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Initial data on the specific heterogeneity found in the bee pollen loads produced in the Pontevedra region (north-west Spain)

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Abstract
Heterogeneous pollen load frequency from a beehive located in the Morcón (Pontevedra, north-west Spain) was recorded. Samples of pollen loads were obtained weekly between May and July using pollen traps, and the accumulated pollen was removed at different times. Of the total analysed pollen loads an average heterogeneity percentage of approximately 5% was recorded. The pollen load heterogeneity consisted of mixed pellets with pollen of several species, but usually with one species (*Erica umbellata* L., *Lythrum salicaria* L., *Lotus corniculatus* L., *Cytisus scoparius*-type, *Castanea sativa* Miller, *Echium plantagineum* L., *Reseda media* Lag., *Erica cinerea* L., *Trifolium arvense* L., *Lonicera peryclimenum* L., *Scandix pecten-veneris* L., *Calystegia sepium* (L.) Br., *Crepis capillaris* (L.) Wallr., *Eucalyptus globulus* Labill., *Plantago lanceolata* L., -*Raphanus raphanistrum* L., *Rubus ulmifolius*- type or Poaceae) having higher values than others. It appears that mixed loads are not the result of flower deception but related to a decrease in pollen availability from plants the bees are currently foraging on that result in mixed foraging behaviour on other taxa.

Keywords: *Pollination, pollen loads, bee pollen, Galicia (Spain)*

Honeybees, *Apis mellifera* L., collect the pollen necessary for the hive from the flowering plants that grow around the apiary. Bees avoid certain species and collect different amounts from selected species present in the area (Betts, 1920, 1935). If the surrounding vegetation is abundant and attractive, foraging trips for pollen are usually short (within 2 km). However, the bees can fly from 5 to 13.5 km looking for the most attractive pollen (Schua, 1952; O'Neal & Waller, 1984). Bees adapt well to the local flora if the latter is abundant and meets the conditions of colours and aromas that serve as inducements and guide signs (Waddington & Holden, 1979; O'Neal & Waller, 1984; Dobson, 1987; Seeley, 1994; Díaz-Losada et al., 1996; De Sá-Otero et al., 2001). Other important factors in flower selection include large and resistant corolla that provides a landing platform (Dafni, 1992), nutritional value of the pollen grains (Bosi & Ricciardelli, 1975; Ricciardelli, 1979; Saa-Otero et al., 2000) and ornamental requirements (Vaissiére & Vinson, 1994).

Honeybees use the flowers as a source of pollen and nectar, and bees behave as any other insect visitor. In species with entomophilous pollination the corolla shape and colour, morphological characters that the honeybee perceives as a whole (Waser et al., 1996), and the nectar accessibility determine the class of pollinator insect that visit the flower. On the other hand, the bees require a flower form that permits landing, and flower colour and the shape that attracts its attention, and the smell of pollen to be easily identified (Proctor & Yeo, 1973). Other works have studied the importance of the corolla colours (Armesto, 1998; Osche, 1983) and shapes as an element of attraction to the pollinating insects.

When foraging, bees collect pollen from many flowers of the same species of plant until small pollen loads, of approximately 5 mm^3, are obtained. These pellets are then transported to the colony in the pollen

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baskets (Betts, 1920, 1935; Free, 1963; Maurizio, 1968). This process guarantees a great degree of effectiveness in collection, which increases the beehive production and the pollination efficiency. These pollen loads have been analysed as basic information about the biology of the honeybee (Hodges, 1984; Le Thomas et al., 1988; Lobreau-Callen et al., 1988).

Some researchers (Free, 1963; Waddington & Holden, 1979) consider that, when bees collect pollen, they remain faithful to the species of plants that it has visited and will only visit one species of flower until it completes its load, producing mono-specific pollen loads. The existence of heterogeneous pollen loads has led researchers to determine the bee's degree of fidelity to a plant species, by studying the frequency of appearance of mixed loads carried in its the pollen baskets (Clements & Long, 1923; Brittain & Newton, 1933; Thomson, 1981; Waddington, 1983).

Betts (1920, 1935), Maurizio (1949), Singh (1950), Free (1963), placed the percentage appearance of heterogeneous pollen loads at around 6%. The reasons why bees form mixed pollen loads are explained by several hypotheses that the bees become dissatisfied with the production from their habitual species and go for others. On the other hand, Bolchi & Salvi (1986) attribute this phenomenon to a confusion of the foragers when they collect pollen from species that have flowers that are similar in form, colour and size, as occurs with *Calystegia* (R.) Br. and *Convolvulus* L.

Our intention is to determine the frequency and composition of mixed pollen loads and to consider the different factors that could determine the lack of homogeneity within them and to relate the results obtained to other studies of the same nature made in a close geographic proximity (De Sá-Otero et al., 2005).

