# RISK FACTORS ANALYSIS FOR HONEYBEE COLONY MORTALITY IN NORTH WESTERN ITALY (PIEDMONT, LIGURIA AND VALLE D'AOSTA REGIONS) \*

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#### ABSTRACT

In recent years, large-scale losses of colonies and a high mortality in bees were reported in all countries.

In 2012 the French Agency for Food, Environmental and Occupational Health Safety (ANSES) published a pilot project with an active epidemiological surveillance protocol for honeybee colony mortality in 17 European member states, including Italy. Data for North Western Italy (NWI) were collected and analysed in order to study colonies mortality and apiary management. The colony mortality rate in NWI amounted to 5.8 (CI95% 3.8-7.8). Every region presented an over winter colony mortality rate: 6.2% in Liguria, 3.2% in Piedmont and 11.8% in Valle d'Aosta, respectively. The difference between regions was not statistically significant ( $\chi^2$ : 4.54, p = 0.10). At apiarian level, the highest mortality rate was observed in the spring, as 41.6% of observed apiaries showed at least one dead colony, while in the summer the apiaries with at least one dead colony were only 12.5%. The difference was statistically significant (McNemar test: 3.90 p = 0.0482). No statistically significant association was found between bee colony mortality rate and various factors considered in the questionnaire.

Keywords: Beekeeping, Mortality, Colony.

#### RESUME

Depuis plusieurs années, l'augmentation de la mortalité des abeilles est observée partout dans le monde.

En 2012, l'Agence nationale de sécurité sanitaire de l'alimentation, de l'environnement et du travail (Anses) a rédigé un projet pour comparer l'état de santé des ruchers dans 17 Pays de l'Union Européenne, dont l'Italie. Les données de l'Italie du Nord-Ouest (NWI) ont été recueillies dans une base de données et analysées pour évaluer l'association de la mortalité avec les activités de gestion des ruchers. Les Régions du NWI présentaient un taux de mortalité printanière moyenne respectivement de 6,2 % (Ligurie), 3,2 % (Piémont) et 11,8 % (Vallée d'Aoste). La différence n'était pas statistiquement significative (test du  $\chi^2$ : 4,54, p = 0,10). La mortalité d'ensemble du NWI a été de 5,8 % (CI95 % 3,8-7,8). Au niveau des ruchers, le taux de mortalité le plus élevé a été observé pendant la visite printanière : 41,6 % des ruchers observés ont montré au moins une colonie morte, tandis que pendant l'été, les ruchers avec au moins une colonie morte ont été de 12,5 %. Cette différence était statistiquement significative (test du 12,5 %. Cette différence était statistiquement significative (test de 12,5 %. Cette différence était statistiquement significative (test de nac Nemar = 3,90 p = 0,0482). Aucun facteur de risque présent dans le questionnaire et considéré dans cette étude, n'était statistiquement significatif.

Mots-clés : apiculture, mortalité, colonie.

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## I - INTRODUCTION

Beekeeping is a worldwide activity, important for production of honey and other apiary products, as pollen, wax, propolis and royal jelly. In the last years in Europe, beekeeping has become an increasing activity with a high proportion of non-professional beekeepers [Chauzat et al., 2013]. In Italy beekeeping is a practice of ancient tradition; the development of this activity is due to favourable climate conditions and to the suitable productivity of local breed *Apis mellifera ligustica* [Contessi, 2009]. There are approximately 72,000 beekeepers, of which about 10% are professional.

The average annual production of honey is approximately 9,000 - 12,000 tons [Mieliditalia, http://www.mieliditalia.it].

Bees play an important role in natural and agricultural ecosystems for the maintenance of environmental balance: their pollination of crops and spontaneous plants contribute to maintain ecosystems biodiversity. The Food and Agriculture Organization of the United Nations (FAO) estimates in fact that about 70% of the crop species that provide 90% of food worldwide, are pollinated by bees [European Commission, 2010; FAO, 2005].

Bee's population protection and management is essential, especially given global bee declines. In the last decade, declines in bee populations has been reported in many countries and is related to several factors [Formato, 2009; Porrini et al., 2008; Van Engelsdorp et al., 2008]. Environmental changes, the effects of enlarged use of pesticides and chemical products in agriculture, climate changes, and the development of pathologies due to bacteria (MELISSOCOCCUS PLUTONIUS AND **PAENIBACILLUS LARVAE** the respective CAUSATIVE AGENTS of European foulbrood and American foulbrood), viruses (chronic bee paralysis virus or CBPV, deformed wing virus or DWV, acute bee paralysis virus or ABPV) and invasive parasite species as varroosis (Varroa destructor), the Asiatic hornet (*Vespa velutina*), the small hive beetle (*Aethina tumida*) and nosemosis (*Nosema apis* and *Nosema cerane*). To stop bee's mortality, the knowledge about bee management and others factors related to bee mortality is required.

