# SUMMARY REPORT

## International Honey Commission

## Melissopalynology Workshop

**10-11 April 2013, Thessaloniki, Greece**

The International Honey Commission with the cooperation of the Laboratory of Apiculture of Aristotle University of Thessaloniki organised a melissopalynological workshop in Thessaloniki on 10 and 11 April 2013. The main goals of the workshop were the addressing of melissopalynological problems regarding the botanical and geographical origin of honey and the harmonization of the melissopalynological methods and results among the analytical laboratories.

In total, thirty-five scientists from Austria, Belgium, Bulgaria, Croatia, France, Germany, Greece, Italy, Poland, Serbia, Switzerland and Turkey participated. The participants were welcomed by the president of the IHC Gudrun Beckh and the leader of the Melissopalinology Working Group Prof. Andreas Thrasyvoulou, who also reviewed briefly the main tasks of the workshop.

The workshop began with the presentation of the pollen frequently found in Italian honeys and the methodology that is applied for their analysis by Maria Lucia Piana. Afterwards, Paola Ferrazzi pointed out the main problems of the bee flora in Italy. Panagiota Gotsiou, Maria Dimou and Sofia Karabournioti mentioned the main honey types produced in Greece, while Dragan Bubalo talked about the botanical origin and characterization of Croatia honeys. Rudolf Moosbeckhofer and Helmut Heigl discussed the Austrian honeys and demonstrated the pollen data base PONET. Panagiota Gotsiou, Maria Dimou and Sofia Karabournioti summarized the data of the lists of the over-represented, under-represented and nectarless pollen grains frequently found in honeys, which was based on the lists that were previously sent by the participants. The presentation was followed by a discussion about the possibility of creating a pollen atlas of the main taxa found in honeys produced in Europe and other countries, in order to harmonise the results given among the analytical laboratories. Later, the first evaluation results about monofloral honeys having been artificially mixed with honeys with over-represented pollen were demonstrated by Chrysoula Tananaki, Maryanna Rodopoulou and Maria Dimou. Next, a discussion about the problems that might arise during the application of accreditation criteria to ISO/IEC 17025:2005 for melissopalynological analysis were coordinated by Maria Dimou, Chrysoula Tananaki and Andreas Thrasyvoulou. The first day of the workshop ended with a short tour at the beekeeping laboratory of the Aristotle University of Thessaloniki and an introduction walk to the city center.

The second day began with the presentation of the monofloral honeys in Switzerland by Katharina Bieri. A detailed presentation about the Turkish bee flora followed given by Asli Sunay, Tuğçe Dastan and Taylan Samanci. Dariusz Teper, Carine

Massaux and Patricia Beaune presented the botanical origin of honeys produced in Poland, Belgium and France respectively. After a short break, a sensorial analysis of honeys artificially or naturally mixed with overrepresented honeys was performed by the participants to combine the melissopalynological and organoleptical characteristics of the honeys. The workshop ended with an extensive discussion which began by brainstorming and grouping the main tasks raised in the workshop such as the the presence of over-represented pollen in honeys, the definition of the over-represented, under-represented and nectarless pollen grains, the nomenclature of the pollen types and the accreditation of the analytical laboratories.

In brief, the main conclusions and tasks raised from the workshop were:

a) The production of a standard list of the over-represented, under-represented and nectarless pollen grains frequently found in honeys, supported by references, in order to harmonise the results given from different laboratories.
b) The production and utilisation of a standard list among the analytical laboratories using pollen forms instead of families; genus and species classification was suggested in order to harmonise the results given from different laboratories.
c) The organization of an inter-laboratory test for pollen analysis in honeys among the participants' laboratories and other interested colleagues of the IHC was decided, given the lack in the market for efficiently standardized proficiency testing schemes (PTS) for quality assurance of the melissopalynological analysis.
d) The application of ISO/IEC 17005:2005 in melissopalynology and the use of an extended pollen database were discussed.
e) The creation of a mailing list of melissopalynology experts besides the ones participating in the workshop was suggested.

