerife.*

Honeys from Tenerife just got the Protected Designation of Origin. Environmental conditions in our Island offer great diversity of flora with many endemic species. However, these same conditions make Tenerife be called "Island of Eternal Spring", allow colonies breed around year, but unfortunately, varroa can go on reproducing continuously. So a Integrated Pest Management to control varroa has been proposed by veterinarian services of APITEN (Beekeeping Association in Tenerife). The principal objective will be avoid Critical Phase after harvest. The basic sanitary calendar pays special attention to adapt therapeutic treatment to environmental conditions and timing of blooms, the multiplication methods and the supplied feeding.

This IPM will be a sinergistic strategy because, at the same time, we will try to control varroa and improve good practices in beekeeping. So, honeybee colonies will be able to reach adequate performance, vigor and tolerance to diseases and beekeepers will increase productivity and save time and money.

Finally, in this sense, it is necessary to consider if the grants for beekeepers in European Union, based on the number of hives, is according to Farrar's rule. A major European coordination of Vets, Beekeeping Technicians and Researchers would be convenient to share criteria, experiences and participate in an European Experimental Network to control Varroa.

SUBMITTED ABSTRACTS

Population dynamics of *Varroa destructor* in colonies of *Apis mellifera intermissa* in Algeria

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Population dynamics of *Varroa destructor* were studied for two years (March 2012–March 2014) in 12 *Apis mellifera intermissa* colonies located in Blida (center of Algeria, 36° 34' 00" N 2° 55' 00" E). The number of bees, the amount of open brood and capped, daily natural mortality, level of infestation of adult bees and level of infestation of the brood, was monitored.

The brood cycle and behavior of reproduction in *Apis mellifera intermissa* is set by exceptional and seasonal contrasts in climate: dry summer (June to September), with an almost complete stop brood. Autumn, relatively wet causes a second peak of activity and brood development. The values of the infestation rate of brood and bees show two peaks in August (29 % and 12.5%), this period when there is less of bees and brood in the colony.

In all colonies, the population of *Varroa* presented during the spring curve of exponential growth, which is explained by the continued presence of brood. In the growth phase, followed by a collapse of populations of mites, which in our experimental conditions, occurred from early summer, along with a weakening of colonies phase. Successive brood cycles allow the population growth of *Varroa*, while the absence of brood during the summer months has the opposite effect of reducing populations of *Varroa*. It appears that the level of *Varroa* infestation in colonies varies according to climatic conditions (seasonal) and internal conditions of each colony. In Mediterranean climates of Algeria, the milder winter climatic conditions and the possibility to collect food resources during a considerable part of the winter account for the permanent brood-rearing activity of honey bee colonies, which is relevant for the intrinsic growth rate of *Varroa* in these regions.

Interplay between climatic variables and *Varroa destructor* on the physiological performance of *Apis mellifera*

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Apis mellifera is the most important pollinator in the world. Indeed, almost 75% of crops use bees for pollination services. Nowadays, their population densities and abundances are jeopardized. In Chile the mortalities have reached between 50-60% of winter losses. For instance during 2013 we recorded about 500,000 hives less for pollination. This situation became an ecological and economic problem, especially because Chile has reached high levels of exportations of fruits and vegetables worldwide.

There are several causes for these mortalities, namely: a) monocultures, b) GMO, c) pesticide residues in the environment and inside the hives, d) new pathogens, e) increased pathogenicity and lethality of known pathogens, f) loss of genetic diversity, g) global warming, h) failures in the beekeeping management, among others. Along with these factors, the mite *Varroa destructor* is the most important sanitary problem in the beekeeping activity in Chile, but also worldwide. Indeed, these mites parasite inside the hive, drink the hemolymph in worker and brood, and kill the hive in 2-3 years if the hive is kept without treatment. Since early XXI Century, new reports indicate that this parasite is moving to new ecosystems, became complex to control, and is more aggressive and lethal for the bees than before. Until now, is not clear why. On the other hand, current environmental climate variability associated to global change poses one of the greatest threats to biodiversity. Anthropogenic impacts on the earth's climate and habitats will likely increase not only in mean temperature but in the frequency of extremely high temperatures and seasonal/daily variability in certain regions. As expected these impact are also affecting honey bees all around the world, hypothetically both directly affecting their physiological performance and tolerances as well as indirectly through an effect on the interaction *Varroa*- bees. Consequently, in this thesis, we will test the effect of *Varroa* on the physiological performance in bees when they are exposed to different conditions of ambient temperature. We will also check how the relation between host/parasite change (or not) when ambient conditions also change and how different Chilean ecotype of bees may (or not) respond to these new environmental conditions.

***Varroa destructor* mite infestation level in honey bee colonies**

Bienkowska M., Panasiuk B., Gerula D., Wegrzynowicz P., Skwarek E., Białek T.

Research Institute of Horticulture, Apiculture Division in Puławy, Poland

Varroa mite infestation level was monitored in honey bee breeding apiary in the Research Institute of Horticulture, Apiculture Division in Puławy. The aim of the research was monitoring the *varroa* mite infestation of bee colonies and comparing 4 methods used to assess the infestation level: natural mite fall, flotation method, icing sugar shaking and dead mite fall after Bee Vital application. The research was conducted from February to August 2014 in total 23 bee colonies: 10 caucasica and 13 carnica. Statistical differences in mite level infestation were stated when different methods of assessment were used.

Mapping European Nectar Flows using MODIS Satellite data and Honey Bee Scale Hives

William Blomstedt

University of Edinburgh

Insect pollination is vital to both the health of our ecosystem and food production and it is important to understand how the phenology of both pollinators and plants will be influenced by global climate change. Satellite remote sensing has frequently been used in phenological studies, but it is difficult linking the imagery with in situ events due to difference in point/pixel scale. Due to their generalized foraging of many plants over 100 km² surrounding their hive, honey bees operate at a scale which can be compared to satellite data. The MODIS Aqua and Terra satellites produce the Normalized Differentiation Vegetation Index (NDVI) which is able to show the “green-up” or start of spring (SOS). Locally, the phenology of nectar-producing flowers can be seen through the honey bee nectar flow (HBNF), which is measured by the daily weight gain of a colony of honeybees. This research investigates the link between smoothed, gap-filled NDVI data, and scale-hives from 2009-2012 in Slovenia, Switzerland, Belgium and Finland. Preliminary results show no correlation between the SOS and HBNF start day, but a notable relationship ($R^2=0.73$) between NDVI SOS and HBNF midpoint (50% day) within the Broadleaf forests of central Europe. This indicates that HBNF can be inferred with continental MODIS climate models, and that nectar flows have been advancing in central Europe.

