Bee Hope project

1. A1 Introduction:
Like numerous natural species, European honeybee populations have faced increasing challenges imposed by three main groups of interacting drivers: (i) Environmental stressors (i.e. malnutrition, agrochemical molecules and residues, apicultural mismanagement, urbanization, climate change, habitat loss and fragmentation); (ii) Genetic diversity and vitality; (iii) Parasitism [1, 2] (see Fig. 1). Moreover, as the honeybee is a species of agronomical interest, its natural structure has been disturbed by deliberate introduction of foreign queens and swarms, which tend to homogenize the natural genetic diversity thereby endangering many native honeybee subspecies. The natural distribution of Apis mellifera subspecies and purebred subspecies kept by beekeepers and professional breeders have probably been influenced for many decades by (i) beekeeping techniques, (ii) international trade of honeybees (ex.: colonies, queens, drones), and also (iii) honeybee products used to counter declines in many countries [1, 3]. For instance, beekeeping has favoured the distribution of subspecies with specific apicultural traits outside of their natural range, usually to the disadvantage of other subspecies or honeybee species leading to a decline of genetic diversity required to face native or invasive pathogens introduced by commercial colonies [1, 4, 5]. Furthermore, the international and European trades of queens and of colonies have also spread the associated cortege of pathogen strains. One of the most documented example of pathogen spread, is the host shift of the invasive acarian species, Varroa jacobsoni, who was transferred from the sister species Apis cerana to Apis mellifera during the 80s. All these anthropic, biotic and abiotic factors, briefly summarized in the previous lines, are affecting the honeybee biology, and different combinations of these factors are probably involved in the important colony losses observed all over the world [1, 2]. While numerous studies have already been published to determine single (pesticides, varroa, microsporidia, virus) or multiple associated factors involved in honeybee decline, paradoxically no particular protocol, especially at ecosystem and subspecies distribution levels, has been developed to counter the effects of colony losses. Moreover, only a few publications have suggested genetic introgression as a putative supplementary cause of colony losses in Europe [6]. Beekeeping practices could favor the distribution of the preferred commercial subspecies outside of their native range. In that case the introduction of artificially maintained unadapted genes into the environment by beekeeping may directly contribute to the increase of mortality, and lead to losses of local genetic diversity through reductions in effective population size and through disruption of co-evolved gene complexes, as a consequence of mating with foreign unadapted subspecies [7]).

1. A2 Objectives, originality and novelty of the project
An interesting assumption is that current honeybee declines observed in European apiaries might be associated with trade of commercial honeybees by (i) the introduction of unadapted and artificially maintained colonies assumed to have interesting apicultural traits, and (ii) the spread of allochtone and invasive pathogens carried by allochtone honeybees [1, 3, 8]. Eco-ethological and genetical surveys have demonstrated that some populations of honeybee subspecies are adapted to local climate and flora [9,10, 11, 12]. Those populations are therefore particularly interesting to study and preserve in a context of sustainable beekeeping. The aim of our proposal is to set up, according to a North/South gradient, genetic conservatories of original naturally distributed honeybee populations. These honeybee preservation areas will have as missions: (i) to characterize the genetic and eco-ethologic diversity of honeybees from the Western European lineage (M lineage), (ii) to protect the genetic diversity of those populations, (iii) to constitute a reserve of diversity usable by the honeybee industry and beekeepers, (iv) to study the impact of the domesticated honeybee in the maintenance of local floristic diversity, and (v) to be able to use the honeybee as a bio-collector and as a biological indicator of environmental quality. Our proposal, BEEHOPE, thus comprises four Work Packages (WP) which join to form a unit
based on fundamental research on genetic and behavioral diversity of local honeybee populations and more applied aiming at answering a societal problem which is the preservation of a key species for the environment and human being by ensuring the pollination ecosystem service in agro- and natural ecosystems (see Fig. 1). BEEHOPE is structured to make an optimal use of the expertise of each European Partner.

(i) WP01 (impact study): The objectives of WP01 are: (i) to characterize the genetic diversity, level of introgression, (ii) to estimate the level of introgression, and (iii) to infer the genetic structure of 6 populations belonging to the two single subspecies of the western European lineage (*A. m. mellifera* and *A. m. iberiensis*). Thus, in WP01, we will employ molecular tools to perform an impact study and therefore determine whether each area is appropriate for setting up a preservation center. We will monitor many neutral markers (unselected markers) based on the protocol developed by Partner 1. Partner 1 has a recognized international expertise on this kind of survey.

(ii) WP02 (eco-ethological survey): The objective of WP02 is to develop a protocol for the preservation of genetic and adaptative traits of local populations and follow-up the evolution of the preservation centers in a context of global changes.

(iii) WP03 (spatio-temporal dynamics of pathogens): The objective of WP03 is to assess the diversity of the pathogen cortege associated with the geographical origin and history of the honeybee populations (spatial approach) and survey the evolution of the cortege during the project (temporal approach). We will monitor the spatio-temporal dynamics of key parasites involved in the arms race in each studied area (i.e. Varroa, virus, microsporidia, and bacteria) by using classical detection methods combined to ‘omics’ tools (ex.: PCR, qPCR).

(iv) WP04 (sustainable beekeeping & preservation centers): The main objective of WP04 is to develop efficient solutions (i.e. adapted honeybee ecotypes) based on results of WP01, WP02 and WP03 to ensure an efficient pollination of natural and agricultural plants, and potentially softening the pressure of climate change. Each European preservation honeybee center involved in BEEHOPE will have the duty of informing all beekeepers from their area about the advantages to breed honeybee ecotypes, and working with them to develop programs promoting the use of honeybee ecotypes in European apiaries to counter the honeybee decline, and reduce the risks of introduction of alien pests and pathogens. Finally, we will teach current beekeepers, and also to the next-generation of beekeepers via training courses offered by the agricultural high schools involved in this proposal, how to start and maintain a honeybee preservation center.

**Figure 1: Schematic overview of the “BEEHOPE” proposal.** Beige boxes represent the three main groups of drivers associated with honeybee decline; red bracket represent direct pressures on honeybee lineages from drivers; green arrows represent interactions between drivers; and blue arrows represent interactions within drivers. In mauve, BEEHOPE proposal with aim to counter honeybee decline by using the sustainable beekeeping as a key strategy.
BEEHOPE will bring pioneer data and key information for two themes of the BIODIVERSA call: (i) how manage and use the actual honeybee biodiversity to better support agro-ecosystems and agricultural production systems in terms of multifunctionality and outcomes in a global change context; (ii) knowledge and key data, thank to a dynamic interaction between different interlocutors working on honeybee decline (research centers, universities, preservation centers, beekeepers, beekeeping industry, and agricultural high schools), to help decision-makers in creating policies and governance systems promoting the emergence and support of agro-ecosystems and agricultural production systems benefiting from and beneficial to biodiversity and ecosystem services, here the pollination by honeybee.