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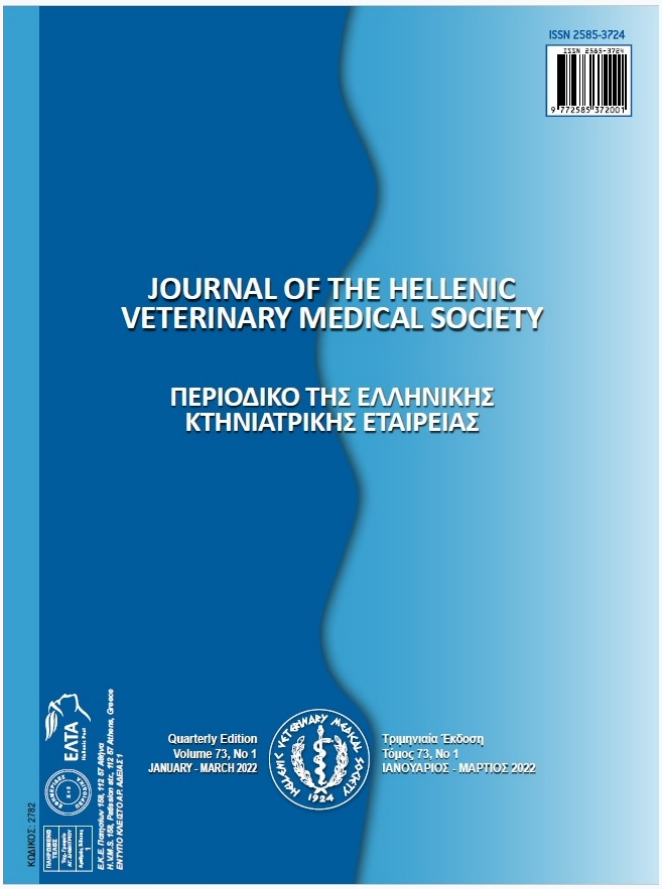

Comparing protein content of pollen and his impact on the lenght of life of honeybees

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Research article
Ερευνητικό άρθρο

Impact of the protein content in pollen on honey bees (*Apis mellifera* L.) lifespan

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ABSTRACT: The objective of the research work was to determine the protein content in identified pollen grains and the lifespan of worker honey bees fed with pollen with different protein content. In this study the number of the identified floral species was nine. The percentage of the total protein content in the bee-collected pollen pellets ranged from 11.5% for *Chondrilla juncea* to 25.1% for that of *Brassica napus*, and the average value was 18.5%. The average protein content of entomophilous plants was 18.5%, whereas it was 18.4% in anemophilous plants. Protein is one of the main components in pollen, and vary among different floral species source. The lifespan of worker bees ranged from 25 days when they consumed pollen from *Brassica napus* (25.1% protein content) to 14.3 days when they consumed pollen from *Zea mays* (17% protein content). The present study was performed to draw a comparison between the lifespan of worker bees fed with pollen with different protein content and significant differences were found. The worker honey bees lived longer when they consumed pollen with higher protein content. There is prolonged lifespan of bees fed with pollen from *Cirsium* sp., *Helianthus annuus* and *Chondrilla juncea*, as compared to bees fed with pollen from *Zea mays*, despite the higher protein content in the latter's pollen.

Keywords: pollen nutrition; protein needs; honey bees

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INTRODUCTION

Pollen satisfies the dietary needs of bees for protein (Herbert, 1992). Pollen differs in its nutritional value to bees (Vivino and Palmer, 1944; Standifer, 1967; Haydak, 1970; Crailsheim, 1990). Protein content in pollen ranges from 3.8% to 40.8% (Stanley and Linskens, 1974; Herbert and Shimanuki, 1978; Loper and Berdel, 1980). According to Kleinschmidt and Kondos (1977) protein content in pollen ranges from 7% to 37%, and for Roulston et al. (2000) ranged from 2.5% for *Cupressus arizonica* to 61.7% for *Dodecatheon clevalandii*. Liolios et al. (2015) showed that the protein content of pollen ranged from 12.8% for pollen from *Smilax* sp. to 30.1% for that of *Fallopia* sp., with an average of 20.8%. According to Kleinschmidt and Kondos (1976) protein content in pollen less than 20% cannot satisfy colony requirements for optimum brood rearing.