Monitoring of melliferous plants biodiversity using pollen loads samples from commercial bee hives

Božič Janko, Podrižnik Blaž

University of Ljubljana, Biotechnical Faculty, Department of Biology

We proposed a method for monitoring biodiversity of melliferous plants using commercial bee hives as a result of research during INTERREG project AmcPromoBID, which involved regions from Slovenia and Austria. The method was based on the comparison of field investigation of melliferous plants in the foraging area of the experimental hives and microscopical determination of botanical origin of pollen loads samples. High variability were observed between hives at the same location. This can be interpreted as a colony specific foraging strategy. Regardless of that, it is possible to correlate diversity of pollen loads with the diversity of melliferous plants. Higher diversity index of pollen loads corresponds to the higher number of melliferous plants species at the foraging site. Samples from the hive are not only indication of the diversity of actual collected food source but also an indication of diversity of melliferous plants. Bees are good samplers of the environment and can be employed for general monitoring of diversity changes at the studied location or to compare different locations at the same time. The method was partially adapted according to the C.S.I. Pollen workshop from the begging of this year and disseminated with the results of AmcPromoBID project. Results encourage us to join C.S.I. task force and to seek for future development of research methods for monitoring of biodiversity with honeybee colonies.

Comparison of apiculture and winter losses of honey bee colonies in Austria and Czech Republic

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Austria and the Czech Republic have historically shared beekeeping thanks to geographical as well as cultural proximity. Although the number of beekeepers (48132) and colonies (540705) is higher in the Czech Republic than in Austria (25207 beekeepers and 312740 colonies, respectively) the structure of apiculture is similar. For example, the average number of colonies per beekeeping operation is about 11 and 12 in Czech Republic and Austria, and the vast majority of beekeepers in both countries can be considered being hobbyists. The density of honey bee colonies in the Czech Republic (6.9 colonies / km²) is almost double than in Austria (3.7 colonies / km²). Winter loss data dating back to 2008 is only available for Austria. Supported by AKTION (project 69p11) cooperation between Austria and the Czech Republic started 2014 to compare apiculture of both countries and for the first time evaluate winter losses in the Czech Republic. The international standardized COLOSS questionnaire on winter losses of honey bee colonies was distributed and promoted through beekeeping journals, internet and at meetings between February and May 2014. For the internet survey, LimeSurvey 1.91 was used. In total, 562 Czech and 1021 Austrian beekeepers answered the questionnaire. Average operation size of participants in both countries was about 18 colonies. Winter loss rate was 12.5% (95% confidence interval: 11.4-13.6%) in Austria and 6.5% (95% CI: 5.5-7.6%) in the Czech Republic. Both datasets were submitted to the international coordinator of COLOSS to facilitate understanding of risk factors for honey bees. Further cooperation between Austria and the Czech Republic will focus on the renewal of livestock (i.e. the production of new queens and colonies) during summer 2014.

Removing heavy summer varroa infestations with queen caging and oxalic acid

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Uncontrolled varroa populations, pharmacoresistance and need of organic acaricides are chief problems in many countries. This is particularly true in warm regions, where extended broodright periods let the infestations grow considerably and highly effective control concepts must be applied.

The oxalic acid may be used best once dissolved in sucrose solutions and trickled onto the colony. However the treatments attain high efficacy only in broodless conditions. Indeed, quick pharmacokinetics and incapability to penetrate the brood seals at effective doses make the substance unsuitable to hit reproductive mites.

Full summer infestations must be tackled with efficient treatments. However sufficiently active miticides may not be unavailable. This fostered us to search a new organic method to control the summer infestations, when large amounts of brood are normally present. The queen is caged for 25 days within the colony to let all the brood hatch and the mites turn into phoretic. An oxalic acid treatment is made thereafter.

The trials started in 2007. Initially the effects of oxalic acid administered by trickling and of queen confinement were tested separately.

Subsequent comparative field tests were made in North and South Italy against controls. Api Bioxal, an oxalic acid based acaricide recently registered in Italy, was taken into consideration as well.

The tests repeatedly confirmed high miticidal activity, good tolerability for the individual honey bees and for the colonies, non-significant effects of caging on the queen survival.

In the different trials the efficacy averaged in the range 94-99%.

According to our results, this combination between a bio-technique and a treatment with a soft chemical may consistently reduce the severe varroa infestations often experienced in warm countries. To our knowledge, this is one of the most effective methods that can be used in an organic control concept against varroosis.

Introducing the Research Network for Sustainable Bee Breeding, taskforce within COLOSS

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The Research Network for Sustainable Bee Breeding was formed by a group of scientists who closely cooperated within the COLOSS working group “Diversity and vitality” during the COST funding period. The aim of the task force is to improve honey bee welfare by developing and disseminating comprehensive breeding strategies that include colony vitality and the conservation of locally adapted populations. The results of our work on genotype - environment interactions show that local bees survive longer than imported bees, most likely due to complex interactions with pathogens, phenological patterns and weather conditions (the complete results are published in a special issue of the Journal of Apicultural Research). The results support our aim to improve honey bee health by considering the bees’ genetic origin and how they interact with the surrounding environment. We believe that the native honey bee populations represent an important resource for breeding gentle and productive bees adapted to environmental challenges. An important focus area of RNSBB is the transfer of our findings to a popular form: we need to communicate the results of our research to beekeepers and to support them in revising their management accordingly. Recommendations to encourage breeding of well adapted bees must include simple methods on how to find and identify breeding material, methods for selecting bees for vitality and indications on conservation strategies. These recommendations can be disseminated by using multiple channels.

As well as being a task force within COLOSS we have set up a webpage www.beebreeding.net to reach out to scientists, technicians and beekeepers. Our working approach includes cooperation, exchange of ideas and mutual support, in a positive and friendly atmosphere. Our ambitious aim is to disseminate and implement sustainable breeding strategies on a global level, and to achieve this we will welcome new ideas and contributions that are in line with our constructive approach. The RNSBB currently includes the following 34 members from 17 countries: Sreten Andonov (Macedonia), Malgorzata Bienkowska (Poland), Maria Bouga (Greece), Ralph Büchler (Germany), Eliza Cauia (Romania), Leonidas Charistos (Greece), Cecilia Costa (Italy), Dariusz Gerula (Poland), Bjørn Dahle (Norway), Raffaele Dall’Olio (Italy), Janja Filipi (Croatia), Roy Mathew Francis (Denmark), Anna Gajda (Poland), Lionel Garnery (France), Pierre Giovenazzo (Canada), Ales Gregorc (Slovenia), Fani Hatjina (Greece), Evgeniya Ivanova (Bulgaria), Mateja Janes (Croatia), Nikola Kezic (Croatia), Hrisula Kiprijanovska (Macedonia), Per Kryger (Denmark), Marina Meixner (Germany), Nebojsa Nedic (Serbia), Beata Panasiuk (Poland), Hermann Pechhacker (Austria), Alice Pinto (Portugal), Sasa Prdjun (Croatia), Slađan Rašić (Serbia), Adrian Siceanu (Romania), Gabriele Soland (Switzerland), Adam Tofilski (Poland), Aleksandar Uzunov (Macedonia), Jerzy Wilde (Poland).

