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Research Article

Floral Choices, Parasites and Micro-organisms in Natural Populations of Bumblebees (Apidae: Hymenoptera) in Ankara Province

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Abstract: In this study, the flower choices of different bumblebee species in Ankara province regarding their associations among various parasites and micro-organisms were determined. A total of 21 different species of the genera *Bombus* Latreille and *Psithyrus* Lepeletier were examined. The motile hypopus of the mite species *Acarus farris* (Oudemans, 1905) (Acaridae) was found to be phoretic on *Bombus* (*Bombus*) terrestris, *Megabombus* (*Megabombus*) *argillaceus*, *Megabombus* (*Thoracobombus*) *zonatus* and *Pyrobombus* (*Sibiricobombus*) *niveatus* queens. *Nosema bombi* was also determined to be an internal parasite of *B. terrestris*. Finally, a nematode species belonging to the family Allantonematidae (Tylenchida) was isolated from *Megabombus* (*Thoracobombus*) *sylvarum citrinofasciatus*. The flower choices of the discussed *Bombus* and *Psithyrus* species were examined by using diversity and richness indices. It was observed that *Anchusa leptophylla*, *Onopordum anatolicum*, *Echium italicum*, *Trifolium pratense*, *Galega officinalis* and *Astragalus* sp. were the most preferred plant species of bumblebees in Ankara province.

Key Words: Bombus, Psithyrus, Flower choice, Acarus farris, Nosema bombi

Ankara'daki Bombus Arısı Doğal Populasyonlarının (Apidae: Hymenoptera) Çiçek Tercihleri ile Bunlarda Saptanan Parazit ve Mikro-organizmalar

Özet: Bu çalışmada Ankara'da saptanan farklı bombus arısı türlerinin çiçek tercihleri ile değişik parazit ve mikro-organizmalar ile etkileşimleri birlikte incelenmiştir. *Acarus farris* (Oudemans, 1905) (Acaridae) türü akarın hareketli hipopeal formunun *Bombus* Latreille ve *Psithyrus* Lepeletier cinslerine ait 21 türden *Bombus* (*Bombus*) terrestris, *Megabombus* (*Megabombus*) *argillaceus*, *Megabombus* (*Thoracobombus*) *zonatus* ve *Pyrobombus* (*Sibiricobombus*) *niveatus* kraliçelerinde foretik olduğu tespit edilmiştir. *Nosema bombi*'de *B. terrestris* türünde internal parazit olarak gözlenmiştir. Ayrıca Allantonematidae (Tylenchida) familyasından bir nematod türü de *Megabombus* (*Thoracobombus*) *sylvarum citrinofasciatus* kraliçelerinden izole edilmiştir. İncelenen *Bombus* ve *Psithyrus* türlerinin çiçek tercihleri de çeşitlilik ve tür zenginliği indeksleri kullanılarak ortaya konmaya çalışılmıştır. *Anchusa leptophylla*, *Onopordum anatolicum*, *Echium italicum*, *Trifolium pratense*, *Galega officinalis* ve *Astragalus* sp. türlerinin, Ankara bölgesinde bombus arıları tarafından en çok tercih edilen bitki türleri olduğu saptanmıştır.

Anahtar Sözcükler: Bombus, Psithyrus, Çiçek tercihi, Acarus farris, Nosema bombi

Introduction

Bumblebees (Hymenoptera: Apidae) live in annual colonies that are founded by single, overwintered queens (1). They are primitive-eusocial bees (2) in which the queen raises the first batch of workers on her own, but restricts herself to egg-laying and other nest activities after the colony starts to grow in worker numbers (3). Bumblebees are economically and ecologically important pollinators of many temperate plant species (4,5). More than 300 species have been recorded worldwide (5), of which 48 have been found in Turkey (6). Extensive commercial rearing of bumblebee colonies, especially in New Zealand, Belgium, the Netherlands and the United States, has naturally affected Turkey. In 1998, four companies started to import queens and colonies from abroad and so far the tendency to use bumblebees has increased in eco-agriculture, mainly for the pollination of greenhouse crops, especially tomatoes, strawberries and aubergines.