Material and methods

The study site was a beehive located in Pontevedra (Morcón), [UTM 29 X531655,29 Y4696059,48 *alt.* 42°24′57″N (−42.41591°) & 8°36′57″W (−8.6159°)], north-west Spain (Figure 1), a region located in Galicia (NW Spain), which is a part of Eurosiberian biogeographical region (Rivas et al., 1987) Samples were collected from May to July at different times of the day 12:00, 14:00, 16:00 and 18:00, using a 10% effective pollen trap with a round-holed grill, placed at the entrance to the hive (Louveaux, 1958). This period coincides with the flowering of plants foraged by bees in the study area (Table I).

Sample units consisted of pollen loads collected at each sampling hour. Samples were dried and weighed using a PRECISA 40SM-200A set of

Figure 1. Location of studied site.

scales. Each sample was then split into half. Using the Pantone Guide 747XR as a reference, pollen loads from half of the sample were separated into different colour types, and the optical microscope was used for corroboration of the specific pollen included in the mixed loads. Twenty-five pollen loads from each different colour unit were analysed (Díaz-Losada et al., 1998). Each 25 pollen load was broken down on a slide, with an inoculating needle, covered with glycerol-gel, and its homogeneity was checked using an optical microscope. The diversity and the importance of each species in the mixed pollen loads were studied. The components of each pellet were identified and their percentages were calculated from a total of 1 200 pollen grains by load (400 per slide), following the method proposed by Louveaux et al. (1978) for the calculation of pollen in honey. A total of 1 579 pollen loads, corresponding to 14 collections from the hive, were analysed.

The other half of the sample was used to determine the relative importance of each pollen species harvested using a percentage pollen analysis (Hodges, 1984; Díaz-Losada, 1995). The pollen loads were first homogenized and then acetolysed (Erdtman, 1969). The residues were mounted on phenolated glycerin, and 1 200 grains were counted per sample using preferential lines. In spite of efficiency gains in the recently developed method of Jones & Bryant (2004), we have decided to use the habitual method in our laboratory as it will provide comparability with all our data. Pollen was identified using the pollen reference collection of the Faculty of Sciences in Ourense and the use of the Palynology identification keys (Moore & Webb, 1978; Valdés et al.,1987; Ramil et al., 1992; Saa-Otero et al., 1996).

Table I. Flowering period (⊛) of the species located close to the hive (updated from de Sá-Otero et al., 2001).

Taxa	February	March	April	May	June	July	August	September	October
Adenocarpus complicatus (L.) Gay				⊛	⊛	⊛	⊛	⊛	
Anthemis arvensis L.				⊛	⊛	⊛	⊛	⊛	
Arenaria montana L.				⊛	⊛	⊛			
Brassica napus L.				⊛	⊛	⊛			
Brassica oleracea L.				⊛	⊛	⊛	⊛		
Calystegia soldanella (L.) R. Br.						⊛	⊛		
Cistus psilosepalus L.				⊛	⊛	⊛			
Crataegus monogyna Jacq.				⊛	⊛				
Crepis capillaris (L.) Wallr.					⊛	⊛	⊛	⊛	⊛
Cytisus multiflorus (L'Hér) Sweet			⊛	⊛	⊛				
Cytisus scoparius-type			⊛	⊛	⊛				
Cytisus striatus (L.) Rothm.	⊛	⊛	⊛	⊛	⊛				
Echium vulgare (L.)				⊛	⊛	⊛	⊛		
Erica arborea L.		⊛	⊛	⊛					
Erica australis L.	⊛	⊛	⊛	⊛	⊛	⊛	⊛		
Erica cinerea L.				⊛	⊛	⊛	⊛	⊛	
Erica umbellata L.			⊛	⊛	⊛	⊛	⊛	⊛	
Eucalyptus globulus Labill.			⊛	⊛	⊛	⊛	⊛		
Genista tridentata L.				⊛	⊛	⊛			
Halimium alyssoides (Lam.) K. Koch			⊛	⊛	⊛	⊛	⊛		
Hedera helix L.								⊛	⊛
Iris pseudacorus L.					⊛	⊛			
Lonicera peryclimenum L.						⊛	⊛		
Lythrum salicaria L.					⊛	⊛	⊛	⊛	
Lythrum junceum Banks & Solander					⊛	⊛	⊛		
Lotus corniculatus L.				⊛	⊛	⊛	⊛	⊛	
Lotus pedunculatus Cav.				⊛	⊛	⊛	⊛		
Muscari comosum (L.) Miller				⊛	⊛	⊛			
Plantago lanceolata L.			⊛	⊛	⊛	⊛	⊛	⊛	⊛
Prunus spinosa L.		⊛	⊛	⊛					
Quercus pyrenaica Willd.			⊛	⊛					
Quercus robur L.			⊛	⊛					
Ranunculus repens L.				⊛	⊛				
Raphanus raphanistrum L.			⊛	⊛	⊛	⊛	⊛	⊛	
Reseda media Lag.		⊛	⊛	⊛	⊛	⊛	⊛		
Rubus L.					⊛	⊛	⊛	⊛	
Rumex acetosa L.				⊛	⊛	⊛	⊛		
Salix fragilis L.		⊛	⊛						
Scandix pecten-veneris				⊛	⊛				
Scilla autumnalis L.							⊛	⊛	⊛
Scrophularia canina L.		⊛	⊛	⊛					
Sedum acre L.		⊛	⊛	⊛					
Solanum nigrum L.					⊛	⊛	⊛	⊛	⊛
Spergularia rubra (L.) J. Presl. & K. Presl.				⊛	⊛	⊛	⊛	⊛	
Stellaria media (L.) Vill.		⊛	⊛	⊛	⊛	⊛	⊛	⊛	
Trifolium pratense L.				⊛	⊛	⊛	⊛	⊛	
Trifolium repens L.				⊛	⊛	⊛	⊛	⊛	⊛
Ulex europaeus L.			⊛	⊛	⊛	⊛			
Ulex minor Rothm.						⊛	⊛	⊛	
Vitis vinifera L.				⊛	⊛				
Zea mays L.						⊛	⊛	⊛	⊛