The panel agreed to continue the work through web communication and present the first results in the next meeting of the International Honey Commission in the Ukraine in September 2013. The project will be coordinated by Panagiota Gotsiou and Maria Dimou under the supervision of Andreas Thrasyvoulou.

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## "Open invitation"

The IHC Melissopalynology Group so far numbers over 40 members worldwide. The main scopes are to discuss melisopalynological issues and problems, and to develop cooperation among melissopalynological laboratories.

If you are working on melissopalynology and wish to join the IHC Melissopalynology Group, please contact:

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# ABSTRACTS

## BOTANICAL ORIGIN OF THE MAIN HONEY TYPES IN GREECE

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Greece is one of the most important honey producing countries in the EU with ~15.000 tonnes produced per year from ~20.000 beekeepers and ~1,5 million beehives. There are around twelve monofloral honey types with the main ones having the following botanical origin: pine honeydew (mainly from coccoid insect *Marchalina hellenica* on *Pinus brutia* and *P.halepensis*), fir honeydew (mainly from coccoid *Physokermes hemicryphus* on *Abies cephallonica* and *A.borisii*), thyme (mainly *Thymus capitatus*), chestnut (*Castanea sativa*), heather (*Erica* spp.), orange blossom (*Citrus* spp), sunflower (*Helianthus annuus*), cotton (*Gossypium hirsutum*), *Eucalyptus* spp. Significant amounts of multifloral honeys are also produced, as well as natural blends of nectar and honeydew honeys, mainly in Crete.
Problems with over-represented pollen, principally from *Castanea, Eucalyptus* and *Myrtus* are often observed in the case of thyme honey. This is primarily due (besides the primary pollution from nectars rich in pollen) to tertiary pollution taking place during honey extraction by centrifugation (from combs with stored pollen).

## BOTANICAL ORIGIN AND CHARACTERIZATION OF CROATIAN HONEYS

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Croatia has favourable conditions and long tradition in apiculture, particularly for honey production. In honey production process, besides the beekeeper and high quality of honey bee breed, an important factor for the preparation of the product for the market, is the evaluation of specific area's natural potentials. The term natural potentials primarily refers to floristic composition and climate conditions that greatly influence honey type variability. Geographical position of Croatia is specific and the territory spreads over three different regions: Pannonian, Mountain and Adriatic region. Each region has specific vegetation, relief and climate, continental, semi-highland/highland and mediterranean climate, respectively. Diversity of climatic zones and biocenological disposition represents a unique national resource that is rarely found in Europe. Biodiversity of a nectar flow plant species from distinctive climatic and geographical areas provides a great potential for the production of a common, but also a specific, rare and unique unifloral honey types. It is known that sage (*Salvia officinalis* L.) cannot be found on such large areas amongst the native flora in any of the Mediterannian countries, as in Croatian Adriatic region. The black locust (*Robinia pseudoacacia* L.), lime (*Tilia* spp.) and sweet chestnut (*Castanea sativa* Mill.) honey originating from Pannonian region, are the most represented on the Croatian market. According to the botanical origin, it is possible to produce in total 30 nectar honey types and 10 types of honeydew honey. Specifically, 14 nectar honey types can be produced in Pannonian region, 4 in Mountain region and 12 in Adriatic region. With respect to honeydew honey, it is possible to produce the highest number of different types (6) in the Pannonian

region, which is followed by 2 types of honeydew honey frequently occurring in the Mountain region, and 2 in the Adriatic region.

## THE RESULTS OF POLLEN ANALYSIS OF UNIFLORAL HONEYS EXAMINED IN THE BEE PRODUCT QUALITY TESTING LABORATORY IN PULAWY (POLAND) IN 2012