Bumblebees require pollen and nectar to support the colony (7). The flower choice strategy of bumblebees is to begin their foraging careers by visiting a number of different plant species and subsequently to concentrate on the most rewarding plants. Selecting the most remunerative flower species from the many available in a habitat is a major problem confronting foragers (8). The scent of the flower, energy economics, proboscis and corolla tube length with some behavioural aspects like

buzzing are the major factors in the mutual interactions of bees and plants (9). The aim of this study was to have an idea about the importance of different bumblebee species on the pollination of various plants in terms of the most preferred flowers. It is known that parasitized bumblebees prefer different flowers than unparasitized ones do (4). To date, according to the literature found, little data is available on flower preference, abundance and ecological effects of parasites and micro-organisms in natural populations of bumblebees in Turkey. In this study, we report the results of field surveys in Ankara. The province is generally steppe-like (ca. 900 m) and surrounded by high mountains (ca. 1800 m) covered mainly by *Pinus nigra* Arn. ssp. *pallasiana* (Lamb) Holmboe, different species of *Quercus* sp., *Picea* sp. and *Juniperus* sp. with *Cedrus libani* A. Richard (10). This kind of geographical structure seems to be very favourable for bumblebees of high and low altitutes and for forest and steppe species.

Materials and Methods

A total of 1547 specimens from 21 different species of the genera *Bombus* Latreille and *Psithyrus* Lepeletier were collected from Ankara province, which is located in the central Anatolian region of Turkey, between 1996 and 1998. Bumblebees of the following species were encountered in the study area: *Pyrobombus* (*Sibiricobombus*) *niveatus* Kriechbaumer, 1870, *P.* (S.) *vorticosus* Gersteacker, 1872, *P.* (Kallo*bombus*) *soroeensis* (Fabricius, 1793), *Megabombus* (*Megabombus*) *hortorum* (L., 1761), *M.* (M.) *argillaceus* (Scopoli, 1763), *M.* (*Melanobombus*) *lapidarius* (L., 1761), *M.* (*Rhodobombus*) *mesomelas* Gersteacker, 1869, *M.* (R.) *armeniacus* Radoszkowski, 1877, M. (R.) *pomorum* (Panzer, 1805), *M.* (*Thoracobombus*) *zonatus* Smith, 1854, *M.* (T.) *pascuorum* (Scopoli, 1763), *M.* (T.) *sylvarum citrinofasciatus* Vogt, 1909, *M.* (T.) *mlokosiewitzi* Radoszkowski, 1877, *M.* (T.) *humilis* Illiger, 1806, *M.* (T.) ruderarius (Müller, 1776), *M.* (*Subterraneobombus*) *fragrans* (Pallas, 1771), *M.* (*Laesobombus*) *laesus* Morawitz, 1875, *Bombus* (*Bombus*) *terrestris* (L., 1758), *B.* (B.) *lucorum* (L., 1761), *Psithyrus* (*Metapsithyrus*) *campestris* (Panzer, 1801) and *P.* (*Allopsithyrus*) *maxillosus* Kluger, 1817. The present nomenclature follows that used by Rasmont and Flagothier (6).

The bumblebees were all caught on plants while they were searching for nectar or pollen. The plants were also collected and the altitude, humidity, time and temperature were noted (5). In the laboratory, only the queen bees were checked, freeze-killed, dissected and examined for the presence of various internal and external parasites and micro-organisms (1,4,11). Five species out of 21 were found to be infested by different agents. These were *B. terrestris*, *M. sylvarum citrinofasciatus*, *M. argillaceus, M. zonatus* and *P. niveatus*.

A total of 37 different plant species were collected during the study period. All the flowers observed visited by bumblebees were brought to the laboratory in numbered glass vials. These were all checked for the presence of questioned agents. The Shannon diversity (H) (6) and species richness (d) (12) indices were used in order to find out the most preferred plant species in Ankara province by the given formulas,

$H = - \Sigma P_i \log_2 P_i$ where $P_i = N_i / N$

N_i = Number of specimens of a bumblebee species visits the i plant

N= Total number of specimens of the examined bumblebee species

$d = (S - 1) / \log N$

S = Number of species

N = Number of individuals

Results

Parasites and micro-organisms

In this study, various parasites and micro-organisms were determined to be associated with different bumblebee species in Ankara (Table 1).