Results

Pollen load heterogeneity

The amount of pollen collected varied according to the hour and it was noticed that more pollen was collected in the middle hours of the day (16:00) (Table II). The average frequency of heterogeneous pollen loads found is around 6% for the whole period studied. The percentage of heterogeneous pollen loads in the total analysed samples varied in the different days and hours of collection. Most of the samples have a heterogeneous pollen loads percentage of less than 5%, except the samples from 19 June at 16:00 and 18:00, and 5 July at 16:00 and 18:00 in which percentages of 6.48 from 9.94 was observed (Table II).

The study of the pollen flora collected by the bees revealed that bees collected the most pollen from the following plants: *Scandix pecten-veneris* in the middle of May, *Cytisus scoparius*-type at the end of May, *Rubus ulmifolius*-type in the middle of and at the end

Table II. Weight (g) of pollen obtained in each harvesting collection, the total number of pollen loads analysed in, the number of species identified (diversity), the total number of heterogeneous pollen loads, the percentage of heterogeneous pollen loads in each collection and the number of species in heterogeneous pollen loads of each harvesting that were found.

Collections	Weight total (g)	No. of pollen loads analysed	Pollen types no by sample	No. of heterogen. loads	Heterogen in the sample (%)	Species no. by heterogen. loads
May 15/16:00	8.857	48	4	1	2.08	5
May 15/18:00	8.078	125	6	2	1.60	6
May 22/16:00	7.310	107	6	5	4.67	12
June 5/12:00	2.995	125	12	6	4.8	9
June 12/16:00	6.276	159	16	8	5.03	13
June 12/18:00	3.502	103	8	4	3.88	8
June 19/16:00	10.353	134	8	10	7.46	10
June 18/18:00	4.910	108	6	7	6.48	9
June 29/16:00	5.164	117	5	4	3.42	7
June 29/18:00	6.292	78	6	3	3.85	5
July 5/16:00	9.021	161	6	16	9.94	8
July 5/18:00	4.409	114	6	10	8.77	7
July 23/16:00	6.360	91	8	1	1.10	2
July 23/18:00	2.570	109	8	4	3.67	8

of June and *Zea mays* at the end of July (Table III). Species that had relatively lower values include *Cytisus scoparius*-type that was collected in middle of May and beginning of June and July. *Scandix pecten-veneris* and *Eucalyptus globulus* was collected towards the end of May, and *Erica umbellata* in June. *Plantago lanceolata* was collected at beginning of June, *Echium plantagineum* at middle of June and the end of July, *Castanea sativa* in the beginning of July and *Erica cinerea* at the end of July (Table III).

Small amounts of *Reseda media* pollen was collected in May and in the first half of June, *Echium plantagineum* in the middle and the end of May, *Eucalyptus globulus, Vitis vinifera, Scandix pecten-veneris* and *Castanea sativa* in the beginning of June. *Plantago lanceolata* and *Lonicera peryclimenum* was collected in the middle of June, *Cytisus scoparius*-type in the middle and the end of July, and *Lotus corniculatus* at middle of June to beginning of July (Table III).

Within the heterogeneous pollen loads, 24.69% of the total consisted of mixed pollen loads in which the major pollen was *Erica umbellata*, 16.04%. In decreasing order of abundance were heterogeneous pollen loads of: *Lythrum salicaria*; 13.58%, *Lotus corniculatus*; 7.04%, *Cytisus scoparius*-type; 7.40%, *Castanea sativa*; 6.17%, *Echium plantagineum*-type, 3.70% *Reseda media*; 3.70%, *Erica cinerea*; 3.70%, *Trifolium arvense*; 2.46%, *Lonicera peryclimenum*; 2.46%, *Scandix pecten-veneris*; 1.23%, *Calystegia sepium*; 1.23%, *Crepis capillaris*; 1.23%, *Eucalyptus globulus*; 1.23%, *Plantago lanceolata*; 1.23%, *Raphanus raphanistrum*-type; 1.23%, *Rubus ulmifolius*-type and 1.23%, Poaceae.

In terms of frequency all the samples have heterogenous pollen loads. However, the collection made on 5 July at 16:00 had the highest number of heterogeneous loads (16) with the majority containing *Trifolium arvense* as the most abundant pollen.

