

ORIGINAL ARTICLE

Diversity of predaceous arthropods in the almond tree canopy in Northeastern Portugal: A methodological approach

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Abstract

The almond tree is an economically important crop in Mediterranean regions. However, knowledge about the biodiversity of natural enemies that may be useful as biocontrol agents is scarce. The objectives of this work were: (i) to study the diversity of predaceous arthropods; and (ii) to establish a suitable sampling protocol for arthropods of the almond tree canopy. Between April and October of 2007 and 2008, 25 randomly selected trees were sampled in an organic almond grove located in the north-east of Portugal using the beating technique. The specimens collected were counted and identified and the sampling protocol was established by using the accumulation curves and the seasonal richness peaks of the most abundant groups of natural enemies. A total of 1856 and 1301 arthropods were captured, respectively, in 2007 and 2008, where Araneae, Coccinellidae and Formicidae were the most abundant groups. A total of 14 families and 29 species of spiders were identified as Linyphiidae, Philodromidae, Thomisidae, Araneidae and Oxyopidae, the five most abundant families in both years. In the Coccinellidae and Formicidae communities 15 and 13 species were identified, respectively. According to taxa accumulation curves, the minimum sampling effort that provided a reliable picture of the biodiversity was established in 11 samples. Moreover, considering the seasonal richness distribution, it would be advisable to concentrate the sampling period from the beginning of July to the harvesting of almonds. This protocol might generate accurate replicate samples to estimate species richness when the effect of agricultural management is studied.

Key words: Araneae, Coccinellidae, Formicidae, *Prunus dulcis*, sampling protocol, species accumulation curves.

INTRODUCTION

Almond production represents an economically important agricultural resource in the Mediterranean countries (Monteiro *et al.* 2003). Characteristic climatic conditions occurring in this region, with temperate winters and hot summers, are required for a high-quality production of almond. In Portugal, Trás-os-

Montes is the major region for almond production, where the high quality of the fruit constitutes an important motive for good marketing and maintenance of populations in marginal areas (Monteiro *et al.* 2003; Anon. 2007).

Nevertheless, almond production still faces problems due to a regular occurrence of arthropod pests representing the main cause of the depletion of both crop yield and foodstuff quality (Monteiro *et al.* 2003). The most serious pests that attack the almond tree *Prunus dulcis* (Miller) D.A. Webb are the European red mite *Panonychus ulmi* (Koch) (Acari: Prostigmata), web-spinning mites such as *Tetranychus urticae* Koch (Acari: Prostigmata), the peach twig

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borer *Anarsia lineatella* Zeller (Lepidoptera: Gelechiidae), the lace bug *Monosteira unicostata* (Mulsant & Rey) (Hemiptera: Tingidae), and aphid species such as *Brachycaudus amygdalinus* (Schouteden) (Hemiptera: Aphididae) and *Myzus persicae* Sulzer (Hemiptera: Aphididae) (Liotta & Mamiglia 1994; Russo *et al.* 1994; Zalom 1994).

The growth of pest populations can be influenced by both climatic conditions and multitrophic interactions with the complex of arthropods that coexist on the almond tree canopy and contribute in maintaining pest levels below the economic thresholds. However, knowledge about the biodiversity of those natural enemies associated with this crop is scarce. In a prior survey carried out in Turkey, Bolu *et al.* (2008) identified a total of 21 species of spiders belonging to 16 genera and 9 families. The species composition for other predator groups is quite unknown.