The motile hypopus stage of the mite species *Acarus farris* (Oudemans, 1905) (Acaridae) was found on *B. terrestris*, *M. argillaceus*, *M. zonatus* and P. *niveatus* queens. They were located on the meso and metanotum (interalar band and scutellum) of the thorax both in the dorsal and lateral parts, at the base of the wings and also on the first segment of the abdomen attached side by side. In *M. zonatus*, the average individual number observed on one queen is 90.36 ± 13.99 where on *M. argillaceus* 45.89 ± 11.56, on *B. terrestris* 23.84 ± 8.12 and on P. *niveatus* 31.00 ± 9.34 (Mean ± St.dv.). *Acarus*

Table 1. Parasites and micro-organisms associated with natural populations of *Bombus* species (queens only) in Ankara. The infestation percentages were given in paranthesis.

Bombus species (queens)	n	*Acarus farris*	Allantonematidae	*Nosema bombi*
P. (*Sibiricobombus*) *niveatus*	43	(13.95%)	-	-
M. (*Megabombus*) *argillaceus*	39	(15.39%)	-	-
B. (*Bombus*) terrestris	67	(20.90%)	-	(74.63%)
M. (*Thoracobombus*) *zonatus*	30	(56.67%)	-	-
M. (*Thoracobombus*) *sylvarum citrinofasciatus*	7	-	(57.14%)	-

farris is mainly known as a field species, but it has also been recorded from barley, hay, cheese and poultry food in spite of the fact that the hypopus may be found clinging to larger mites and to insects such as scatopsid flies (13). It was also found in the nests of *Cinclus cinclus aquaticus* Bech. (14). *A. farris* sometimes may be confused with *A. siro*, which was recorded from bumblebees before (1), but the hypopus differs in the position of a sucker and the adjacent genital seta (13). During the hypopeal stage, because the mites do not feed and their mouthparts are considerably reduced, they are generally thought to be phoretic than external parasitic for the *Bombus* species (15,16).

In this study, the only Protozoon observed that affects bumblebee queens was *Nosema bombi* (Table 1). It was found in *B. terrestris* queens and develops primarily in malphigian tubules, then extends secondarily to the midgut, tracheal matrix, connective tissue and eventually the fat body (17).

Sphaerularia bombi is the only known parasitic nematode species in bumblebees and it has been found in 22 European, 15 American *Bombus* and 7 *Psithyrus* species (1,4,17). In spite of this, in Ankara province, this species has not been observed but another nematode species has been isolated from 4 specimens of *M. sylvarum citrinofasciatus*. The nemotode is from the family Allantonematidae (Tylenchida). Unfortunately, these specimens could only be determined to family level. It is known that some species of this family are known to be used in the biological control of some insect species, such as tobacco thrips *Frankliniella fusca* (Thysanoptera) (18).

Flower choices

During the study period, 1547 specimens from 21 different species were collected from 37 plant species (Table 2). By using the Shannon index it was observed that *M. argillaceus*, *M.* armeniacus, *M. zonatus* and P. *niveatus* do not choose particular plant species but *B.*

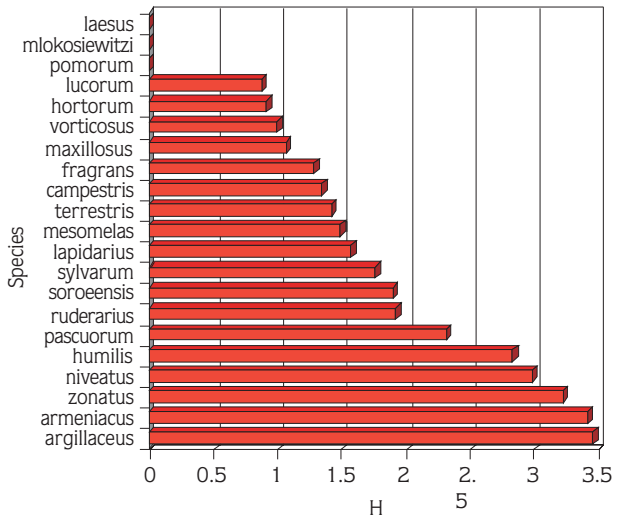

Figure 1. The diversity of the bumblebee species according to the Shannon diversity index.