Since 2009 EFSA underlined the need of a harmonized surveillance system on bees at European level [Hendrikx et al., 2009]. To accomplish this task, in 2012 the French Agency for Food, Environmental and Occupational Health Safety (ANSES), designed a pilot surveillance project at European level [European Reference Laboratory on Honeybee Health, 2011]. The ANSES project is an active epidemiological surveillance protocol on honeybee colony mortality (EPILOBEE) based on randomly selected apiaries and colonies. 17 European member states, including Italy, took part at this project. A questionnaire was proposed to beekeepers. Many data on beekeeper (age, type and duration of activity, to be part of association groups), bees (breeds, type of production) farming practice (seasonal migrations, treatment, presence of crops, lands or wood, harbour or airport near hives) and clinical manifestations of the main disease (varroosis, American foulbrood, European foulbrood, nosemosis and chronic paralysis and the early detection of Aethina tumida and Tropilaelaps spp.) were collected in three different visits performed by local inspectors (autumn, spring and summer, during the beekeeping season). Mortality rates, management information and disease prevalence were calculated according to a standardized method in all member states and descriptive results were recorded into a web database [https://honeybees.anses.fr].

In this paper we present results on colony mortality observed in NWI (Piedmont, Liguria and Valle d'Aosta) during the period 2012-2013; the association between mortality rate and beekeeping management has been addressed.

# **II - MATERIALS AND METHODS**

## 1. STUDY AREA

Piedmont is in the northwest corner of Italy, bordered by Switzerland to the north and France on the west. The territory is of 25,402.46 squares Km divided in three zones: alpine, hill and land. Almost 40% of Piedmont is covered in mountains. Three High Mountain honeys from the Alpines are fir honeydew, rhododendron and wildflower. Wildflower mountain honey, a multi-floral honey collected from the flowering meadows and mountain pastures, at high altitude, can contain more than 30-35 of botanical species. Beekeepers is widespread: there are more than 5,000 apiaries.

Liguria is a coastal region of NWI, bordered by France to the west. The territory (5,421.55 squares kilometres) is mainly mountainous. Liguria's natural reserves cover 12% of the entire region. There are 1,100 apiaries. Valle d'Aosta is a small (3,263.24 square kilometres) mountainous region in northwestern Italy, bordered by France to the west, Switzerland to the north and the region of Piedmont to the south and east. There are 394 apiaries; beekeepers activity is mainly related in honey. It's produced from blooms in late spring-summer with the predominant use of the technique of moving the hives to higher altitudes (above 1,200 meters) for the mountain flowers.

#### 2. STUDY DESIGN

A random sample of 24 apiaries, 15 in Piedmont, 5 in Liguria and 4 in Valle d'Aosta were visited by trained bee inspectors from autumn 2012 to summer 2013 to monitoring colony mortality (figure 1). A proportional, stratified random sampling design was followed for apiaries. Stratification was carried out by health district according to spatial representativeness.

Figure 1 The map shows the sampled apiaries into the study area



Information on potential risk factors for bee colony mortality at apiary level were recorder through

EPILOBEE questionnaire, that was administered to beekeepers by trained bee inspectors at the time of

apiary visit. The following part of EPILOBEE questionnaire was considered for the statistical analysis:

- General data on the apiary: region (Piedmont, Liguria or Valle D'Aosta), size (number of colonies in the apiary), beekeepers age (< = 45, >45 years), beekeeping experience: (number of years in the activity), scale of beekeeping (hobbyist, professional beekeepers).
- Data on honeybees: (breed), production (honey, pollen, royal jelly, queens, swam); seasonal migration practice.
- Data on environment around apiaries (within a radius of 3 km): presence of farmlands, orchards, wild natural flora, forest/wood, towns, industries;
- Risk factors for exotic arthropod introduction: queen import, international harbour or airport at less of 10 km.

Data were extracted from ANSES web-based data base [https://honeybees.anses.fr/].