The first heterogeneous loads were recorded in the middle of May, with two samples at different hours from the same day (15 May: 16:00 and 18:00). The percentage of heterogeneity is low, around 2% (Table II) with the number of mixed pellets recorded being 1 and 2 respectively. In day 22 May, a sample was collected at 16:00, its percentage of heterogeneity is 4.67 (Table II) with five heterogeneous pollen loads. Although the absolute number of pollen loads is small it can be seen that the relative percentages of different pollen types that comprise the mixed pollen loads vary with a diversity of species being collected (Figure 2).

The samples with the most number of heterogeneous pollen loads were collected in the beginning July and in the second half of June (5 June at 16:00 and 18:00. and 19 June at 16:00 with 16, 10 and 10 mixed pollen loads respectively, Table II). The mixed pollen loads with *Lythrum salicaria, Lotus corniculatus, Echium plantagineum, Raphanus raphanistrum* and *Erica umbellata* as the principal component were formed during June (Figures 3, 4).

The number of heterogeneous pollen loads brought to the beehive by the bee determines the degree of constancy for a particular plant species (Thomson, 1981; Waddington, 1983). At Morcón (Pontevedra), the degree of constancy shown by the bees for shrubland species is high. Only 5% of the pollen loads are heterogeneous, during the period studied.

Pollen flora

As established in the material and methods section, the sample of each harvesting has been divided into two fractions; half of sample has been utilized for heterogeneity analysis of pollen loads (described above). The other half has been homogenized, acetolysed and a pollen percentage analysis has been

Table III. Shows the relative importance of each species by sample, identified in the microscopic slides of the 50% utilised for pollinic percentage analysis. (*) Percentage <0.1%.

	Samples													
	15-V		22-V	5-VI	12-VI		19-VI		29-VI		5-VII		23-VII	
Pollen types	16 h	18 h	16 h	12 h	16 h	18 h	16 h	18 h	16 h	18 h	16 h	18 h	16 h	18 h
Anthemis arvensis	*	*	*		*		*	*						
Calystegia sepium														
Castanea sativa				3.7	*	*	1.6	1.5	0.2		41.3	36.7	0.4	
Crepis capillaris					1.2	1					*	*		
Cytisus scoparius-type	38.4	42.9	46.8	14	2.7	1.9	3.6	1.6	9	2.3	16.6	1.2	12.5	57.8
Echium plantagineum	0.3	6.4	3.8	5	36.16	30.5	8.7	22.4		1.1	1.4	0.9		15.2
Erica umbellata				24.7	32.05	30.1	6.4	14.2	16.2	*	4.4	7.8		
Erica cinerea													16.9	
Eucalyptus globulus	1.6	9.8	13.9	7	0.6	0.3								
Iris pseudocorus					0.4									
Lotus corniculatus						0.7	0.5	2.4	2.2	4.6	1.6	6.1	1.5	
Lonicera peryclimenum					3.2									
Lythrum salicaria				0.8		*	*		*					
Lythrum junceum				*			*		*					
Muscari comosum					0.2									
Olea europaea					0.2									
Plantago lanceolata		*	0.8	15.5	6.5			*			*		2.03	
Pinus pinea			*											
Poaceae			*	8	2	*	*	*	*	*	*			
Raphanus raphanistrum-type							0.5							
Reseda media		3.6	13.1	9	10.2	2.5	0.5							
Rubus ulmifolius					0.3	33.2	78.2	58	72.4	91	35	47.3		1.7
Rumex acetosa			*											
Scandix pecten-veneris	59.7	37	21.6	5.4										
Spergularia arvensis		0.3		0.8	0.8									
Trifolium arvense												*		1.07
Vitis vinifera					5.8									
Zea mays													84.3	5.4

done, in order to gauge in advance the relative importance of each harvested pollen species by the honeybee (Table III). The species whose pollen has been recollected in abundance were *Cytisus scoparius*-type which constitutes an important part of honeybee pollen from May to July with varying percentages depending on the month and hour collected. For example, on 23 July bees harvested more intensively at 16:00 compared to 18:00. The other important taxa are: *Scandix pecten-veneris* which is harvested in May only; *Reseda media*, *Plantago lanceolata* and *Eucalyptus globulus* that are harvested at end of May and beginning of June; *Erica umbellata* and *Echium plantagineum* which are important in June. *Rubus ulmifolius*-type is one of the more important species in June and beginning of July and *Castanea sativa* at the beginning of July. The harvesting of *Zea mays* on 23 July is anecdotal.

Discussion

A comparison between the abundance of the species collected as part of the total pollen flora and composition of the heterogeneous pollen loads reveals important differences. For this, let us suppose that heterogeneous pollen loads frequently have a mix of pollen from plants which were not relevant as an important source of pollen for the honeybee. The sample from the middle of May (15 May at 16:00), shows the heterogeneous pollen loads contain *Reseda media* pollen as principal taxon. In contrast the total pollen flora harvested by the honeybees on 15 May (16:00) does not contain this species (Table III). Thus it is not likely that the formation of mixed pollen loads is due to physical contact of different pollen loads present in the trap drawer from which the samples are collected for analysis.