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The Bee Product Quality Testing Laboratory, established at the Institute of Horticulture, Apiculture Division in Pulawy, is the only laboratory in Poland, which performs service of pollen analysis of honeys and has been accredited for this method.
In 2012, 361 pollen analyses of nectar honeys, delivered by the beekeepers associations, companies engaged in the purchase and packaging of honeys, and by beekeepers and consumers were made. Among the delivered samples there was 204 samples declared as unifloral honeys, but, only for 93 samples it was confirmed by pollen analysis (Table). Declarations were limited only to the varieties mentioned in the Polish Standard - Honey Bee (PN-88/A-77626).
Comparison of the number unifloral honeys declared by beekeepers with the number of unifloral honeys confirmed by the pollen analysis

| Unifloral honey | Number of honeys declared as unifloral | Number of unifloral honeys confirmed by pollen analysis |
|---|---|---|
| *Brassica napus* | 47 | 41 |
| *Robinia pseudoacacia* | 41 | 11 |
| *Tilia* spp. | 63 | 22 |
| *Fagopyrum esculentum* | 39 | 7 |
| *Calluna vulgaris* | 14 | 12 |
| Total | 204 | 93 |

Besides honeys mentioned in the Polish Standard also were determined honeys: *Phacelia* (39 samples), *Centaurea cyanus* (10 samples), *Rubus* (4 samples), *Solidago* (4 samples), *Salix* (2 samples) and *Lythrum salicaria* (1 sample). Besides unifloral honeys characteristic for Polish climate zone also were determined honeys: *Helianthus* (3 samples from China, 1 sample from the south of Europe), *Echium* (3 samples from the south of Europe), *Salvia* (1 sample from the south of Europe). Moreover three multifloral honeys from southern Europe were found which contained high percent of *Castanea sativa* pollen and one which contained numerous pollen grains of *Sofora japonica*.
The discrepancies between the declaration of beekeepers, and the results of pollen analysis in 33 samples was probably due to overrepresented pollen which came from the bee bread. During microscopic examination of honeys, a high total number of pollen grains of plants flowering in the earlier time of the beekeeping season or even in the autumn of the previous year was found too.
In the case, when pollen from bee bread comes into the honey, pollen analysis cannot be reliable classification parameter of unifloral honeys.
The remaining honeys (78 samples) were incorrectly identified by the beekeepers.

## THE MONOFLORAL HONEYS IN SWITZERLAND, BOTANICAL ORIGIN, DISTRIBUTION AND POLLEN ANALYSES

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Most of the honeys in Switzerland are blended flower or forest honeys. The production of monofloral honeys is of minor importance. Many beekeepers keep the bees in houses that stand on fixed places. That is one of the reasons why the production of monofloral honey is difficult. Another reason can be found in the farming practices. In Switzerland you find a lot of small fields and pastures with a big variety of different plants side by side. But despite of this situation we do find some monofloral honeys: These are rape, robinia, rhododendron, dandelion as floral honeys, lime and chestnut as floral or honeydew honeys or mixtures of them and fir, spruce and deciduous trees honeys.
The geographical distribution of the plants which produce monofloral honeys is shown. Robinia, Chestnut and lime honeys are mainly produced in the southern part of Switzerland; lime honey can also be found in the Jura mountains. Dandelion honeys can be harvested in the central plateau, the prealps and the Jura mountains. Rhododendron honey is limited to the alpine region. Rape honey can be found north of the alps mainly in the central plateau. The main area of distribution of spruce today is in the prealps, the Alps and in the Western Jura mountains. Honey from the silver fir can be harvested mainly in the Western Jura, the central plateau and the Prealps. Deciduous trees appreciate the milder climate of the lowlands. We find them on the central plateau, the Jura and large parts of the Prealps and on the southern side of the Alps.

## HONEY PLANTS OF TURKEY

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Turkey has a rich range of flora with, approximately 10.000 different plant species. About 350 taxa of these plants produce nectar and are important for beekeeping. Honey bees usually collect nectar, pollen or both from plants to produce honey. This special relationship does not only produce bee products but also contribute to the environment by increasing pollination between many plants, enabling them to reproduce.
It is possible to determine the production region and botanical origin of honey by identification of pollens in honey. In this study, plant sources used for the production of honey in Turkey are described. A list is given with details like; flowering periods, location and how they are used by bees as nectar or pollen source. Results of field and microscopic studies on honey samples showed that 131 plant species belonging to 47 families are used by bees for production of honey in Turkey. Among them, 18 species belong to *Fabaceae*, 15 species belong to *Asteraceae*, 9 species to *Lamiaceae*, 7 species to *Brassicaceae,* 7 species to *Rosaceae*, 5 species to *Boraginaceae*, 5 species to *Ericaceae*, 4 species to *Apiaceae* and 61 species to others. Among those 131 species, 8 are only nectar source, 29 are only pollen source and 94 are both nectar and pollen source plants.

