# PRELIMINARY ANALYSIS OF A BEE POLLEN SAMPLE COLLECTED DURING AUTUMN IN THE CORNETU AREA (ILFOV COUNTY)

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#### Abstract

The present study describes the results of a pollen analysis carried out using light microscopy of a sample of bee pollen collected from a stationary apiary situated in the commune of Cornetu (Ilfov County) in October 2022, during a period with low temperature during the night, and after a summer characterized by very hot weather and severe drought conditions. To determine the preference of pollen sources by the honey bee colony, identification of melliferous plants in the apiary vicinity and up to the periphery of the commune of Cornetu was carried out. Cultured crops such as artichoke, buckwheat, cabbage, common vetch, mustard, phacelia, rape and several autumn-flowering garden plants were identified in the foraging area around the beehive. After sorting the pollen pellets based on colour, the results indicated several origins of pollen, including pollen of Brassicaceae, Asteraceae, Rosaceae, Portulace and also pollen of the highly allergenic anemophilous species ragweed - Ambrosia artemisiifolia, known as a popular source of pollen for bees.

Key words: autumn pollen; bee pollen; optical microscope; pollen morphology, stationary apiary.

## INTRODUCTION

The development of rural areas through bee colonies and pollination activities could ensure the balance of the agricultural ecosystem in order to conserve environmental resources.

Romania ranks second in the European Union, in terms of the number of beehives of bee families and first in the production of bee honey (European Commission, 2022; European Parliament, 2018). With a view to the rural development of the beekeeping sector in Romania, a particular importance is aimed at intensifying the potential of beekeeping determined by the pedoclimatic and melliferous base conditions in Romania.

The melliferous potential of Romania includes a series of plants from the spontaneous or cultivated flora, which through nectar and/or pollen secretions provide bees with the raw material necessary for the maintenance and development of bee families.

By flowering from February to March and until the end of October, melliferous plants from the S-W region of Ilfov County provide bee families with pollen and nectar throughout the active beekeeping season.

In addition to the melliferous sources mentioned above, bees also collect manna (honeydew) during certain periods of the active beekeeping season, this frequently occurs in the case of forests (deciduous and coniferous).

As a honey source is also considered the highly concentrated sugar secretion (50-80%) of flower buds and fruit bushes before the opening of flowers, with impact on the development of bee colonies (Iordache et al., 2008). The plants that bees search for nectar, manna or pollen form together the melliferous base (Beekeeping Law no. 383/2013 updated 2020 - Romania).

In an article, Spulber et al. (2020), pointed out the need for a study of the pollen types and their antioxidant properties for the whole geographical area of Romania.

The country's climate is mainly temperatecontinental with excessive nuance, with hot and dry summers and cold winters, dominated by the frequent presence of cold continental air masses from the East or Arctic air from the North and strong blizzard winds, the average annual amount of precipitation is 460-500 mm. The current research describes the results of a pollen analysis carried out using light microscopy of a sample of bee pollen collected from a stationary apiary situated in the commune of Cornetu (S-W of Ilfov County, Romania) in October 2022, during a period with low temperature during the night, and after a summer characterized by very hot weather and severe drought conditions (Figure 1), as part of a larger study that monitors both the melliferous power of the local crops and the amounts of honey and pollen obtained per hive.

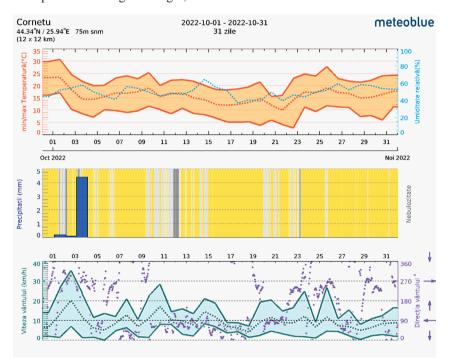


Figure 1. Meteorological records (temperature, precipitation and wind speed) in Cornetu, Ilfov County, Romania, October 2022 (Source: Romanian weather archive - retrieved from https://www.meteoblue.com/ro)

### MATERIALS AND METHODS

Buckwheat (*Fagopyrum esculentum* "Zita"), mustard (*Sinapis alba* "Maryna"), phacelia (*Phacelia tanacetifolia* "Stala"), common vetch (*Vicia sativa*) and camelina (*Camelina* sp.) crops were sown in July 2022, at the location of the apiary used in the current study, in the commune of Cornetu located in the southwest of Ilfov County, on the left bank of the Argeş river (where it forms the Mihăileşti reservoir, Giurgiu County) on Sabar riverside with an area of 17.12 km<sup>2</sup>.

Ilfov County is located in the south-southeast part of Romania, in the center of the Romanian Plain. Like a protective wall that surrounds a fortress, the county extends around the capital of Romania, Bucharest, being surrounded in turn by the neighboring counties of Prahova to the north, Dâmbovița to the west, Giurgiu to the southwest, Călărași to the southeast and Ialomița to the east (Figure 2).