It was not possible to establish a relationship, based on the few cases of mixed loads, between the amount of total harvested pollen in each sample, and the number of heterogeneous pollen loads. Samples that weighed 8.857 g and 6.360 g had one heterogeneous pollen loads and samples with a higher number of heterogeneous pollen loads (16) weighed 9.021 g but, another with a mean number (10), weighed 10.353 g of pollen (Table II).

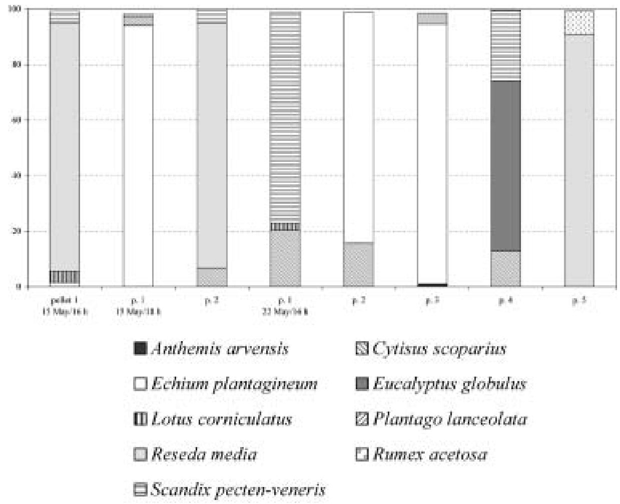

Figure 2. Relative composition of the heterogeneous loads identified in the harvest of May.

A relationship does not seem to exist between the diversity of species visited and the frequency of heterogeneous pollen loads. Thus, there were collections in which five and two different species were identified in a total sample by microscopic analysis and the number of mixed pollen loads was one pollen load in both cases. In other collections the number of pollen types identified per collection, were 8, 6, and 6 and the number of heterogeneous pollen loads was 10, 16, and 10 respectively in samples of 19 June at 16:00, 5 July at 16:00 and 18:00 h (Table II).

There may be a relationship between the number of species in flower and heterogeneity. Thus, in May, the number of species in flower with interest for the honey bee was 22 (Table I) and the percentage of heterogeneity in the pollen loads was 1.6–4.67%. In July, 26 species were in flower and the percentage of heterogeneity varied between 1.1 and 9.94%. Whereas, in June, 25 species were in flower and the percentage of heterogeneity varied from 3.42 to 7.5%.

The availability of pollen from certain species could also be an interesting factor related to foraging process. For example, in the collection in which the amount of *Rubus ulmifolius*-type brought to the hive was 78.24% of the total (Table III), the percentage of heterogeneity was 7.46% (Table II). Whereas, in the collection in which *Rubus ulmifolius*-type represented 57.98% of the total, the heterogeneity percentage was 6.48%. It is probable that the number of *Rubus ulmifolius*-type flowers with available pollen was low, and the bee needed to visit a larger number of flowers to complete its pollen load, and therefore had more contacts with floral stigmas that could have been contaminated with pollen from other species.