According to Avetisyan (1983) are 20 amino acids in pollen, but for honey bee's body 10 amino acids-threonine, valine, methionine, isoleucine, leucine, phenylalanine, histidine, lysine, arginine and tryptophan are essential of their diet (De Groot, 1953).

High protein content in pollen contains necessary amino acids in high concentrations (Wille et al., 1985). On turn most pollen types contain all necessary amino acids (Herbert, 1992). Individual amino acids are important in bee development (Herbert et al., 1970). There is a close relation between the nutritional value of pollen and the development, reproduction, and productivity of the bee colonies (Radev et al., 2014).

The honey bee (*Apis mellifera* L.) is using for aging research (Rueppell et al., 2004a). The lifespan of honey bees is influenced of different factors (Maurizio, 1961; Brucner, 1980; Winston and Katz, 1982; Milne, 1982, 1985). High level of protein content in pollen gives a long-lived bees, while low protein content minimises the longevity of bees (Kleinschmidt and Kondos, 1978; Schmidt et al., 1987). According to Levchenko (Stashenko, 1988), the level of protein content in pollen significantly affects the bee biology. Low protein content in pollen reduces the resistance of honey bees to diseases (Matilla and Ottis, 2006). Deficiency of quality protein in the diet of the bee colony can be one of the reasons for the emergence of invasion and infectious diseases (Bilash, 1990). One of the best ways to fight against dangerous diseases is by feeding the bees with pollen (Lavrehin and Pankova, 1983).

The purpose of this study was to determine the lifespan of worker honey bees according to consuming identified pollen with different protein content. In the literature there is not much information about pollen protein content of identified floral species and lifespan of honey bees.

MATERIALS AND METHODS

Pollen collecting

Pollen bottom traps (Figure 1) were placed at five bee hives and the pollen pellets harvested every second day from April until September in area of Belozem (Bulgaria) (N 42.201860/E 25.049330). The pollen loads from each hive were analyzed carefully. Four hundred and ten samples of bee-collected pollen pellets were separated over white paper according to their colour, shape, and texture (Figure 2).

Figure 1. Inside pollen trap

Figure 2. Separated pollen for identifing

Sampling

The labeled pollen samples were stored in separate vials in a freezer at -18 °C. The plant species represented by each pollen sample was identified through a microscopic examination of pollen grains (Figure 3). Palynological analysis was carried out using a similar methodology as Louveaux et al. (1978). To identify the pollen, is referred to the database of the laboratory of Apiculture-Sericulture of the Agricultural school of Aristotle University in Thessaloniki, Greece.

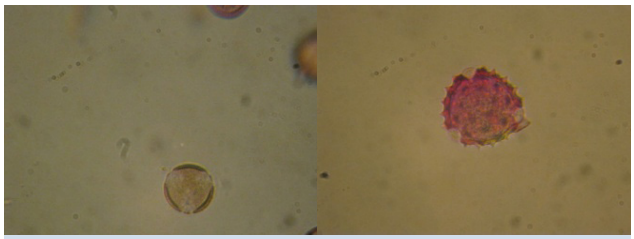

Figure 3. Microscopic pollen grains

Kjeldahl method

For nitrogen content determination, pollen was analyzed using the Kjeldahl method. The crude protein content was estimated using the factor 5.60 (Rabie et al., 1983) times the volume of HCL added to the pollen. In this case, the following formula applied: protein (%) = 0.791*V/m, where: V = the volume (mL) of HCl used for titration and m = the amount of pollen (g). Two replicates from each sample were analyzed, and the results were averaged.

Lifespan of worker honey bees

Nine groups with three cages 10*10*10 cm each were created (Figure 4). One hundred newly emerged worker honey bees were placed in each cage and fed with 50% sugar syrup (1:1 sugar/water). By pipet (Figure 5), a quantity of 20 ml sugar syrup containing two grams pollen from each identified floral species for each cage group were added. The mixed syrup was replaced every day with new one. All cages were placed under the same conditions, covered for darkness in a room at temperature 28 °C. In pre study fed only with sugar syrup, honey bees lived longer than 10 days at this temperature. The alive worker honey bees were checked every day and the last remaining found dead in each cage was reported for lifespan.

Figure 4. Cage with wax comb and honey bees

Figure 5. Pipet with sugar syrup and pollen

Statistical analysis

The results were statistically processed by used a Student's T-test and "Anova".