In Ilfov, the agriculture occupies an important place, having great reserves and possibilities for development determined by the qualities of the soils, climate factors, technical and material endowments and the adequate labor force.

Approximately 95% of the agricultural potential of Ilfov county is held by arable land, the remaining 5% being occupied by trees, vineyards, meadows and hayfields (rasfoiesc.com, 2023).

Due to the geographical positioning of Ilfov county in the southern part of the country, there are relatively varied natural conditions, which are also reflected in a variety of soil types (Stănilă & Parichi, 2011).

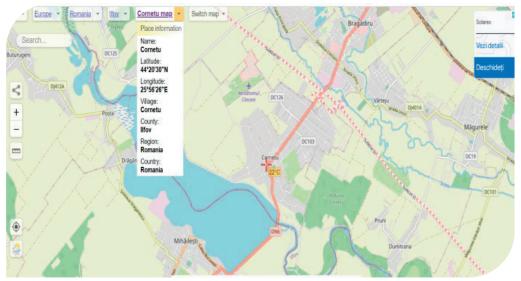


Figure 2. The location of Cornetu, S-W of Ilfov County, Romania (Source: Map retrieved from https://cornetu-if.pe-harta.ro/)

To determine the preference of pollen sources by the honey bee colony, identification of melliferous plants in the apiary vicinity and up to the periphery of the commune of Cornetu was carried out.

The apiary in the commune of Cornetu consists of 25 bee families, horizontal hives, with a capacity of up to 20 frames 1/1. This type of hive is very easy to maintain and use. The apiary is of a stationary type located on extraurban land with a large proportion of spontaneous vegetation.

Figure 3 illustrates the activities of mechanized processing by plowing and shredding the plots of land intended to be sown with buckwheat, mustard, phacelia, common vetch and camelina in spring 2022.



Figure 3. Ploughing at the location of the apiary used in the current study, in the location commune of Cornetu, Ilfov County, Romania (10.03.2022)

To obtain information on what flowers the bees are visiting at different points in the season,

beekeepers can use pollen traps on hives, and collect pollen samples at regular intervals

(Waters, 2014). In the present work pollen was collected in October 2022 (for the period September-October 2022) and was analysed at the Laboratory of Biology of the Faculty of Biotechnologies. from University of Agronomic Sciences and Veterinary Medicine of Bucharest, using the method and equipment described previously (Enache et al., 2019). In addition, a digital S-Eye 2.0 microscope camera was also used. For the present study, the pollen descriptions were compared to those found in the literature, including: Diethart & Heigl (2020); Halbritter (2016); Halbritter et al. (2020a: 2020b); Heigl (2020); Sam et al. (2020): ScientificBeekeeping (2023): Stebler (2023); Serbănescu-Jitariu et al. (1994); Tarnavschi et al. (1981; 1987; 1990).

## **RESULTS AND DISCUSSIONS**

Cultured crops such as artichoke (Cynara sp.), buckwheat (Fagopyrum esculentum "Zita"), cabbage (Brassica oleracea), common vetch (Vicia sativa). mustard (Sinapis alba "Maryna"), phacelia (Phacelia tanacetifolia "Stala"), rape (Brassica napus) and autumnflowering garden plants (dahlia - Dahlia sp., common zinnia - Zinnia elegans, pendant amaranth - Amaranthus sp.) as well as dog rose species and park rose varieties were blooming at the moment when the current study was carried out. Also, uncultivated areas colonized by allergenic plants were identified in the foraging area around the apiary.

After sorting the pollen pellets based on colour, the results indicated several origins of pollen, as follows: pollen pellet colour - yellow, pollen: tricolpate, prolate shape, with medium size (P = 35-40  $\mu$ m, E = 27.5  $\mu$ m), reticulate ornamentation, thick sporoderm (Figure 4), the images are similar to those of the pollen of white mustard var. Marvna (Gardenseedsmarket.com, 2023) collected in Cornetu and analysed in the laboratory previously (Figure 5). Pollen pellet colour - black/dark brown, pollen: tricolporate, oblate-spheroidal shape, with medium size (P = 40-45 um, E = 45-50 um), echinate ornamentation, the surface of echini has granules, possible ruderal Asteraceae (Figure 6).

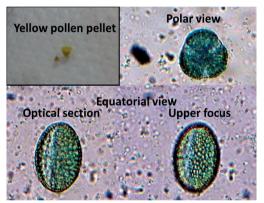


Figure 4. Yellow pollen pellet showing tricolpate pollen (Brassicaceae)



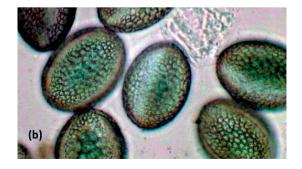


Figure 5. (a) Sinapis alba "Maryna" from the commune of Cornetu and (b) its pollen, 29.09.2022

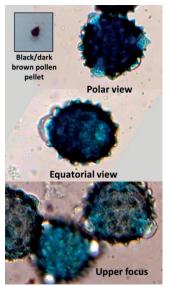


Figure 6. Tricolporate echinate pollen of medium size, possible ruderal Asteraceae

Pellet colour - orange, spores of Pucciniales (rust) fungi (fungal parasites that grow on some plants) (Figure 7) - they are sometimes collected by bees.