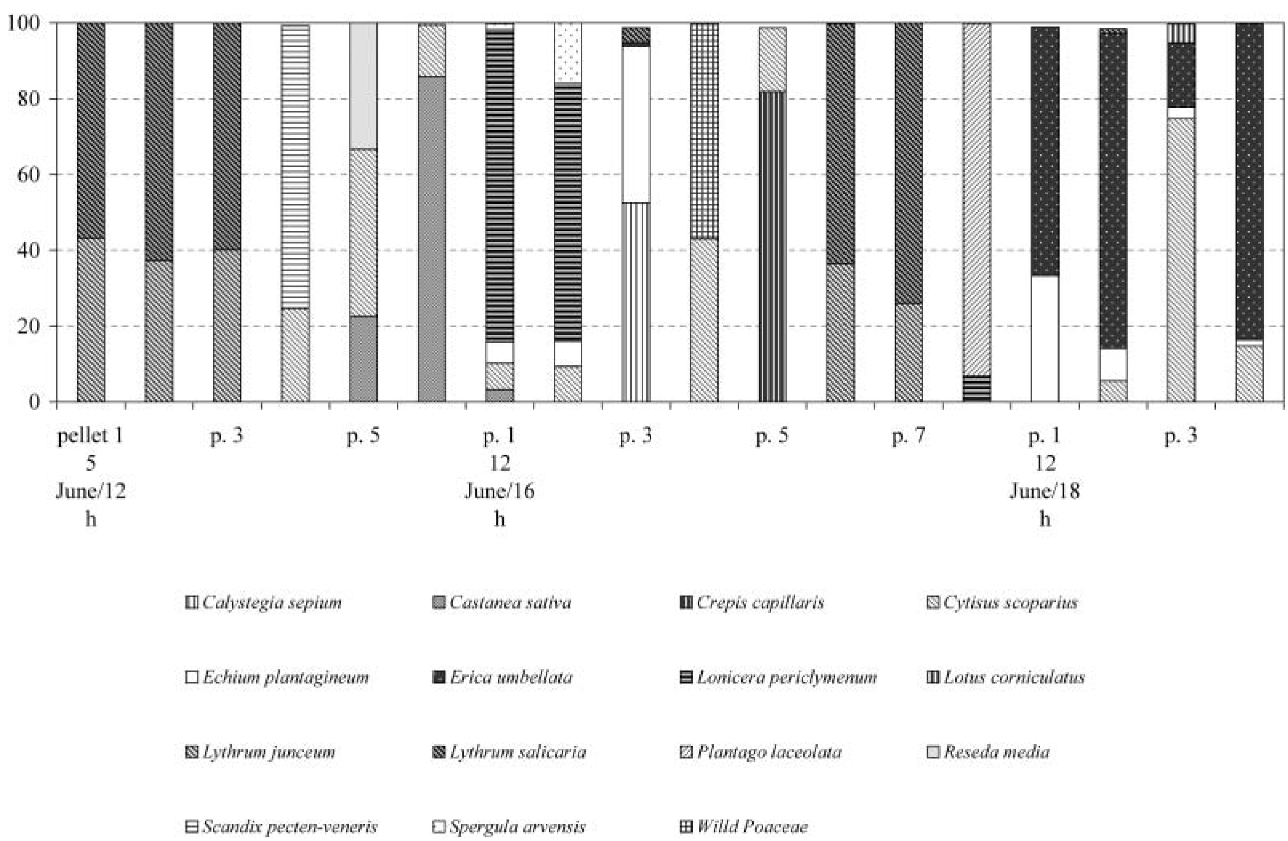

Figure 3. Relative specific composition of the heterogeneous loads identified in the harvest of 1–15 June.

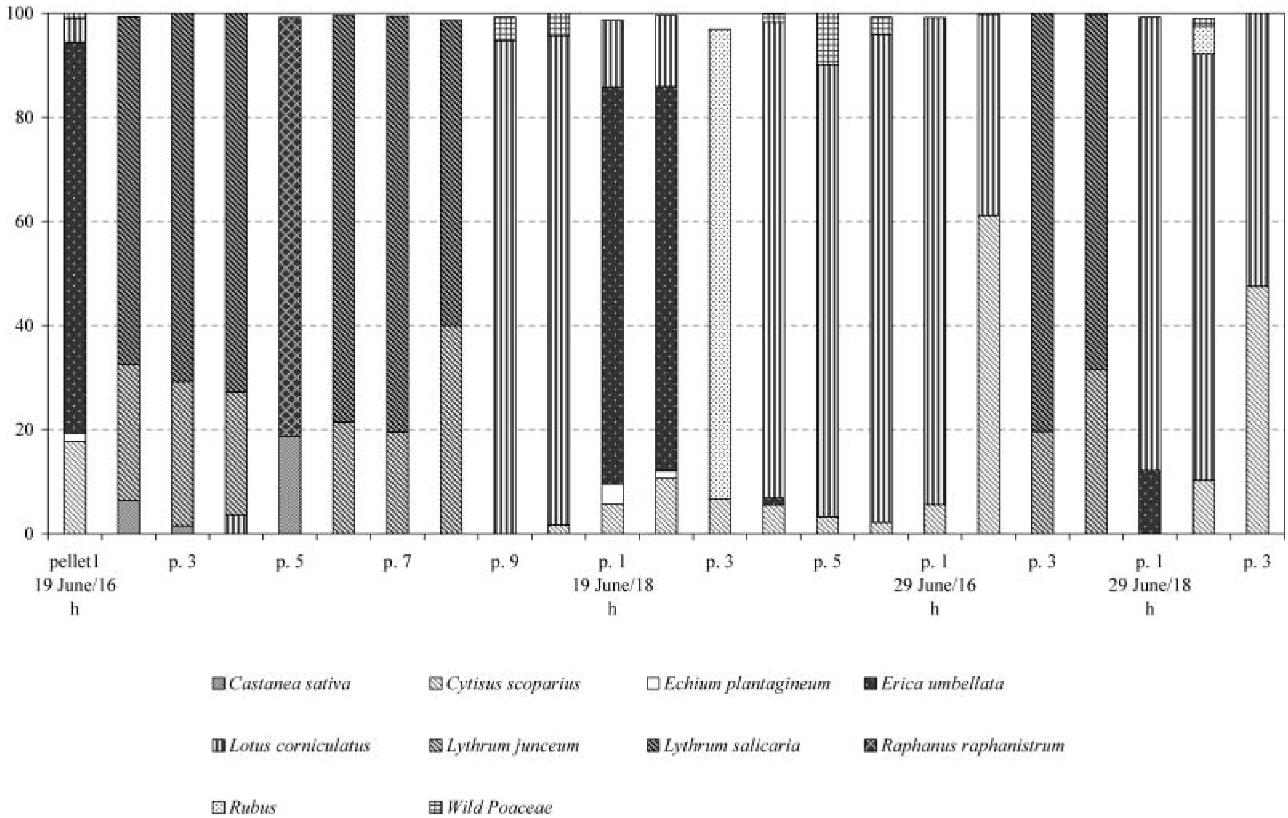

Figure 4. Relative specific composition of the heterogeneous loads identified in the harvest of 15–30 June.