## 3. STATISTICAL ANALYSIS

Descriptive statistics were used to display bees dead distribution by questionnaire data. According to ANSES the mortality rate at colony level was calculated as weighted average of dead bees, by the apiary size. If at least one death colony was found in the apiary the apiary was classify "bees dead presence". The association between questionnaire data and apiary was estimated by Fisher exact test or Pearson chi square test of independence ( $\chi$ 2). The mortality rate seasonality in spring and in summer was compared by McNemar test. All data analysis was performed by SAS v9.2.

## **III - RESULTS**

A total of 38,228 bees belonging to 134 colonies were analysed in Piedmont, 15,296 bees from 68 colonies were sample in Liguria and 14,400 bees from 48 colonies were analysed in VDA. At apiary level 11 apiaries were found to have at least 1 dead colony (45.8%; Cl95% 25.6-67.2). Every region presented an over wintering mean colony mortality rate respectively of 6.2% (Cl95% 2.1 – 12.3) Liguria, 3.2% (Cl95% 1.6-6.1) Piedmont and 11.8% (Cl95% 6.6-19.6) Valle D'Aosta for apiary. The difference was not statistically significant ( $\chi^2$ : 4.54, p = 0.10).

The overall study area mortality rate was 5.8 (Cl95% 3.8-7.8).

On spring it was observed a greater number (41.6%) of apiaries with at least one colony dead than on summer (12.5%), with a statistically significant difference (McNemar test = 3.90 p = 0.0482).

Questionnaire data are shown on table 1, data analysis showed not statistical significant association.

# **IV – CONCLUSIONS**

The overwintering colony mortality rate found in NWI was 5.8% (CI95% 3.8-7.8), similar to that reported at Italian national level. According to EPILOBEE, colony mortality rate lower than 10% is considered acceptable.

Data on beekeeping management, as it is underlined on table 1, point out that the majority of

beekeepers is aged between 45 and 65 years old and practices this activity since more than 5 years.

Information on beekeepers could, that is still extensively exercised since many time thanks to the optimal environmental conditions and the high productivity of *Apis mellifera ligustica* that is the mainly breed farmed.

## Table 1

FACTOR	Category	No. of apiaries	Mortality yes (%)	p value
Bees breed	A. m. ligustica Other	9 15	33% 53%	0.23
Bees production	only honey others	20 4	50% 45%	0.4
Apiaries size	< = 96 colonies >96 colonies	17 7	42% 57%	0.27
Beekeepers age	30-45 45-65 Over 65	5 15 4	60% 40% 50%	0.73
Beekeepers activity	Hobby Part-time Professional	10 4 9	40% 50% 56%	0.79
beekeepers experiences	< = 5 years > 5 years	5 18	80% 39%	0.12
Beekeepers Organization member	yes no	4 17	25% 59%	0.31
Beekeepers training	yes no	8 14	63% 43%	0.23
Enviromental farmland	yes no	13 11	54% 36%	0.23
Enviromental orchards	yes no	10 14	43% 50%	0.3
Enviromental wood	yes no	20 4	45% 50%	0.4
Enviromental town	yes no	14 9	44% 43%	0.63
Enviromental industries	yes no	7 16	37% 57%	0.65
Airport	yes no	5 19	40% 47%	0.37
Harbour	yes no	3 21	33% 48%	0.43
Region	PMT LIG	15 5	47% 25%	0.57

#### Bivariate analysis on factors related to beekeeper questionnaire and honeybee mortality

Although the regions show different mortality rates according to climatically and territorial differences, the sample size (24 apiaries) was not able to account for statistical difference, as the power of the study is low related to sample size. For that reason analysis on factors implemented in questionnaire (apiary management, environment around colonies...) resulted not statistically significant for colonies losses.

Overwintering and seasonal mortality rates are statistically different: this result is consistent with data reported by EPILOBEE study where overwintering mortality rate ranged from 3.5 to 33.6 and seasonal mortality rate from 0.3 to 13.6 among European countries [Chauzat *et al.*, 2014].

Further analyses are required in order to take into account others risk factors including healthy colonies conditions and use of veterinary treatments as well to identify factors conditioning honeybee colony losses.

Despite the small sample size, the study was useful to gather interesting information on beekeeping in

NWI and on beekeeper profile: apiaries have lowmedium size (median 96 colonies) and are located mainly in wood; beekeepers are professional, with more than 5 experience years, but not specifically trained on beekeeping. The pilot project increased the relationship between beekeepers and public health veterinary, improving the collaboration among stakeholders.

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