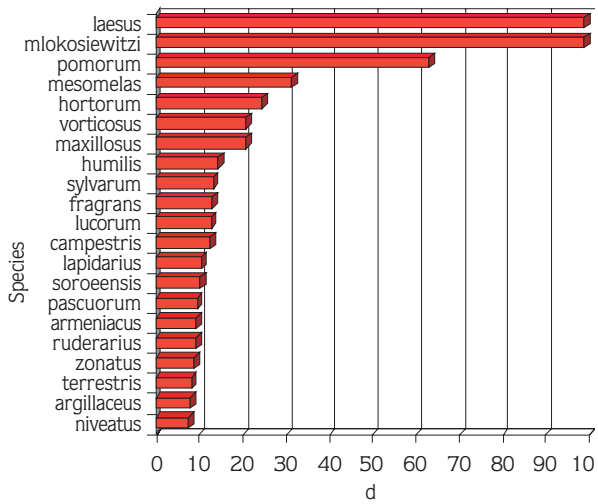

Figure 2. The richness of the bumblebee species according to the species richness index.

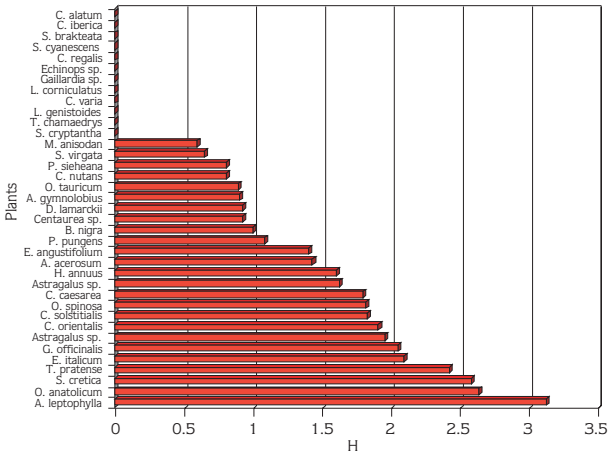

Figure 3. The diversity of the plant species preferred by bumblebees according to the Shannon diversity index.

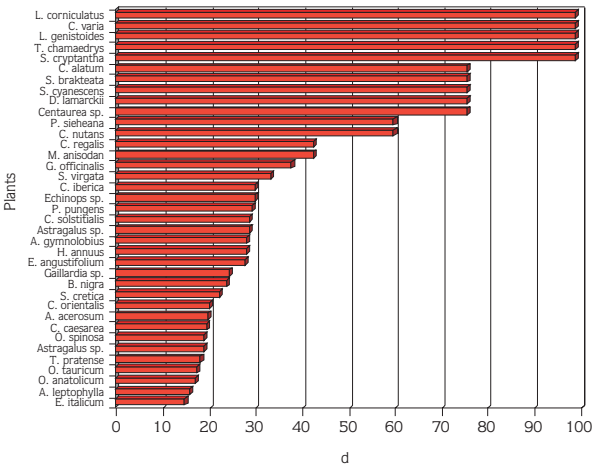

Figure 4. The diversity of the plant species preferred by bumblebees according to the species richness index.

Table 2. The flower choices of bumblebees. H refers to the Shannon diversity index and d refers to the species richness index.