American and European Foulbrood diagnosis using debris of the hives and qPCR

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At the end of Winter 2014 the Apiculture Unit of Istituto Zooprofilattico Sperimentale del Lazio e Toscana (IZSLT) asked to 15 beekeepers to sample the debris of their stronger and weaker beehive in order to investigate in this matrix the presence of *Paenibacillus larvae* (pathogen responsible for American Foulbrood- AFB- of the honeybees) and *Melissococcus plutonius* (pathogen responsible for European Foulbrood – EFB- of the honeybees).

A total of 60 samples were analyzed: 30 for AFB and 30 for EFB.

The AFB investigations were performed by both cultural and PCR methods, according to OIE Manual of Standards, while the EFB investigations were performed by qPCR analysis.

While all of the samples resulted negative for AFB, 6 (of 30) samples resulted positive for EFB.

In 2 apiaries only the stronger family resulted positive for EFB (*Melissococcus plutonius* 1,72 x 10⁴ DNA copies and 1,65 x 10⁸ DNA copies), while in 2 other apiaries both the stronger and the weaker family resulted positive for EFB (*Melissococcus plutonius* 1,68 x10³ DNA copies in the stronger hive and 1,75 x 10⁷ copies in the weaker hive of one apiary; *Melissococcus plutonius* 1,72 x10⁷ DNA copies in the stronger hive and 1,7 x 10⁴ copies in the weaker hive of the other apiary).

The American and European Foulbrood investigation using the beehives debris with PCR methods could represent a useful tool for quick and preventive diagnosis of the diseases. Positive hives could be shook swarmed during spring time, avoiding the pathogens spreading within the apiary.

Temperature effects on *Deformed Wing Virus* (DWV) titers in adult honey bees

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The prevalence of viruses in honey bee colonies can present seasonal variations. However little is known about the effect of temperature on virus titers. Since some viruses can be affected by high temperatures, we tested whether increased rearing temperatures had an effect on virus titers in adult bees. Young bees naturally infected with *Deformed wing virus* (DWV) were reared *in vitro* at different temperatures ranging from 28°C to 37°C, with some groups being daily exposed to acute heat treatments. During 2 weeks, survival of bees was recorded, and DWV titers were assessed by quantitative RT-PCR. In parallel, as a field reference, we measured virus titer of young bees (of the same age than the experimental bees) reared in colonies. High temperatures significantly decreased DWV titers but increased mortality. Our results do not support the hypothesis that temperature could be used as a tool to eliminate viruses, but may contribute to reduce virus titers in further experimental studies. This suggests also that temperature changes could be involved in seasonal variations of virus titers.

Diffusion of *Vespa velutina* in Italy

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Vespa velutina Lepeletier 1836 was detected in France in 2005 in the area of Bordeaux and since its arrival it has spread in a great part of the country and in neighbouring countries. A monitoring activity for intercepting *V. velutina* has started since 2007 in the Italian regions of Piedmont and Liguria which border on France to the West. The main communication routes that join Italy and France, and freight traffic patterns were taken into account. Bottle traps baited with lager beer were used and the trapped insects were removed and determined weekly throughout the active season. The first *V. velutina* specimen, a male, was trapped at Loano (Savona Province, Liguria Region) on 19th November 2012. As a consequence of this catching, the monitoring activity continued in 2013, focusing on the Ligurian coast, and the local beekeeper associations were alarmed. These activities permitted to ascertain the presence of yellow-legged hornet workers in several localities in the provinces of Imperia (west Liguria) and Cuneo (southern Piedmont); active nests of *V. velutina* were also detected and destroyed in some of such localities. The further diffusion of *V. velutina* in Italy is presently monitored in a larger part of the country.

Findings in *Apis mellifera* L. (Hymenoptera: Apidae) mated queens with problems in their performance

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In the current context of environmental fragility, the quality of the mated queen bees is a key factor for the survival of honeybee colonies. It is well known the influence of genetics, nutrition and health on quality attributes. In recent years increasing problems have been reported in the quality of queens, related to supersedure queens prematurely, increased winter mortality of colonies or higher proportion of queen less colonies during the flowering season. In the present work sixty three queens during 2011 - 2013 with abnormal egg laying after mating were examined. The visual observation led to detect defects at macroscopic level and dissections of the queens gave information about internal organs: diameter, aspect of the spermatheca; symmetry and adhesions of the ovaries. Thirty queens were free of defects (N) at the macro and microscopic examination. In the remaining 33 queens defective queens (D) various defects were detected: Deformed Abdomen (DA) was the most frequent defect found (n=19) described as a slight indentation or external waist tergites of abdomen that not always was continued in the internal organs. Last tergite opened (LTO) (n=6) was registered concomitantly with defects in the spermatheca and ovaries; deflected abdomen (DA) (n=6) presented asymmetry in the abdomen and ovaries, gelatinous consistency, with adhesions and / or defective spermatheca . Presence of hematoma in queen abdomen (H)(n=2) corresponded to a queen with small spermatheca and a queen with defective ovaries This information is helpful for queen breeders to take decisions in the moment of caging mated queens, taking in account that defects at the abdominal level could cold explain 52 % of the problems in queen performance.