RESULTS

In the present study, identified pollen grains from nine floral species were used. They represent plants which are favored by honey bees. The largest number of taxa come from Asteraceae - 5, and 4 families by one representative (Figure 6). The results underline the fact that pollen from different plant species has a greatly varying protein content. The percentage of the total protein content in bee-collected pollen grains ranged from: 11.5% for *Chondrilla juncea* to 25.1% for *Brassica napus*, and the average value was 18.5%.

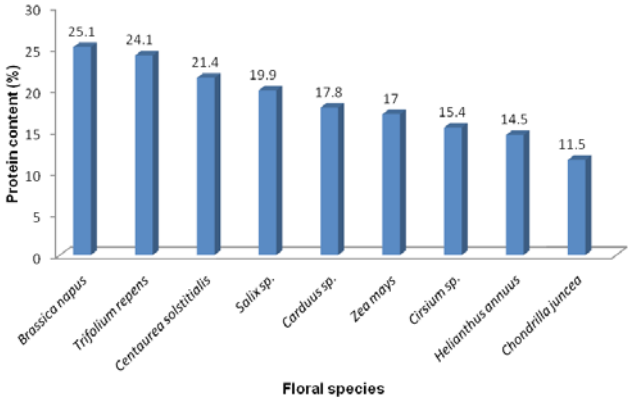

Figure 6. Protein content (dry matter %) of 9 different pollen species

Significant difference in the protein content in pollen of entomophilous plants - 18.5% compared to protein content in pollen of anemophilous plants - 18.4% was not found ($P < 0.05$, Student's t-test). Salix sp. was presented as an anemophilous plant in this study. The protein content values of Salix sp. and the other anemophilous plant *Zea mays* were pretty high. The result obtained for *Zea mays* could be attributed to hybrid variety.

Table1. Average lifespan (in days) of worker bees fed with sugar syrup and pollen of different plant species from Bulgaria

	Lifespan of bees I cage	Lifespan of bees II cage	Lifespan of bees III cage	Average lifespan of bees
Worker bees fed with sugar syrup and pollen from *Brassica napus*	26	24	25	25 a
Worker bees fed with sugar syrup and pollen from *Trifolim repens*	24	23	25	24 a
Worker bees fed with sugar syrup and pollen from *Centaurea solstitislis*	23	23	24	23.3 a
Worker bees fed with sugar syrup and pollen from *Salix* sp.	23	24	23	23.3 a
Worker bees fed with sugar syrup and pollen from *Carduus* sp.	23	24	22	23 a
Worker bees fed with sugar syrup and pollen from *Zea mays*	15	14	14	14.3 b
Worker bees fed with sugar syrup and pollen from *Cirsium* sp.	18	20	22	20 ac
Worker bees fed with sugar syrup and pollen from *Helianthus annuus*	17	19	16	17.3 c
Worker bees fed with sugar syrup and pollen from *Chondrilla juncea*	17	16	18	17 c

The data in Table 1 shows a slight reduction of the lifespan of worker bees when they consumed pollen with lower protein content. The longest lifespan was observed in the bees which consumed pollen from *Brassica napus* (25 days), while the shortest - in bees which consumed pollen from anemophilous plant species *Zea mays* (14.3 days). The present study was performed to make a comparison between the lifespan of worker bees fed with pollen from: *Brassica napus* (25.1% protein content), *Trifolim repens* (24.1% protein content), *Centaurea solstitialis* (21.4% protein content), *Salix* sp. (19.9% protein content), *Carduus* sp. (17.8% protein content), *Zea mays* (17% protein content), *Cirsium* sp. (15.4% protein content), *Helianthus annuus* (14.5% protein content) and *Chondrilla juncea* (11.5% protein content).

When comparing the results (Anova: Single factor), significant differences were found between the lifespan of worker bees fed with pollen from *Brassica napus* (25 days), *Trifolim repens* (24 days), *Centaurea solstitialis* (23.3 days), *Salix* sp. (23.3 days) and *Carduus* sp. (23 days), as compared to bees fed with pollen from *Zea mays* (14.3 days), *Helianthus annuus* (17.3 days) and *Chondrilla juncea* (17 days) ($F > F$ crit). Significant differences were also found between the lifespan of worker bees fed with pollen from *Brassica napus* (25 days) and *Trifolim repens* (24 days), compared to bees fed with pollen from *Cirsium* sp. (20 days) ($F > F$ crit).