	Stachys cretica L.	*Carduus nutans L.*	*Anchusa leptophylla Roemer&Schl.*	*Phlomis sieheana Rech.fil.*	*Centaurea sp.*	*Echium italicum L.*	*Salvia cryptantha Montbret & Aucher.*	*Astragalus sp.*	*Acanthollimon acerosum (Willd) Boiss.*	*Teucrium chamaedrys L.*	*Onobrychis spinosa L.*	*Lotononis genistoides (Fenzl) Benth.*	*Phlomis pungens Willd.*	*Onopordum tauricum Willd.*	*Onopordum anatolicum (Boiss.) Eig.*	*Centaurea solstitialis L.*	*Convolvulus varia L.*	*Digitalis lamarckii Ivan.*	*Trifolium pratense L.*	*Galega officinalis L.*
P. (Sibiricobombus) silvestris	2		26					8	47		47			105	1					
M. (Megabombus) argillaceus	6		58	3	2	58	1	2	4		5		12	1	5	3		1	1	1
B. (Bombus) terrestris	5	3				143														4
M. (Thoracobombus) sonanus			29			26		1	3		15	1	1	1	2	10				
M. (Thoracobombus) ruderarius	12										1				46		1		34	
M. (Rhodobombus) armeniacus	6	1	3	1	1	6		7		1	10		4	4	8	2				
M. (Thoracobombus) pascuorum			25			26									18				19	1
P. (Kallobombus) soroeensis			29												15				15	
M. (Melanobombus) lapidarius			24												21				18	
Pat. (Metapsithyrus) campestris			19			6														
B. (Bombus) lucorum	9																			
M. (Subterraneobombus) fragrans						1			9						18	1				
M. (Thoracobombus) sylvarum citrinofasciatus	2		12			5								8						
M. (Thoracobombus) humilis			3			5					2					2			4	2
P. (Sibiricobombus) variegatus									4					4						
Pat. (Allopsithyrus) maxillosus			6																	1
M. (Megabombus) hortorum																		2	4	
M. (Rhodobombus) mesomelas			2																	
M. (Rhodobombus) pomorum																			2	
M. (Lasiobombus) lassus						1														
M. (Thoracobombus) mlokosiewitzi											1									
TOTAL	42	4	236	4	3	277	1	18	67	1	81	1	17	123	134	18	1	3	97	9
H	2.59	0.81	3.13	0.81	0.92	2.1	0	1.63	1.43	0	1.82	0	1.08	0.89	2.64	1.83	0	0.92	2.43	2.05
d	22.18	59.79	15.7	59.79	75.45	14.74		28.68	19.71	-	18.86	-	29.26	17.23	16.92	28.68	-	75.45	18.12	37.73

Table 2. The flower choices of bumblebees. H refers to the Shannon diversity index and d refers to the species richness index.

	Echium angustifolium Miller	*Lotus corniculatus L.*	*Galium sp.*	*Echinops sp.*	*Cousinia cariensa Boiss. & Bull.*	*Cousinia orientalis (Dry) Schrad.*	*Helianthus annuus L.*	*Astragalus gymnolobius Fischer*	*Astragalus sp.*	*Cousinia regalis S. F. Gray*	*Bellevia nigra L.*	*Salvia virgata Jacq.*	*Salvia cyanescens Boiss. & Bal.*	*Salvia bracteata Banks. & Sol.*	*Centaurea iberica Trev. & Spreng.*	*Cirsium alatum (Gmelin) Bobrov*	*Marrubium anisodon C. Koch*	Total	H	d
P. (Sibiricobombus) niveatus					24	25	11	13	28									337	2.99	7.52
M. (Megabombus) argillaceus					27	20	2		8	7	17	2	3	3				252	3.46	7.91
B. (Bombus) terrestris	1		30	16					1									202	1.42	8.24
M. (Thoracobombus) zonatus					22	6	4		1						16	3		141	3.23	8.84
M. (Thoracobombus) ruderarius											16							110	1.92	9.31
M. (Rhodobombus) armeniacus					2	11			24			10					6	107	3.42	9.36
M. (Thoracobombus) pascuorum	6	1																96	2.32	9.58
P. (Kallobombus) soroensis	12																	71	1.9	10.26
M. (Melanobombus) lapidarius																		63	1.58	10.56
Psi. (Metapsithyrus) campestris								6										31	1.35	12.74
B. (Bombus) lucorum									21									30	0.88	12.86
M. (Subterraneobombus) fragrans																		29	1.29	12.99
M. (Thoracobombus) sylvarum citrinofasciatus																		27	1.77	13.27
M. (Thoracobombus) humilis					1		2											21	2.84	14.37
P. (Sibiricobombus) verticosus																		8	1	21.04
Psi. (Allopsithyrus) maxillosus	1																	8	1.07	21.04
M. (Megabombus) hortorum																		6	0.92	24.42
M. (Rhodobombus) mesomelas						1											1	4	1.5	31.56
M. (Rhodobombus) pomorum																		2	0	63.12
M. (Laesobombus) laesus																		1	0	-
M. (Thoracobombus) mlokosiewitzi																		1	0	-
TOTAL	20	1	30	16	76	63	19	19	82	7	33	12	3	3	16	3	7	1547		
H	1.4	0	0	0	1.8	1.91	1.61	0.9	1.96	0	1	0.65	0	0	0	0	0.59			
d	27.67	-	24.37	29.9	19.4	20.01	28.15	28.15	18.81	42.6	23.71	33.36	75.45	75.45	29.9	75.45	42.6			

terrestris was observed to be more specific and preferred fewer and certain ones (Figure 1). According to the data obtained from the species richness index, Ankara province may thought to be rich in P. *niveatus*, *M. argillaceus*, *B. terrestris* and *M. zonatus* and the other species arranged in Figure 2. When it is looked at with regard to the plants, it was found out that *Anchusa leptophylla*, *Onopordum anatolicum*, *Echium italicum*, *Trifolium pratense*, *Galega officinalis* and *Astragalus* sp. were the most preferred plant species of bumblebees either in species (Figure 3) or in density of individuals (Figure 4).