Management practices associated with acaricide failure during autumn treatment against *Varroa destructor* in honey bee colonies from Argentina

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Although the use of chemical acaricides could be associated with resistance, it is still the most preferred method for controlling *V. destructor* during autumn. Acaricides efficacy should be improved by identifying variables affecting treatment outcome. The aim of this study was to identify the risk factors associated with acaricide failure during autumn treatment of *V. destructor*. Mite infestation after treatment was evaluated in 62 apiaries and data regarding management practices were collected by means of a questionnaire. A mixed-effects model was constructed to associate management variables with the risk of acaricide failure occurrence. Colonies with high mites level prior to treatment and owned by beekeepers who did not frequently replaced queens were associated with a higher risk of acaricide failure ($P= 0.002$; $P= 0.001$). Other beekeeping practices indirectly improved acaricide efficacy. The influence of the geographical zone on risk factors such as queen replacement ($P= 0.001$) and percentage of *Varroa* prior to treatment ($P= 0.017$) might either indicate a direct effect of climate on mite fertility or a coordinated and regionalized response from Beekeepers concerning mites control measures. Percentage of *V. destructor* infestation prior to treatment and queen replacement are factors associated with acaricide failure occurrence in mite control during autumn. An integrated strategy for controlling mites that include chemotherapy and suitable beekeepers management is needed to keep mite populations low during winter.

Diversity of pollen sources: Preliminary data from CSI Pollen in Spain

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Coordination of CSI Pollen in Spain has achieved a relatively satisfactory answer, with several partners from different places and beekeepers with apiaries located in different habitat types, climatic zones and vegetation diversity. There are about 20 beekeepers whose answers, samples and shipping is regularly continues. Small differences are observed in foraging among the three colonies from each hive. Given the irregular weather we are experiencing this year in the Iberian Peninsula, some beekeepers have chosen, in unfavorable cases, the option to vary up to a week sampling dates. Pollen loads mainly collected and the protein content of these main taxa that has been obtained so far are: Cistaceae (15.07 ± 0.27), Compositae (18.57 ± 2.81), Brassicaceae (21.97 ± 2.93), Fagaceae (20.72 ± 2.75 , where the values corresponding to Castanea are 22.51 ± 0.34), Fabaceae (31.19 ± 0.16) y Rosaceae (24.42 ± 1.24), Ericaceae y Labiatae (still small number of samples analyzed). We note: 1. the variability in protein content, 2. diversity and availability of pollen sources throughout the year 3. foraging variability between colonies of the same apiary. We are waiting for with all this, determine the variability of the protein values given by the same types of pollen source located at different locations and also compared with that provided over different years. Important factors in climatology, both the current period and in consecutive periods are taken into account. Continuity of the project is expected, provided that the adequate funding is achieved.

Colony loss monitoring surveys in Scotland: winter loss rates and the online versus postal approach

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In Scotland we have carried out surveys of beekeepers since 2006, mainly by post. Since 2008 we have used geographically stratified random sampling of the membership records of the Scottish Beekeepers' Association, excluding those who opted out of surveys, to select the invited participants. In 2013 our survey was conducted online, as far as possible, using the LimeSurvey software (<http://www.limesurvey.org/>) for the first time. This was repeated in 2014.

In 2013, 300 participants were selected from 1094 possible participants, of whom 218 (73%) had email contact details. Postal questionnaires were used for the others, those whose email failed or where there were technical problems: 94 questionnaires were posted and 3 sent electronically for postal return.

Analysis of the 2012-2013 loss data gave 108 beekeeper respondents with valid loss data, of whom 56.5% experienced winter losses. The overall loss rate was 31.6% (158 colonies lost out of 500 colonies wintered). There were differing winter loss rates of 19.3% for the postal respondents and a much higher 36.6% for the online respondents. This difference is highly significant (Fisher's exact test gives a p-value of 0.0001297). Eleven (50%) of the 22 postal respondents and 50 (58.1%) of the 86 online respondents with valid loss data experienced losses, though this difference was not significant. The profile of the beekeepers differed between the two groups: the postal respondents were typically much more experienced beekeepers than the online respondents. It is therefore important for us to include the postal element for beekeepers not contactable by email, even though the response rate for the online survey was higher (65% online; 36% postal; 55% overall).

In the 2014 survey, 350 participants were chosen randomly from 1224 possible participants, 273 (78%) had email contact details, and 95 questionnaires were posted, allowing for 17 failing emails and one person with difficulty accessing the survey.

Preliminary results, based on 118 valid responses collected so far in LimeSurvey from beekeepers, are that 41 (34.7%) experienced losses over winter 2013-14, and that the loss rate was 13.2% (74 colonies lost from 560 wintered colonies). The final loss rates including postal responses may be expected to be lower.

The 2012-13 winter loss rate of 31.6% was the highest since our surveys began, the next highest being 30.9% over winter 2009-10. It seems likely that the much lower loss rate this last winter can largely be attributed to the different weather conditions.

Biodiversity survey and monitoring wildlife bees (Hymenoptera, Apoidea) in different ecosystems of the province of Córdoba.

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During the spring and summer of 2010 and 2011 carried out a study aiming to determine bee diversity, abundance and phenological curve, from both natural and anthropogenic ecosystems, in the province of Córdoba,

Following environments were selected, sorted by increasing degree of alteration of the original environmental characteristics: "Los Villares", "Pedroches", "IFAPA", "Campiña" and "Torre Malmuerta". In each of these monitoring stations a couple of traps like "Moericke" (or "pan-traps") were installed, one on storey and the other one metre above it, painted real yellow color.

The traps were installed in 2010 from 26 April to 12 July and, in 2011, from 2 May to 8 July.

The overall results (based on the first year of sampling, where data are more consistent) show that:

- The greatest diversity was obtained in the ecosystem, "Campiña" (Countryside), with 47 species,
- The lowest in the "Tower", with 17 species.
- The location with the highest number of catches was "Country, with a total of 3726 copies, but with a dominance of genera Panurgus and Eucera.
- The lower number of captures was the "Tower" with 84 copies.
- Regarding the height setting of traps, generally the most effective was the placed 1 m above ground level.
- The general phenology curves indicate that the main activity of Apoidea fauna in all ecosystems studied in Cordoba province extends from the last week of May to the third week of June, with maximum scores in the first two weeks of the latter month.
- As for the genera represented, was the predominant Eucera, which showed an excessive peak in the last week of May and early June in the ecosystem, "Campiña " (Countryside), followed by Panurgus, whose peak is focused on the second week of June in the same place.

Melliferous potential assessment of species: a useful tool to enhance the low diversified bee-food in agricultural habitats

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Agro-ecosystems are the major habitat of honey bee colonies in countries concerned by the colony losses. Floral resources in farming landscapes are not continuous and may result in periods of food shortage. Biodiversity in general, weeds and plants in particular, are declining in farmland habitats while several studies have shown their importance in bee health, e.g. through supplying food. The availability of nectar in the environment is not constant and depends on environmental conditions as well. The melliferous potential of flower taxa can be assessed by measuring the nectar secretion with the capillary method. Some species are known as excellent nectar producers while others are considered as secondary nectar providers. Crops can also become a major food source for bees, depending on agronomic interest expressed by farmers.