In order to estimate the abundance and species composition of natural enemies in biodiversity assessment studies, reliable field sampling programs capable of collecting the biodiversity present in an agroecosystem are needed (Jiménez-Valverde & Lobo 2006). To develop such programs, appropriate sampling efforts and the seasonal dynamics of the studied assemblages must be well known. Species accumulation curves have been used as a tool where the cumulative sampling effort carried out in an area is related to the numbers of species that are being sequentially added to the inventory (Soberón & Llorente 1993; Jiménez-Valverde & Hortal 2003). The slope of the curve at each point determines the species accumulation rate at that sampling level (Hortal & Lobo 2005). The choice of the sampling period is another important strategy that should be used in order to optimize the required field effort and must be done, bearing in mind the goal of the study. In this context, Jiménez-Valverde and Lobo (2006) suggested limiting the surveys to the richest season, therefore reducing the seasonal coverage of samplings. Moreover, arthropod inventories ideally should be prompt, repeatable, quantitative and as cost-effective as possible (Oliver & Beattie 1996; Fisher 1999). Thus, in the case of pest control, these issues can be achieved by directing the sampling effort in a way as to characterize the community of arthropods associated with the same vertical stratum of the pest in the crop (Marc *et al.* 1999).

In this context, the objectives of the present work were: (i) to study the diversity of predaceous arthropods in the almond tree from Trás-os-Montes region (north-eastern Portugal); and (ii) to establish a suitable sampling protocol for arthropods of the almond tree canopy where the minimum sampling effort and the best time for sampling were assessed.

MATERIALS AND METHODS

Study site

The study area was located in an organic almond grove of approximately 17 years of age in Vilarinho dos Galegos, Mogadouro (north-eastern Portugal) (41°15'52"N, 6°37'48"W). The grove covers an area of 2 ha, the planting density is of 5 m × 7 m and the predominant cultivars are Ferraduel and Ferragnes. During the sampling years, trees were slightly pruned and the soil was ploughed superficially. No phytosanitary treatments were done during the experiments and soil management was conducted by planting leguminous plants and grasses between the trees as a measure to increase fertilization.

Sampling of arthropods

The sampling period occurred between April and October of 2007–2008 and arthropods were collected by the beating technique on a two-weekly basis. Each sample was collected by the same operator beating two branches per tree over a rectangular cloth (0.6 m × 0.5 m) between 09:00 and 10:00 hours. In each sampling period, samples were collected from 25 randomly selected trees. Collected arthropods were immediately transferred to a box with ice to diminish their activity. Rainy days were avoided in order to prevent a reduction in the efficiency of the sampling method. All individuals were sorted, identified and counted using a binocular microscope.

Due to their abundance and importance as natural enemies, spiders, coccinellids and ants were identified to species level and used for sampling effort estimation. Spiders were identified to species according to Locket and Millidge (1951, 1953), Hubert (1979) and Roberts (1996) and the nomenclatural system according to Platnick (2010). For this group, juveniles that could be taxonomically identified (to family, genera or species level) were included in the data analysis. Ants were identified according to Collingwood and Price (1998). Coccinellid identification was based on external characteristics but extraction and observation of the genitals of some species were needed to confirm the morphological identification. Coccinellid species were identified according to Raimundo and Alves (1986) and Raimundo (1992).

Data analysis

The total abundance of each taxon was calculated for every year of the study. The frequencies (*f*) of the different taxa were calculated as the number of samples in which the taxon was present, and the occurrence (*O*) in

percentage was calculated as $O = (f/n) \times 100$ (Eqn 1), where n is the total number of samples.

Species or family accumulation curves were used to estimate richness in the almond grove. This method illustrates the rate at which new taxa were added to the inventory within a defined area. As the number of samples increases, an increasing number of taxa are sampled, reaching a plateau. The resulting diagram shows the cumulative number of taxa recovered according to the increase of the number of samples considering the sampling period where more individuals and species were captured. The Estimates version 8.2 software (Colwell 2009) was used to calculate the accumulation curves for each taxon (Araneae, Formicidae, Coccinellidae). The curves were modeled using Statistica version 7.0 (StatSoft 2008) and adjusted by the Clench model (Soberón & Llorente 1993; Jiménez-Valverde & Hortal 2003) according to the equation:

$$S_n = a \times n / (1 + bn) \quad (\text{Eqn 2})$$

where S_n is species richness, a represents the increase rate at the beginning of the collection, b is a parameter related with the curve shape and n is the sampling effort.