## MAIN PROBLEMS OF BEE FLORA IN ITALY

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The territory of the Italian peninsula extends in latitude with a wide variety of climates and environments from the Mediterranean Sea to the Alps. Among the species cited by Pignatti (1992) in the Italian flora, excluding Gymnosperms and other families without nectaries, we can count at least 887 genera and 4590 species that could be visited by the bees. The importance of bee plants is very high, considering that in Italy over 30 unifloral honeys are produced; anyway both the flora and several productions of the honey bees are at risk due to direct or indirect effects of human impacts. Global warming has an effect on flora and insects, still not very visible, but already documented. Plants species in the high mountains move upward, while the nival vegetation could be restricted hardly: this might result in extinctions; also in the Mediterranean region imbalances are seen. The vegetation equilibrium is continuously broken by the arrival of new pests, which weigh heavily on the flora not suited to cope with these new species. Recent exempla are the chestnut gall wasp *Dryocosmus kuriphilus* and *Glycaspis brimblecombei*, eucalyptus psyllid, which are heavily reducing the production of these important unifloral honeys. New plants become invasive, stealing land to indigenous species, as *Ailanthus glandulosa* and *Reynoutria japonica*, which, however, allow the production of honey.
One of the main causes of reduction of bee flora is the fragmentation of natural areas and the banalization of the landscape. The great extension of crops little or nothing useful to bees as corn and soybean, reduces the grazing of bees in large areas, especially in the Po valley; furthermore these crops are also potential sources of danger for the bees, because of the plant protection treatments to which they are subjected. The legume crops, which represent an excellent supply of nectar and pollen for *Apis mellifera* thanks to their abundant blooms, are less and less cultivated or they are not allowed to flowering because of the changes in cattle feed.
The decrease of resources adversely affects the health of bees; the beekeepers are forced to look for large displacements to reach zones safe for bees and escape to pesticides. The abandonment of vast hilly and mountain areas and the reduction of the meadows, increasing shrublands and forests, reduces the floristic biodiversity. Land management particularly affects the rarest species, which can characterize the geographic origin of honeys, losing more and more their identity. Among the typical Italian unifloral honeys those at most risk are the honeys of chestnut, eucalyptus, sainfoin, and sulla, while the honey of alfalfa and clover are now almost disappeared.

## MICROSCOPIC, ORGANOLEPTIC AND PHYSICOCHEMICAL CHARACTERISTICS OF BLENDS, OF HONEYS WITH OVER-REPRESENTED POLLEN

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In order to find how the characteristics of thyme honeys change when they are mixed with honeys with overrepresented pollen, we prepared mixtures of thyme honeys of three different levels (20%, 37%, 82%) with chestnut and eucalyptus honeys whose pollen is over-

represented. Pollen analysis along with electrical conductivity have been analyzed. The results show that electrical conductivity is less affected by the presence of over-represented honeys than the pollen grains' percentage. Furthermore, the samples will be analyzed for sugars, volatile compounds and organoleptic characteristics.

## ACCREDITATION CRITERIA TO ISO/IEC 17025: 2005 FOR MELISSOPALYNOLOGICAL ANALYSIS

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During the last years the number of laboratories applying for accreditation is rapidity increasing so that the laboratories can assure their quality but also to satisfy customers' demand. In this work, the application of ISO/IEC 17025: 2005 in melissopalynology is discussed. Special emphasis is given to the validation of the method and the external quality assessment. Further clarifications and guidelines for melissopalynological analysis regarding the technical requirements described in ISO/IEC 17025:2005 are essential to assure the quality of the results of pollen analysis in honeys.