From a large database covering around 50 years of observations within the same region, which can be considered as a long-term study, we review studies concerning herbaceous species in agricultural habitats using the same method of capillary assessment. Based on the results on some species regarding the nectar production per flower and its concentration in sugar, we calculated the sugar production for each species. This survey aims at highlighting the variability encountered for some species and the relevance of this assessment potential for honey.

The introduction of some new species in crops is a valuable way to enhance biodiversity and nectar providing in a farming landscape. The sugar production is one of the best means to classify the melliferous value of species candidates. Our work may offer new perspectives to improve agricultural habitats for honey bees and some information necessary for the implementation of the best management practices of various habitats.

DNA bank establishment from *Apis mellifera* populations inhabiting the territory of Bulgaria

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Creating of a DNA bank with genetic material samples of Bulgarian *A. mellifera* populations is a science project stage with a real perspective for enrichment and development. Its goal is to have a centralized storage facility with DNA material and information, which can be used for future studies and research on a broad scale. The collected DNA could give and have information for the genetic history of the species, its populations and the complicated genetic relationships between and within them.

For the Bulgarian honey bees DNA bank establishment, up to this point have been used 1443 individual DNA samples. They inhabit 26 different populations on the country's territory.

The DNA bank will give new possibilities to study the genetic variability among the honey bee populations in the country, which is important for their selection and conservation. Furthermore, this gives possibilities for studying the phylogenetic relationship between Bulgarian honey bees and other races and ecotypes from different regions of Europa with the idea of characterizing and being clear about the subspecies status on the bred honey bees in Bulgaria.

The individual DNA samples are kept in 2 ml Cryovial tubes and each tube has a linear barcode with individual information about DNA quantity, the concrete bee individual and its origin, the population and the region.

Till the moment, honeybee DNA samples were used for microsatellite DNA analysis of nine microsatellite loci: Ac011; A024; A043; A088; Ap226; Ap238; Ap243; Ap249 and Ap256.

This idea could be successfully used by the scientific COLOSS network system for establishment of the honey bee DNA bank centers in Europe or for creating a honey bee DNA European network in order to facilitate future collaboration in genetic investigation to be done, which is important for conservation and selection of local honey bee populations.

Occurrence of abnormalities and diseases in newly mated and old honey bee queens

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Observation of nonacceptance, frequent replacements and reduced performance of honey bee queens (*Apis mellifera ligustica*), induced to analyse newly mated and old bee queens to detect the causes. The study was carried out on 99 newly mated bee queens, from queen breeders, and on 109 older bee queens or at the end of their career, which showed a poor efficiency and were obtained from beekeepers. The bee queens were dissected to check their reproductive system status and/or the presence of various problems. Anomalies and/or diseases were observed in 39.39% of newly mated bee queens and in 75.23% of older ones. Absence, hypoplasia or atrophy and melanosis of ovaries; absence of oviducts; egg anomalies; missing, abnormally shaped, double or discoloured spermathecae; absence of or abnormal sperms; and enteroliths; also in association with a change in tissues were common. *Nosema* spp. spores were detected in five bee queens and in most of the accompanying workers when present. Ciliate protozoa of the genus *Tetrahymena* and undetermined flagellates were present in the spermatheca of 22 newly mated and 2 older bee queens. The observed differences permitted to highlight the causes of the reduced performance of the bee queens and of the hives they came from. The information obtained would prove useful to improve queen breeding techniques.

Native-like coiled coil silk proteins from *Apis dorsata*

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Silk, a group of natural protein polymer produced from a specific gland in many insects. The most silk that received much attention are silk from silkworm and spider dragline. However, the large size and highly repetitive protein result in inefficient expression that limits the size of the silks produced. The silk from other insects that suitable for express in host has been considered. Our interest focuses on investigate silk associated biosynthesis genes in Giant honeybee (*Apis dorsata*). In this study, silk protein biosynthesis genes from native Giant honeybee (*A. dorsata*) in Northern Thailand were deduced the cDNA by using primers designed based on sequence of silk genes of *Apis mellifera*. The cDNA of silk genes in *A. dorsata* were found. The sequence encodes a amino acids , respectively. Proteins alignment demonstrated high homology to silk proteins of superfamily Apoidea (identity > 94%). The amino acids compositions of silk are highly in Ala that associated with the coiled coil regions in silk. Phylogenetic analysis suggested that all silk proteins from *A. dorsata* belong to honeybee silk with may slightly descended from their clade. This result described for the first time of basic information of silk genes in open-nesting giant honeybee and may lead to understanding the silk evolution in honeybee.

Wing venation teratology in European honey bees

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Honey bee (*Apis mellifera* L.) wing venation is quite distinctive and the resulting pattern is currently used in subspecies discrimination.

In recent years the forewings of worker bees sampled in *A. m. mellifera*, *A. m. ligustica*, *A. m. carnica*, and hybrid colonies were examined and various abnormalities - due to the presence of both supernumerary and defective veins - were observed in about 4% of wings. The supernumerary veins were considered as present only if an evident vein length could be detected, while slight thickenings were ignored. Most colonies did not show any teratology in wing venation, while a few ones provided several workers with abnormal wings; in these cases, some individuals showed two or more abnormalities.

Spurs of various length protruding from the standard veins were the most frequent abnormalities observed; among them an adventitious distal abscissa of the 2rs-m crossvein and the extension of the Rs vein from the marginal cell distal end were rather common. In some cases two opposed spurs tended to join or a single vein branched, thus defining an open or even closed supernumerary cell. In defective veins the missing stretch varied considerably in length so as in a few cases two contiguous cells merged more or less completely. The 1rs-m crossvein appeared rather unstable showing forks in some specimens and defective tracts in others.

Wing teratology should be taken into account when using wing venation patterns for the morphometric distinction of honey bee subspecies and data from abnormal wings should not be acquired, especially if automated procedures are used.

Monitoring Bee Losses in Spain

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In the program of COLOSS Monitoring, questionnaires were disseminated in Spain in 2014. The questionnaires were published in four important beekeeping journals and in the Centro Apícola Regional web site. They were also sent to professional associations and veterinarians and beekeepers attending the formative courses at the Center were asked for collaboration. Finally, J. A. Ruiz was in charge of the dissemination in Las Palmas and Tenerife Islands (Canary Islands).

Despite of this, a low level of participation was obtained. Only around one hundred of beekeepers collaborated in this survey. These beekeepers owned 9203 colonies. The results on the losses will be presented.