The proportions of the recorded taxa were calculated as $F(\%) = S_{obs}/(a/b) \times 100$ (Eqn 3), where S_{obs} represents the total taxa richness and a/b is the asymptote of the curve. The quality of the inventory was calculated as $C_i = a/(1 + bn)^2$ (Eqn 4), where C_i is the slope of the curve at each sampling point. When $C_i < 0.1$ the inventory can be considered complete and reliable. According to Jiménez-Valverde and Hortal (2003), the number of samples (n) that define the quality of the inventory was calculated as the point of the curve that fitted a slope equal to 0.1.

RESULTS

Abundance and richness of natural enemies

A total of 1856 and 1301 arthropods were captured in the almond tree, respectively, in 2007 and 2008 (Table 1). From those, Araneae dominated the community of natural enemies with 957 individuals collected in 2007 and 616 in 2008, followed by Coleoptera, from which Coccinellidae were collected in high abundance, Formicidae, Neuroptera, Heteroptera and Dermaptera. In general, the abundance of each taxon was higher in 2007 than in 2008.

Considering the community of Araneae, a total of 1573 individuals were captured during the two years, belonging to at least 27 genus and 29 species (including morphospecies) in 14 families (Table 2). From the 1573 individuals collected, 41 (2.7%) were adults and 1532 (97.3%) were immature.

In 2007, the community of Araneae was dominated by the family Linyphiidae followed by Philodromidae, Araneidae, Thomisidae, Oxyopidae and Salticidae (Table 2). The less abundant families represented 3.8% of the total. In 2008, the community of Araneae was dominated by Philodromidae, Linyphiidae, Oxyopidae, Thomisidae, Araneidae and Dictynidae. The less abundant families in that year represented 9.8% of the total. The most diverse families were Araneidae with six species, followed by Thomisidae with five and Linyphiidae and Theridiidae with three species. On the other hand, families Clubionidae, Lycosidae, Gnaphosidae and Uloboridae were represented by singleton species. In 2007, the most abundant species were *Nigma puella* (Simon), *Synema globosum* (Fabricius), *Philodromus praedatus* O.P. Cambridge, *Salticus scenicus* (Clerck), *Phylloneta impressa* (C.L. Koch) and *Runcinia*

Table 1 Total number and relative abundance of natural enemies collected in the almond tree canopy during the two years studied (Mogadouro, 2007–2008)

Taxon	2007 ($n = 255$)		2008 ($n = 325$)	
	<i>N</i>	Relative abundance %	<i>N</i>	Relative abundance %
Order Araneae	957	55.3	616	51.9
Order Heteroptera	44	2.5	30	2.5
Family Coccinellidae	94	5.4	86	7.2
Other Coleoptera	479	27.7	380	32.0
Order Neuroptera	43	2.5	44	3.7
Family Formicidae	104	6.0	30	2.5
Order Dermaptera	10	0.6	1	0.1
Total	1856		1301	

n , number of samples; N , total number of natural enemies.

Table 2 Total number of Araneae species, percentage abundance, number of samples where the species occurred and occurrence observed in total samples collected in the almond tree canopy during the two years studied (Mogadouro, 2007–2008)