Attention was drawn to the fact that the specific composition of the heterogeneous pollen loads showed a higher percentage of pollen from the species that were at the end and at the start of their flowering period, and a lower percentage from species that were at their peak. The phenology of each source species with regard to the phenology of other species is an interesting factor also in the pollen foraging process. For example, when *Cytisus scoparius*-type pollen represented 57.79% of the total amount collected, the heterogeneity percentage was 3.67%; whereas on the 5 July at 18:00 collection, when the hive harvested only 1.20% of total pollen from *Cytisus scoparius*-type, and when others pollen sources began to gain importance, the heterogeneity percentage reached its highest peak of 8.77% (Table II).

A high degree of affinity was observed in relation to species of unisexual flowers. In this case, it would be convenient to note that there is no possibility of involuntary contamination with stigmatic pollen. There is a higher risk of formation of heterogeneous pollen loads when the bee visits flowers with a low pollen production in their stamens; either because their anthers are small or reduced in number or because of their low production of pollen. In flowers with a low level of pollen production, the bee must visit a large number of flowers in order to form a pollen load (585 flowers, 106 to 166 flower heads, in *Trifolium repens*, in Percival, 1950).

It should be also taken into consideration that the probability of heterogeneity may be related to the presence of foreign pollen on the stamen of the visited flowers that passes on to the body of the bee which is then included involuntarily, when the bee combs its body, to the pollen load.

The heterogeneous pollen loads with *Rhaphanus raphanistrum* as principal component (Figure 2) also had *Castanea sativa* as the common pollen and other species with different floral morphology, corolla colour, apetalous flowers or anemophilous pollination (Poaceae).

In general it can be said that the heterogeneous pollen loads, no matter what the dominant species (Figures 2–5), were formed by pollen combinations of plants which had floral characteristics that are very different among themselves and with the dominant species. It would appear that the possible confusion of the forager bees between the plants of the similar floral morphology proposed by Bolchi & Salvi (1986) is not a factor that determines the phenomenon of heterogeneous pollen loads. However combinations of species of the some vegetation formations predominate in the heterogeneous pollen loads.

From 5 July the highest percentage of pollen harvested is from *Castanea sativa* and *Erica* species, and during this period the highest numbers of heterogeneous pollen loads are observed. It is possible that *Castanea sativa* and *Erica* sp. pollen production is unsatisfactory, forcing the honeybee to explore new plants resulting in heterogeneous pollen load production

Apis mellifera bees are pollinating insects of a generalist nature; they visit a wide range of plant species, obtaining pollen and nectar from their flowers. They adapt everywhere to the characteristics of the local vegetation, although they have preferences for different species in relation to the nutritional reward offered by each of them, as they are able to distinguish plants by their flower form, size, or colour (van der Pijl, 1960; Fraegri & van der Pijl, 1971; Proctor & Yeo, 1972; Stebbins, 1981; Davis, 1997)

De Sá-Otero et al. (2005) established that heterogeneous pollen loads consisted of species that grow in associated vegetation formations near the beehive. Thus, combinations of different types of heather with species of *Halimium alyssoides* and *Cytisus scoparius*-type; combinations of meadow species with *Lotus corniculatus* pollen together with *Plantago lanceolata*, *Geranium molle* or *Anthemis arvensis* L. pollen, and combinations of rock or roadside species (*Sedum acre* together with *Reseda media* pollen) occur. In this study it has not been possible to reach a similar conclusion, perhaps because the beehives are located in an area where the vegetation communities have been disturbed by human action (Table IV).

Conclusions

The percentage of heterogeneous pollen loads is around 6% and similar values determined by others researchers (for example: Betts, 1920, 1935; Singh, 1950; Free, 1963). It can be concluded that the degree of affinity of the honeybees for the local vegetation is similar to the ones proposed by the above mentioned researchers. The greatest amount of pollen was gathered to 16:00 local time

The species diversity of pollen that constituents the mixed pellets varies from 2 to 13 species by load, and their corolla form and size differ to each other (*Erica umbellata, Lythrum salicaria, Lotus corniculatus, Cytisus scoparius*-type, *Castanea sativa, Echium plantagineum, Reseda media, Erica cinerea, Trifolium arvense, Lonicera peryclimenum, Scandix pecten-veneris, Calystegia sepium, Crepis capillaris, Eucalyptus globulus, Plantago lanceolata, Raphanus raphanistrum, Rubus ulmifolius*-type and Poaceae). This suggests that the origin of heterogeneous pollen loads is not the result of confusion by the bees between morphologically similar flowers of different species.