Piotr Medrzycki

I will present two contributions during the Apitox meeting

Developing A New HopGuard Strip for Improved Efficacy Against The Varroa Mite, *Varroa destructor*

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Varroa destructor is a serious parasite of the Western honey bee *Apis mellifera*. Synthetic miticides, primarily Apistan, Checkmite and Apivar are extensively used to control this parasite. However, Varroa has developed variable degrees of resistance to most applied synthetic miticides in various locations around the world. In order to provide more tools to effectively manage the resistance to applied miticides and effectively control Varroa mites, the efficacy of HopGuard made of hop extracts was evaluated against Varroa mites in Canada. Earlier trials using cardboard strips showed the efficacy of one or two applications of HopGuard (one strip per 5 frames of bees) was approximately 40% (Vandervalk M. Sc. 2013). The current study expanded upon earlier trials by changing the substrate of HopGuard strips to corrugated cardboard strips with double dose of active ingredients (25g/strip). Consequently, the exposure period of bees to the hop material increased.

HopGuard was tested in double brood chamber bee colonies in fall 2013 in Edmonton, Alberta Canada. HopGuard was applied at a dose of one strip for every 5 frames covered with bees. Apivar was used as a finishing treatment to determine the efficacy of HopGuard. The efficacy for the tested treatments was as follows; one application of HopGuard II, two applications at 10 day intervals, three applications at 10 day intervals, three applications of HopGuard at 5 day intervals and no treatment as a control was 80.0%±10.7%, 97.0±1.4, 98.5±0.7, 93.7±4.0 and 22.0±4.0, respectively. Similar results were achieved when HopGuard was tested in single brood chamber bee colonies in Prince Albert, Saskatchewan, Canada. The efficacy was 92.0%±5, 97.0±13.3, and 92.3±4.1 for HopGuard II with one application, HopGuard II with 2 applications and HopGuard with 3 applications, respectively.

A similar experiment is conducted in spring –summer 2014 in Edmonton, Alberta to test the efficacy of variable doses (1/2x dose to 2x dose) of HopGuard on Varroa mites and honey bee populations. Results will be discussed. Overall, the results of these studies show that the substrate of the strips to corrugated cardboard and increasing the applied dose improved the efficacy of HopGuard against Varroa mites. A single application of one strip of HopGuard II for every 5 frames covered with bees will have an efficacy 80.0-92.0% against varroa mites. Moreover, these results show that HopGuard II will become a useful effective miticide that will play a role in managing varroa in honey bees.

The Efficiency of Honeybee Quarantine in Preventing the Introduction of Honeybee's Pests and Diseases in Sudan

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Honeybees in Sudan are usually attacked by different indigenous pests. However, with the modernization of apiculture in the last decades, new species of pests were introduced to the country such as *Varroa destructor* mite, and the bacterial brood disease *Serratia marcescens*. Hence honeybee quarantine would be the first defense line to protect and conserve our native honeybees from the exotic pests and diseases. Therefore this study aims to answer the following question: 1) What are the pests and disease that subjected to honeybee quarantine in Sudan? 2) What kind of technique that used for examine the imported bees? 3) What is the efficiency of the used technique for detecting the introduced pest and diseases of honeybees?

The result of showed that there is no any one of the dangerous diseases (Bacterial, Virus, fungal and Microsporida diseases) recorded to be inspected by quarantine officers in Sudan. The parasitic mite *V. destructor* was found to be the only honeybee parasite that used to be inspected regularly by quarantine officers in Sudan using one technique called Tobacco smoke detection.

The inspection of packages bees contents which was developed in this study appeared to be the most efficient technique for detecting *V. destructor*. High infestation percentage 100% was able to be detected by this technique comparing to 00%, 08%, detecting by Tobacco smoke and Washing technique respectively.

Further confirmation of the efficiently of this new technique was done by inspecting the same imported honeybees in field. The infestation percentage in the established imported colonies in the field ranged from 1.11% among adult bees using Washing technique to 28, 25% using inspection of Brood. These results clearly demonstrated the weakness of the honeybee quarantine in the matter of the type of pests and diseases that subjected to quarantine and in the kind and inefficiency of the techniques that used for detecting these pests and diseases.

Interaction between *Varroa destructor* and viruses: implications on honeybee colony population

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Losses of honeybee *Apis mellifera* colonies have become a major threat to the apicultural industry. In temperate regions, such losses occur mainly during winter and pathogens are considered key factors. In particular, the mite *Varroa destructor* is considered one of the main candidates. This ectoparasite has a direct impact on the bee immune system and also serves as a vector for several harmful RNA viruses which is likely to enhance the deleterious action of the mites on honeybee colonies. Six viruses (DWV, ABPV, CBPV, IAPV, BQCV y SBV) have been detected in Argentina, primarily in asymptomatic honeybee colonies. The purpose of this study was to investigate viral and *Varroa* dynamics and its impact on honeybee population size over two annual cycles. The *Varroa* infestation level, the presence and viral load of six honeybee viruses and honeybee population size of 12 experimental colonies from Buenos Aires (Argentina) were periodically estimated. The relationship between parameters measured was analyzed. In total, four of the six virus analyzed were positive in the experimental colonies and multiple virus infections were detected in a same colony. DWV was the most prevalent virus and the percentages of positive samples for this virus were higher in colonies with elevated *Varroa* infestation levels. The percentage of varroa infestation varied in association with the DWV prevalence through the year, with the autumn samples showed the highest level of *Varroa* and viral infections. The DWV viral loads were significantly higher in colonies with severe *Varroa* mite infestations (above 4%) and the two parameters were significantly higher in colonies with lower honeybee population sizes. The same results were observed during two consecutive annual cycles. Our findings evidence great seasonal variability for viral and *Varroa* infestations and suggest that the interaction between DWV viral load and *Varroa* infestation are harmful to the overall health status and population size of honeybee colonies. The results emphasize the importance of maintaining low levels of *Varroa* mites in order to minimize the circulation of viruses within hives and therefore the impact on colony population size.

The Cage Assay about the Glutamine Effect on Lifespan of Honey Bees:CAGE

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Honey bees are considered as multifunctional organism for not only honey and other bee products but also pollination. There is positive correlation between their production and lifespan of honey bees. In this study, sterile 50 honey bees emerged from brood cells were put into the 4 experimental, 4 control cages. They were fed by 2 gr pollen cake and 5 mL1:1 w/v sugar syrup. It is well-known that Glutamine is a special amino acid support immune defence and effect the life span of some organism. So, Glutamine added the sugar syrup at different concentrations (0.001gr/mL and 0.01 gr/mL) and follow the bees in the experimental cages. The preliminary results showed that there is a difference between the two kinds of bees from the experiment and control cages. We have different measurements belongs to their life span, weight and length of the bodies.