Family/Species	2007 (<i>n</i> = 255)				2008 (<i>n</i> = 325)			
	<i>N</i>	Abundance %	<i>f</i>	O (%)	<i>N</i>	Abundance %	<i>f</i>	O (%)
Family Anyphaenidae								
<i>Anyphaena sabina</i> C.L. Koch		–	–	–	4	0.7	4	1.2
<i>Anyphaena</i> spp.	1	0.1	1	0.4	–	–	–	–
Subtotal	1	0.1	1	0.4	4	0.7	4	1.2
Family Araneidae								
<i>Aculepeira armida</i> (Audouin)	1	0.1	1	0.4	–	–	–	–
<i>Agalenatea redii</i> (Scopoli)	–	–	–	–	3	0.6	3	0.9
<i>Araneus diadematus</i> Clerck	2	0.2	2	0.8	–	–	–	–
<i>Araniella cucurbitina</i> (Clerck)	1	0.1	1	0.4	–	–	–	–
<i>Araniella opisthographa</i> (Kulczynski)	1	0.1	1	0.4	–	–	–	–
<i>Araniella</i> spp.	12	1.4	11	4.3	9	1.6	8	2.5
<i>Mangora acalypha</i> (Walckenaer)	2	0.2	2	0.8	–	–	–	–
Araneidae unid. immatures	46	5.1	41	16.1	18	3.3	15	4.6
Subtotal	65	7.1	50	19.6	30	5.5	25	7.7
Family Clubionidae								
<i>Clubiona</i> sp.	1	0.1	1	0.4	–	–	–	–
Subtotal	1	0.1	1	0.4	–	–	–	–
Family Dictynidae								
<i>Nigma puella</i> (Simon)	20	2.2	17	6.7	–	–	–	–
Dictynidae unid. immatures	–	–	–	–	28	5.2	26	8.0
Subtotal	20	2.2	17	5.9	28	5.2	26	8.0
Family Gnaphosidae								
Gnaphosidae unid. immatures	1	0.1	1	0.4	1	0.2	1	0.3
Subtotal	1	0.1	1	0.4	1	0.2	1	0.3
Family Linyphiidae								
<i>Meioneta rurestris</i> (C.L. Koch)	–	–	–	–	1	0.2	1	0.3
<i>Pelecopsis</i> sp.	1	0.1	1	0.4	–	–	–	–
<i>Styloctetor romanus</i> (O.P. Cambridge)	–	–	–	–	1	0.2	1	0.3
Linyphiidae unid. immatures	487	53.4	125	49.0	145	26.7	85	26.2
Subtotal	488	53.5	125	49.0	147	27.1	87	26.8
Family Lycosidae								
Lycosidae unid. immatures	–	–	–	–	1	0.2	1	0.3
Subtotal	–	–	–	–	1	0.2	1	0.3
Family Miturgidae								
<i>Cheiracanthium</i> spp.	2	0.2	2	0.8	1	0.2	1	0.3
Subtotal	2	0.2	2	0.4	1	0.2	1	0.3
Family Oxyopidae								
<i>Oxyopes</i> spp.	56	6.2	37	14.5	53	9.8	38	11.7
Subtotal	56	6.2	35	13.7	53	9.8	38	11.7
Family Philodromidae								
<i>Philodromus buxi</i> Simon	1	0.1	1	0.4	–	–	–	–
<i>Philodromus praedatus</i> O. P.-Cambridge	11	1.2	9	3.5	3	0.6	3	0.9
<i>Philodromus</i> spp.	27	3.0	23	5.0	–	–	–	–
Philodromidae unid. immatures	140	15.4	72	28.2	186	34.2	102	31.4
Subtotal	179	19.7	85	33.3	189	34.8	104	32.0
Family Salticidae								
<i>Salticus scenicus</i> (Clerck)	10	1.1	7	2.8	6	1.1	4	1.2
Salticidae sp1	–	–	–	–	1	0.2	1	0.3
Salticidae unid. immatures	22	2.4	19	7.5	17	3.1	17	5.2
Subtotal	32	3.5	25	9.8	24	4.4	21	6.5