Discussion

It is known that there are several Acari species which may be parasitic, phoretic or at least mutualistic for different *Bombus* and *Psithyrus* species (1,11,16,17,19). In this study it was found that the relationship between *Acarus farris* and *B. terrestris*, *M. argillaceus*, *M. zonatus* and *P. niveatus* queens is phoresy; the mites are simply carried by the bumblebees. However, the association between mites and bees is well documented, albeit poorly understood, and more biological data are needed (20). The parasitic effect of different Protozoa species is well known. *Apicystis bombi*, *Nosema bombi* and *Crithidia bombi* from Canada, France, Finland, Italy, Switzerland, New Zealand and Denmark (1,17,21), and also a parasitic protozoon belonging to the order Neogregarinida, from Finland and Italy (22) were reported. In this study, only *N. bombi* was observed in *B. terrestris* with a high incidence (74.63%). *A nematode* species from Allantonematidae was determined to be an internal parasite of *Megabombus sylvarum citrinofasciatus* and was recorded for the first time. All the flowers visited by

bumblebees were checked for the presence of different agents. There seemed to by no relationship among the parasites, micro-organisms and flowers. Also, foraging and general ecological behaviour were not observed to be affected by nematodes or acari. However, more laboratory data are needed for *B. terrestris*.

Two main points are essential for bumblebees; first, some species are more species specific than others and second, some plant species are more attractive to foragers. It was observed by using the Shannon diversity index that *M. argillaceus*, *M. armeniacus*, *M. zonatus* and *P. niveatus*, which were common in Ankara (Figure 2), seem to visit different numbers of plant species than *B. terrestris*, *M. lapidarius*, *M. sylvarum citrinofasciatus*, *M.* ruderarius, *M. pascuorum* and *P. soroeensis*. One possible reason for *M. lapidarius*, *M. ruderarius*, *M. pascuorum* and *P. soroeensis* is that these four species are localized in the Kızılcahamam region, which is mainly a high (c.a. 2000 m) foresty area including respectively fewer plant species attractive for bumblebees. For *B. terrestris* and *M. sylvarum citrinofasciatus*, this cannot be stated. It is thought that these two species are more specific and actively choose certain flowers. It was also observed that among 37 plant species, *Anchusa leptophylla*, *Onopordum anatolicum*, *Echium italicum*, *Trifolium pratense*, *Galega officinalis* and *Astragalus* sp. were more preferred by bumblebees. When the different results of the indices were examined it was shown that *Anchusa leptophylla*, for instance, was visited by more bumblebee species (Figure 3) than *Echium italicum*, but *Echium italicum* were more densely visited than *Anchusa leptophylla* in individual number terms (Figure 4). It is reported that bumblebees prefering to visit to *Tilia* and *Echium* fall off in the middle of the day when the nectar is too concentrated (8). Bumblebee researchers generally reach suitable collection places at midday. In order to avoid an erroneous approach which may result from the dietry patterns in foraging activity, we decided to perform collections at randomly different times of the day. Our results are broadly consistent with those of Rasmont and Flagothier in southern Turkey (6). A study in the south of France at Lozère obtained similar results (23). According to that study it was observed that *M. pascuorum* (H = 4.05), *M. lapidarius* (H = 4.03), *M.* ruderarius (H = 3.58), *M. sylvarum* (H = 3.55), *M. humilis* (H = 3.48), B. lucorum (H = 3.17) and P. soroeensis (H = 3.16), which are all forest species, more commonly visited different plant species than other bumblebees. These results are consistent with our data in the Kızılcahamam region, which has a similar structure to that of Lozère. But the other steppe bumblebee species seem to have priority in Ankara province over that of southern Turkey. Later studies on a wider scale are under way for the Palaearctic.

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