The Effect of DEET on honeybee chemosensing by *Varroa destructor*

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Varroa destructor Anderson & Trueman (Acari: Varroidae) is an obligatory ectoparasitic mite of honey bee (*Apis mellifera* and *Apis cerana*) and is considered to be one of the major causes of European honey bee (*A. mellifera*) colony losses almost worldwide. In view of limited success in *Varroa* control, the use of synthetic repellent was evaluated. The major objective of the present study was to investigate the effect of common arthropod repellent N,N-Diethyl-m-toulamide (DEET) on the chemosensing and honeybee detection of the *Varroa* and its hosts, by electrophysiological and behavioural bioassays. The effect of DEET on the European honey bee (*Apis mellifera* L.) was also tested. In electrophysiological assays, the nurse head space served as a positive stimulus for the *Varroa* foreleg, whereas a queen head space was used as a positive stimulus for honey bee antennae. Two effects of DEET on chemosensing were evaluated: short term inhibition and long term inhibition. The inhibition observed in the presence of DEET simultaneously with a positive stimulus was termed "short term inhibition", while inhibition that occurred following the administration of the compound alone was termed "long term inhibition". In *Varroa*, DEET served as a long term inhibitor to the response of the chemosensory organ to nurse bee headspace volatiles, whereas in honey bee it caused short term inhibition of antenna response to queen volatiles. Consistent with electrophysiological studies, DEET significantly inhibited host choice of *Varroa*, whereas even a 10 times higher dose did not alter honey bee behaviours (e.g. antennating, grooming, fanning etc.) or worker attraction to a queen. This study shows a dramatic effect of DEET on *V. destructor* chemosensing and host location, preventing mites from reaching its host in the laboratory conditions. This finding indicates the potential of a compound with such an effect for *V. destructor* control. Still DEET effect on *V. destructor* population at colony remains to be proven along with its safety for the honey bee colony.

Optimization of a method of pollen counting adapted to honeybee diet

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Honeybee diet surveys need an accurate knowledge of the pollen supplies. Our studies concern the flora contribution in different landscape environments based on pollen loads collected in our experimental apiaries along the year, and require a large number of microscopic analyses. Knowledge of bee pollen diet focuses first on identifying the major resources that are related to land use, especially in cropping systems. Rare species are not necessarily sought in this process. Referring to the literature, we aimed in reducing the counted pollen grains in order to decrease the time spent for each sample. For this, we compared counts on whole transects with alternative methods of counting by sub-sampling, using the Louveaux palynological method (1978).

For each pellets sample, a drop from a homogenized suspension was dried, degreased and mounted onto slide in the glycerine gelatinized. The counting is performed through a numeric camera fitted on a microscope. The study had been carried out in two phases. Phase 1 : Test of slides homogeneity, carried out on a 10-slide set with one replicate. For each slide we compared an exhaustive count of each present taxa all along a whole transect with each quarters of it. Phase 2 : Search for an alternative counting. Work performed over the first 6 samples with replicates studied in previous phase retaining exactly the same transect but counting only a microscopic field on three, then four, and finally one on five.

Data analyses were carried out with Chi-square test on species > 5%. Phase1 showed a good homogeneity of the slides in particular for 2nd and 3rd quarters of the transect. In phase2, the comparison of alternatives counting to full transect verified the linearity of the counting results. Suitable results were given by the reading mode "one on three". We improved the accuracy on this method for the particular case of samples containing big pollen grain, e.g. *Zea mais*, and differently disseminated on the slide. This task consists in a general review of their number along the transect in order to establish the exact proportion.

In conclusion we propose a method in palynological analyses which is adapted to the predominant species visited by honeybees for their pollen diet.

Effectiveness of the varroa disease treatment with Beevital Hive Clean

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Because of the possibility of resistance to multiple administrated synthetic acaricides and very variable effectiveness of used treatments, there is a current need for utilization new treatment methods with natural phytopharmaceutical preparations and/or food additives. Also, the effects of residues and their by-products in honey and wax present environmental concerns and are another reason for reducing the use of conventional chemical mite- and parasite-control methods in beekeeping. The aim of this study was to examine the effectiveness of triple application BeeVital Hive Clean preparation in removing varroa mites from honey bee colonies during the active beekeeping season. This is a natural preparation containing oxalic and citric acids, extracts of propolis and different essential oil recipes. It represents an *eco-friendly* preparation in the form of food additive for honey bees.

The number of mites was recorded on three occasions in the pre-treatment period, and then recorded regularly once a day for 24 days after treatments began. The treatments were performed three times with six days apart using the “drench method” according the manufacturer’s instructions. The control group of honeybee colonies was treated with sugar powder.

Mite mortality after BeeVital Hive Clean application in the period from 16th of July to 7th of August, was higher ($P < 0.05$) compared to natural mite mortality in the pre-treatment period. Also, the total number of fallen mites during the experiment was significantly ($P > 0.05$) different between the treated and control groups. After the second treatment we determined 41.72%, and after third treatment 31.30% more fallen mites in comparison with first one. After last treatment there was 75% more of fallen mites in comparison with treatment before that. All treatments together achieved 91.6% efficiency of varroa mite control. Further experiments should be conducted in order to establish how to increase the efficacy of mite control in highly infested colonies during the period of brood rearing under continental climatic conditions.