The statistical analysis showed no significant differences ($F \leq F$ crit) in the lifespan of bees fed with pollen from *Cirsium* sp. (20 days) compared to bees fed with pollen of: 1) *Centaurea solstitialis* (23.3 days), *Salix* sp. (23.3 days) and *Carduus* sp. (23 days) ; 2) *Helianthus annuus* (17.3 days) and *Chondrilla juncea* (17 days).

It is noted, there was a prolonged lifespan for bees fed with pollen from *Cirsium* sp., *Helianthus annuus* and *Chondrilla juncea*, in comparison to bees fed with pollen from *Zea mays*, despite the higher protein content in the latter's pollen.

DISCUSSION

In this study, the results for pollen protein content come from some of worker bees' favorite plant species. Protein content in pollen depends on the plant visited by bees. The knowledge of pollen protein content of floral species in the area of beekeeping is the main aspect of utmost importance related to honey bees vitality and development.

The results presented in this study provided additional insight into the impact of protein content in pollen to honey bees lifespan. This would be useful information for beekeepers and would help them in bee colonies development. There are not many studies about the lifespan of bees fed with identified pollen with known protein content. The lifespan studies are

for bees which are a part of honey bee colonies.

According to: Taranov (Azimov, 1969), Tyunin (Azimov, 1969) and Azimov (1969) the lifespan of bees is about 30 days; El-Dib (Azimov, 1969) the lifespan from the Caucasian breed of bees is 27-29 days; Omholt and Amdam (2004) the life of bees varies from 3 to 4 weeks in the summer to over 6 months in the winter. According to Radev (2013) in a period of intensive rearing of brood the lifespan of honey bees is over 47 days on average.

It was found that the protein content in pollen of *Zea mays* is higher in comparison to that of *Cirsium* sp., *Helianthus annuus* and *Chondrilla juncea*, but the lifespan of the worker bees fed with that pollen is shorter than that of the bees fed with pollen from the other three floral species. This difference could be attributed to hybrid variety of the cultivated plant or other components in the pollen and needs further research. *Zea mays* is a cultivated plant and could be contaminated by a pesticide. Further research must be provided for components in pollen from *Zea mays* to establish why honey bees which consume pollen from it have the shortest lifespan.

According to Höcherl et al. (2012), extensive cultivation of *Zea mays* could force bees to collect mainly of it pollen instead of other kinds, and these honey bees fed maize pollen had shorter lifespan.

This research was a prerequisite for studying the impact of protein content in pollen on honey bees' lifespan. Analysis provided us with exclusive information about the nutrition of honey bees and could be used for further research. Every pollen type contains components such as: vitamins, minerals, enzymes, lipids, sugars and etc. in different amounts and bees are better off consuming mixed pollen rather than only one pollen type.

CONCLUSIONS

Protein is a main component of pollen and its content vary among different floral species. The lifespan of worker honey bees was longer when they consumed pollen with higher protein content.

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CONFLICT OF INTEREST

The author declares no conflict of interest.

REFERENCES

Avetisyan GA (1983) Breeding and keeping of bees. Moscow: Kolos.

Azimov TN (1969) The length of the lifespan of bees. Proc of the Res Inst of Api 13-20.

Bilash NG (1990) Influence the quality of pollen stocks of bees. Beekeeping 4:6-7.

Bruckner D (1980) Hoarding behavior and life span of inbred, non-inbred and hybrid honey bees. J of Api Res 19:35-41.

Crailsheim K (1990) The protein balance of the honey bee worker. Apidologie 21:417- 430.

De Groot AP (1953) Protein and amino acid requirements of the honeybee (*Apis mellifica* L.). Phys Comp et Oecol 3:197-285.

Haydak MH (1970) Honey bee nutrition. Ann Rev of Entom 15:143-156.

Herbert EWJr (1992) Honey bee nutrition. In: Graham, J.M., ed. The hive and the honeybee. Hamilton, IL: Dadant & Sons, : pp 197-233.

Herbert E, Shimanuki H (1978) Chemical composition and nutritive value of bee-collected and bee-stored pollen. Apidologie 9:33-40.