Pollen pellet colour - beige, pollen: tricolporate, suboblate shape, with medium size (P = 25  $\mu$ m, E = 30-32.5  $\mu$ m), psilate surface, possible Rosaceae pollen (Figure 8).

Pollen pellet colour - yellow, pollen: triaperturate, echinate, fenestrate, with medium size (37.5-40 µm), possible ruderal Asteraceae (Figure 9).

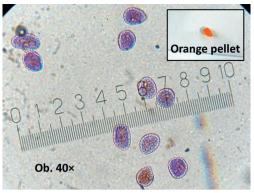


Figure 7. Rust fungi, fresh mount of pellet, toluidine blue stain

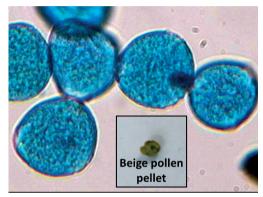


Figure 8. Tricolporate, suboblate, medium-size pollen, possible *Rosa* sp.

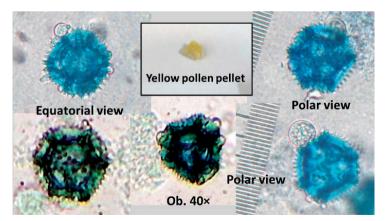


Figure 9. Triaperturate, echinate, fenestrate, medium-size pollen, possible ruderal Asteraceae

Pollen pellet colour - orange, pollen: pantocolpate, spheroidal, with large size (51- $62 \mu m$ ), echinate, with granular cytoplasm and pollenkitt (Figure 10), possible *Portulaca grandiflora* (Portulacaceae) that is traditionally found in villages all around Bucharest, since they are easy-care self-seeding plants, very pretty and drought resistant.

Pollen pellet colour - yellow, pollen: triaperturate, spheroidal, echinate, spines rather flat with a large base, with small size (13-16  $\mu$ m), no pollenkitt (Figure 11), possible pollen of the highly allergenic anemophilous species ragweed - *Ambrosia artemisiifolia* (Asteraceae), that is found in the apiary vicinity, and is known to be a popular source of pollen for bees.

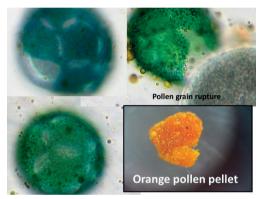


Figure 10. Pantocolpate, spheroidal, echinate, large-size pollen with granular cytoplasm, possible *Portulaca* grandiflora

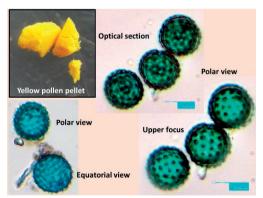


Figure 11. Triaperturate, spheroidal, echinate, small-size pollen with no pollenkitt, possible ragweed pollen

Pollen pellet colour - beige, pollen: periporate, pori with aperture membrane ornamented,

spheroidal shape, with small size (14-22  $\mu$ m) (Figure 12), possible pollen of amaranth (*Amaranthus* sp., Amaranthaceae), a cosmopolitan genus with ruderal species and species that are cultivated as leaf vegetables, pseudocereals, and ornamental plants. Similarly with *Ambrosia* pollen, the pollen of *Amaranthus* is highly allergenic.

Pollen pellet colour – orange-yellow, pollen: tricolporate with echinate surface with  $\sim 6 \mu m$ long echini, medium size spheroidal pollen (Figure 13), similar to sunflower pollen (*Helianthus annuus*), but found in other Asteraceae as well.

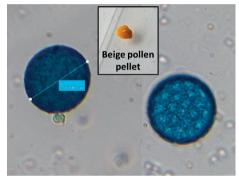


Figure 12. Periporate, spheroidal, small-size pollen, possible pollen of amaranth

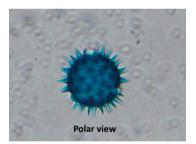


Figure 13. Tricolporate medium-size pollen with echinate surface, possible Asteraceae

### CONCLUSIONS

The aim of the current research was to determine the food resources available to the bee families of the apiary, in the climatic conditions of prolonged autumn manifested in Cornetu in the year 2022. Currently, great importance is given to beekeeping due to the special role that the bee has in preserving biodiversity, therefore special efforts are still required to protect this insect from the aggression of external factors such as pollution, climate changes and various disruptive human activities (such as unregulated phytosanitary treatments or herbicide excess).

The increasingly diverse aspects faced by beekeepers in the practice of this noble activity require complex and diversified information on the problems existing in the new socioeconomic conditions, but also regarding the environment.

The current results indicated several origins of pollen, including pollen of Brassicaceae, Asteraceae, Rosaceae, *Portulaca* sp. and also pollen of two highly allergenic anemophilous species: ragweed and amaranth.

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