Outcomes of the second year of investigation of winter honey bee colonies losses in Poland using COLOSS Questionnaire and randomised sampling

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In Poland winter honey bee colony losses have been a subject of research for 7 years. Since 2009 the COLOSS questionnaire has been used for this purpose. For many years the study was based on self selected sample, when beekeepers sent us questionnaires as a response to our request published in beekeeping journals. The questionnaires were also disseminated during beekeepers meetings and conferences. This way of survey resulted in a disproportion between beekeepers' participation in different regions. In 2013, for the first time we based our investigation mainly on stratified random sampling, where a region (voivodeship) was used as a stratifying factor. The response of beekeepers reached 33%, so we concluded that in the following years sending reminders to beekeepers would be necessary. At the beginning of April 2014 1552 questionnaires (together with envelopes with return address and stamps) were posted to beekeepers' addresses randomly selected from those from Veterinary Inspection lists. As an incentive we promised to send the Beekeeping Encyclopaedia (with authors' signatures) to 3 randomly selected respondents. By June 25th we received 459 filled in questionnaires and 112 return letters with questionnaires which, for various reasons, had not been delivered to the recipients. The incentive did not raise the response rate when we compared the results with those from the previous year. At the end of June we posted 112 letters to new randomly selected addresses and 981 reminders to nonresponders. The deadline for sending the questionnaires was set for 10th of July. The analysis of the data received by the 25th of June showed that the losses experienced by beekeepers were about 7.7% and were the lowest since the winter of 2006/2007. It was revealed that despite our efforts of many years to promote the use of organic acids for *Varroa* control, only 8.3% of beekeepers used them. Also using products based on essential oils was rare. Most beekeepers applied Polish products based on amitraz (74%). 30% of beekeepers claimed that they monitored the *Varroa* infestation level and 45% that they conducted drone brood removal as a supplemental method of *Varroa* control. In 15% of apiaries the symptoms, suggesting occurrence of bee poisoning were observed, usually in May. During the conference, the analysis of data obtained during the full period of conducting the investigation will be presented.

C.S.I. Pollen: Preliminary report of year one

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We will present first results of the Citizen Scientist Investigation (C.S.I.) on pollen diversity available to honey bees. After conducting pilot studies in 2013 and the presentation of this the project-idea at the COLOSS conference in Kyiv, a group has formed that coordinates this investigation in 21 European countries. The main task of these national coordinators is to recruit beekeepers to participate as "Citizen Scientists". We have developed a simple, standardized protocol that has been translated to 15 different languages and serves as instruction manual for beekeepers to collect, sample and analyze corbiculate pollen pellets. In each Apiary, the pollen collected by three honey bee colonies is harvested with pollen traps. Nine samplings between April and September have been scheduled in 2014. Due to the Citizen Scientist approach of this study, we limit the remote analysis by beekeepers to differentiation of discriminable different colors. To assess whether this is a possible factor for pollen diversity at this apiary location, further palynological analysis are needed to establish the relationship between the number of colors and number of different botanical origins in a specific sample; these are second tier analyses. Some countries already have national funding to collect and analyze the samples in the laboratory. A common protocol will be adopted for these analyses in different countries. The beekeepers in that case are trained also to store the pollen samples for the second tier analyses. In 2014, all over Europe more than 400 beekeepers participated in this study, helping in better understanding the pollen diversity in different areas, habitats (e.g. city or forest) and during the season. We welcome all countries to join for 2015 and make this the first and largest project investigating pollen diversity with the help of Citizen Scientists and their honey bee colonies throughout Europe.

Effect of deltamethrin on voltage-gated calcium channels of brain nerve cell in honeybee

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In order to protect bees to pollinate for crops and natural plants, it's necessary to research toxicity mechanism of harmful environmental substances on bees and establish a set of flexible and accurate risk assessment technology. We examined the effect of sub-lethal dosage of Deltamethrin(DM) on free calcium ion concentrate ($[Ca^{2+}]_i$) inside brain nerve cell of honeybee (*Apis mellifera ligustica* Spinola) developed in vitro with the help of Ion Ratio Imaging System.

The result shown as follows:

1. Even DM with low concentration(3.125×10^{-2} mg/L) shall rapidly and obviously impact $[Ca^{2+}]_i$ inside cell;
2. There are obvious differences among tests with different DM concentration:
 - (1) When DM concentration >0.25 mg/L, $[Ca^{2+}]_i$ shall maintain high level after the influence of DM was removed. It shows that high DM concentration cause death of cell due to overload of Ca^{2+} ;
 - (2) when DM concentration is <0.25 mg/L, $[Ca^{2+}]_i$ shall gradually recover without influence of DM. It shows that cell is still alive and has recovery function;
3. Further research were conducted by closing the L shape, T shape voltage-gated gating calcium channel and NMDA receptors of brain nerve cell, specifically and respectively. The results reveled that calcium ion concentration developed less sensitivity to deltamethrin when T shape voltage-gated gating calcium channel was closed, so we confirmed that the action site of deltamethrin was T voltage-gated calcium channel.

The study provides theory basis for honeybee protection of pesticide poisoning represented by deltamethrin strain for further research.

Poster Presentations

Registrant	Abstract title
Adjlane et al.	Population dynamics of <i>Varroa destructor</i> in colonies of <i>Apis mellifera intermissa</i> in Algeria
Aldea et al.	Interplay between climatic variables and <i>Varroa destructor</i> on the physiological performance of <i>Apis mellifera</i>
Blomstedt et al.	Mapping European Nectar Flows using MODIS Satellite data and Honey Bee Scale Hives
Božič et al.	Monitoring of melliferous plants biodiversity using pollen loads samples from commercial bee hives
Crailsheim et al.	Comparison of apiculture and winter losses of honey bee colonies in Austria and Czech Republic
Dalmon et al.	Temperature effects on <i>Deformed Wing Virus</i> (DWV) titers in adult honey bees
Formato et al.	American and European Foulbrood diagnosis using debris of the hives and qPCR
González-Porto et al.	Diversity of pollen sources: Preliminary data from CSI Pollen in Spain
Ivanova et al.	DNA bank establishment from <i>Apis mellifera</i> populations inhabiting the territory of Bulgaria
Lanzavecchia et al.	Interaction between <i>Varroa destructor</i> and viruses: implications on honeybee colony population
Laurino et al.	Occurrence of abnormalities and diseases in newly mated and old honey bee queens
Maitip et al.	Native-like coiled coil silk proteins from <i>Apis dorsata</i>
Manino et al.	Wing venation teratology in European honey bees
Odoux et al.	Optimization of a method of pollen counting adapted to honeybee diet
Odoux et al.	Melliferous potential assessment of species: a useful tool to enhance the low diversified bee-food in agricultural habitats
Palacio et al.	Findings in <i>Apis mellifera</i> L. (Hymenoptera: Apidae) mated queens with problems in their performance
Pardo-Martin et al.	Diversity of pollen sources: Preliminary data from CSI Pollen in Spain
Porporato et al.	Diffusion of <i>Vespa velutina</i> in Italy
Ruiz Martínez et al.	Biodiversity survey and monitoring wildlife bees (Hymenoptera, Apoidea) in different ecosystems of the province of Córdoba.
Topolska et al.	Outcomes of the second year of investigation of winter honey bee colonies losses in Poland using COLOSS Questionnaire and randomised sampling
Wang et al.	Effect of deltamethrin on voltage-gated calcium channels of brain nerve cell in honeybee

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