Herbert E, Bickley W, Shimanuki H (1970) The brood-rearing capacity of caged honey bees fed dandelion and mixed pollen diets. J of Econ Entom 63:215-218.

Höherl N, Siede R, Illies I, Gätschenberger H, Tautz J (2012) Evaluation of the nutritive value of maize for honey bees. J of Insect Physio 2:278-285.

Kleinschmidt G, Kondos A (1976) The influence of crude protein levels on colony production. Austral Beekeeper 78:36-39.

Kleinschmidt GJ, Kondos AK (1977) The influence of crude protein levels on colony performance. Austral Beekeeper 79:357-361.

Kleinschmidt G, Kondos A (1978) The effect of dietary protein on colony performance. Austral Beekeeper 79:251-257.

Lavrehin FA, Pankova SV (1983) Biology of the honey bee. Moscow: Kolos.

Liolios V, Tananaki C, Dimou M, Kanelis D, Goras G, Karazafiris E, Thrasyvoulou A (2015). Ranking pollen from bee plants according to their protein contribution to honey bees. J of Api Res 54:582-592.

Loper GM, Berdel RL (1980) The effects of nine pollen diets on brood rearing of honey bees. Apidologie 11:351-359.

Louveaux J, Maurizio A, Vorwohl G (1978) Methods of melissopalynology. Bee World 59:139-157.

Matilla H, Otis G (2006) Effects of pollen availability and *Nosema* infection during the spring on division of labour and survival of worker honey bees (Hymenoptera: Apidae). Env Entom 35:708-717.

Maurizio A (1961) Lebensdauer und Altern bei Honigbiene *Apis mellifera.* Gerontologia 5:110-128.

Milne CPJr (1982) Early death of newly emerged honeybee workers in laboratory test cages. J of Api Res 21:107-110.

Milne CPJr (1985) An estimate of the heritability of worker longevity or lenght of life in the honeybee. J of Api Res 24:140-143.

Omholt S, Amdam G (2004) Epigenic regulation of aging in honeybee workers. Sci Aging Knowledge Environ 26:28.

Rabie A, Wells J, Dent L (1983) The nitrogen content of pollen protein. J of Api Res 22:119-123.

Radev ZR (2013) Length of life of *Apis mellifera* worker bees in region of Belozem (Bulgaria). J of anim sci 6:103-105.

Radev Z, Liolios V, Tananaki C, Thrasyvoulou A (2014) The Impact of the Nutritive Value of Pollen on the Development, Reproduction and Productivity of Honey Bee (*Apis mellifera* L.). Bul J of Agri Sci 20:685-689.

Roulston TH, Cane JH, Buchmann SL (2000) What governs protein content of pollen: pollinator preferences, pollen-pistil interactions or phylogeny? Ecol Mono 70:617-643.

Rueppell O, Amdam G, Page R, Carey J (2004a) From genes to society: Social insects as models for research on aging. Sci Aging Knowledge Environ 5:5.

Schmidt JO, Thoenes SC, Levin MD (1987) Survival of honey bees, *Apis mellifera* (Hymenoptera: Apidae), fed various pollen sources. Ann of the Entom Soc of America 80:176-183.

Standifer LN (1967) A comparison of the protein quality of pollens for growth stimulation of the hypopharyngeal glands and longevity of honey bees, *Apis mellifera* L. (Hymenoptera: Apidae). Insectes Sociaux 14:415-425.

Stanley R, Linskens H (1974) Amino acids and proteins in pollen, biology, biochemistry, management. Berlin, Heidelberg, New York: Springer-Verlag.

Stashenko VI (1988) Pollen collecting by honey bees. Beekeeping 9:10-11.

Vivino AE, Palmer LS (1944) The chemical composition and nutritional value of pollens collected by bees. Arch of Bioch and Biophys 4:129-36.

Wille H, Wille M, Kilchenmann V, Imdorf A, Bühlmann G (1985) Pollenernte und Massenwechsel von drei *Apis mellifera*-Völkern auf demselben Bienenstand in zwei aufeinanderfolgenden Jahren. Revue Suisse De Zool 92:897-914.

Winston ML, Katz SJ (1982) Foraging differences between cross-fostered honeybee workers (*Apis mellifera*) of European and Africanized Races. Behav Ecol and Sociobi 10:125-129.