May and June is the optimal period for the flowering of species of pollen interest in Galicia (Table I). We observed an increase of mixed pellets and greater diversity of species in composition of mixed pollen loads when the bees have carried large amounts of pollen (12 June at 16:00 and 19 June at 16:00). At the end of July, when the bee carries smaller amounts of pollen to the beehive, a greater

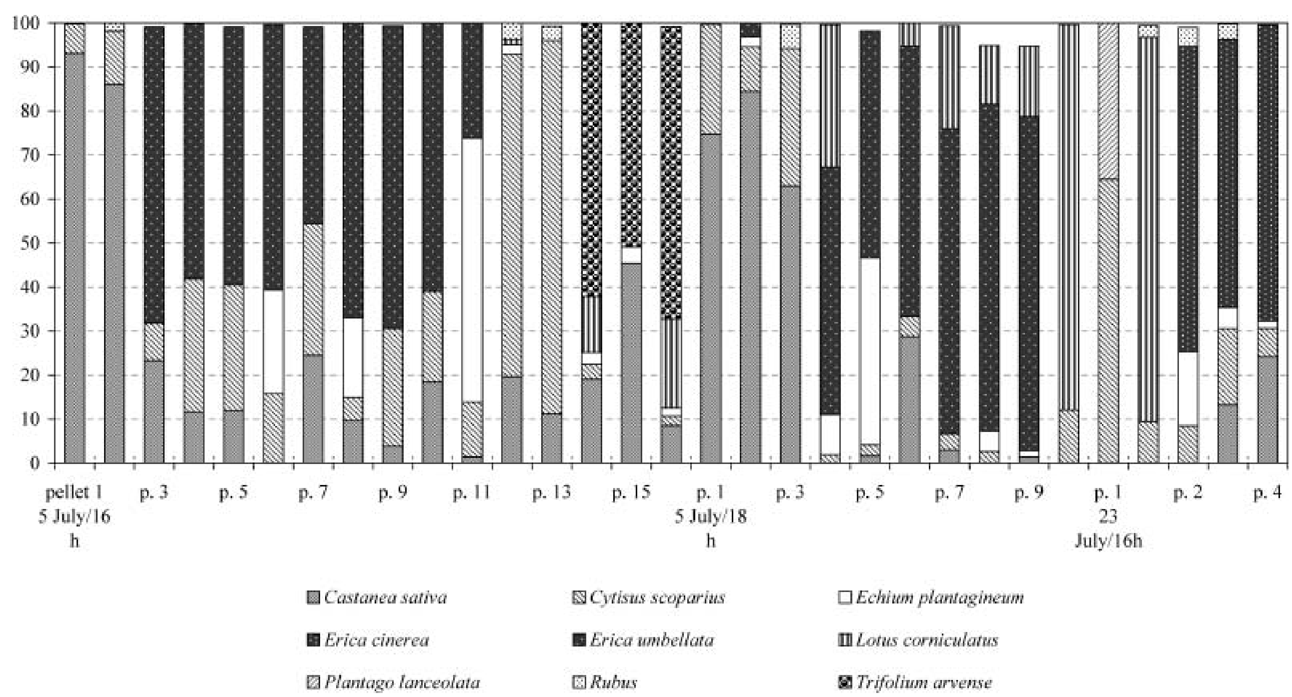

Figure 5. Relative specific composition of the heterogeneous loads identified in the harvest of July.

Table IV. Representation of the combinations of the principal component and other components in the loads found in this study.

Heterogeneous pollen loads

	Other Components															
Principal Component	*Anthemis arvensis*	*Calyst. sepium*	*Cast. sativa*	*Cytisus scopar.*	*Echium plantag.*	*Erica arborea*	*Erica umbell.*	*Lonicera pericly.*	*Lotus cornicul.*	*Lythrum salicaria*	*Lythrum junceum*	*Plantago lanceol.*	*Rubus ulmifol.*	*Rumex acetosa*	*Scandix pec.-ven.*	*Spergula arvensis*
Calystegia sepium					■		■			■						
Castanea sativa				■	■		■						■			
Crepis capillaris		■		■												
Cytisus scoparius			■		■		■		■				■			
Echium plantagineum	■		■	■			■					■				
Erica cinerea			■	■	■								■			
Erica umbellata			■	■	■				■							
Eucalyptus globulus				■											■	
Lonicera periclymenum			■	■	■											■
Lotus corniculatus				■			■						■			
Lythrum salicaria			■								■					
Plantago lanceolata								■								
Wild Poaceae				■												
Raphanus raphanistrum			■													
Reseda media				■					■					■	■	
Rubus ulmifolius			■						■							
Scandix pecten-veneis				■					■							
Trifolium arvense			■	■	■				■							

number of mixed pellets and greater diversity of species in mixed pollen loads occurs (Table II). The data that suggests when the main species of pollen interest are at the end of their period of flowering, and the amount of pollen produced diminishes; the bee is forced to visit greater number of plants to form a pollen load. It is likely the energy required to gather minor amounts of the same species pollen to form a pollen load results in the exploration of new species resulting in a greater number of mixed pellets and greater diversity of species in pollen loads

Pollen loads composition demonstrates that the honeybees harvest pollen preponderantly from the most abundant species in their environment, which form dense populations, but when the pollen offered begins to decline they search for and try out other sources that share the characteristics of abundance and high density and in this way mixed pollen loads are produced.

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