Table 2 Continued

Family/Species	2007 (n = 255)				2008 (n = 325)			
	N	Abundance %	f	O (%)	N	Abundance %	f	O (%)
Family Theridiidae								
<i>Anelosimus aulicus</i> (C.L. Koch)	1	0.1	1	0.4	–	–	–	–
<i>Phylloneta impressa</i> (C.L. Koch)	8	0.9	6	2.4	1	0.2	1	0.3
<i>Platnickina tincta</i> (Walckenaer)	–	–	–	–	1	0.2	1	0.3
Theridiidae unid. immatures	1	0.1	1	0.4	19	3.4	17	5.2
Subtotal	10	1.1	7	2.8	21	3.8	19	5.9
Family Thomisidae								
<i>Ebrechtella tricuspidata</i> (Fabricius)	1	0.1	1	0.4	–	–	–	–
<i>Runcinia grammica</i> (C.L. Koch)	8	0.9	7	2.8	–	–	–	–
<i>Synema globosum</i> (Fabricius)	19	2.1	13	5.1	29	5.3	22	6.8
<i>Xysticus</i> sp1	1	0.1	1	0.4	–	–	–	–
<i>Xysticus</i> sp2	5	0.5	5	2.0	–	–	–	–
<i>Xysticus</i> spp.	5	0.5	5	2.0	7	1.3	7	2.2
Thomisidae unid. immatures	18	2.0	13	5.1	7	1.3	6	1.9
Subtotal	57	6.2	33	12.9	43	7.9	33	10.2
Family Uloboridae								
<i>Uloborus</i> sp.	–	–	–	–	1	0.2	1	0.3
Subtotal	–	–	–	–	1	0.2	1	0.3
Non-identified	45				73			
Total	957				616			

–, zero abundance; f, number of samples where the species occurred; n, number of samples; N, total number of Araneae species; O, occurrence; sp1, sp2, two different non-identified species; unid, unidentified.

grammica (C.L. Koch), whereas *Synema globosum*, *Saliticus scenicus* and *Anyphaena sabina* C.L. Koch were the most abundant species in 2008.

The peak of total abundance of Araneae occurred from the end of June to the middle of September of both years (Fig. 1). In this period, the maximum abundances in 2007 were registered on 10 July and 28 August and in 2008 the maximum was reached on 21 August and September.

Regarding the coccinellid community, although the number of individuals captured was exactly the same in both years, the relative abundance of the subfamilies was different (Table 3). In 2007, the subfamily Coccinellinae dominated the community, followed by the subfamilies Scymninae and Coccidullinae. In 2008, the subfamily Scymninae dominated the community, followed by the subfamilies Coccinellinae and Coccidullinae. A total of 15 coccinellid species was identified in the two years of the study. *Oenopia conglobata* (L.), *Rhyzobius chrysomeloides* (Herbst), *Stethorus punctillum* Weise and *Adalia bipunctata* (L.) were the most abundant species in 2007, whereas *Scymnus interruptus* (Goeze), *S. apetzii* Mulsant, *O. conglobata* and *S. punctillum* were the most abundant species in 2008. The main peak of abundance of coccinellids occurred from the middle of July to the end of September of both years

(Fig. 1), with the maximum abundance observed on 11 September 2007 and 7 August 2008.

Ant abundance was considerably higher in 2007 with 104 individuals, than in 2008 with 31 specimens collected (Table 4). This community was distributed throughout three subfamilies, the Myrmicinae being the most abundant, followed by Formicinae and Dolichoderinae. In both years, a total of 13 ant species were collected, with 11 species collected in 2007 and eight in 2008. The subfamily Formicinae obtained the highest species richness with eight species.

In 2007, the most abundant species were *Crematogaster auberti* Emery and *C. scutellaris* (Olivier), while in 2008, *Tetramorium semilaeve* Andre and *Tapinoma nigerrimum* (Nylander) dominated the ant community.

The peaks of abundance occurred from the beginning of August to the middle of September 2007 and in August 2008 (Fig. 1). The maximum abundances were reached on 28 August 2007 and 7 August 2008.

Sampling protocol

The minimum sampling effort was calculated based on the parameters that are shown in Table 5. The rate of fauna registered varied among the three groups of natural enemies studied. In the case of Araneae families, a high

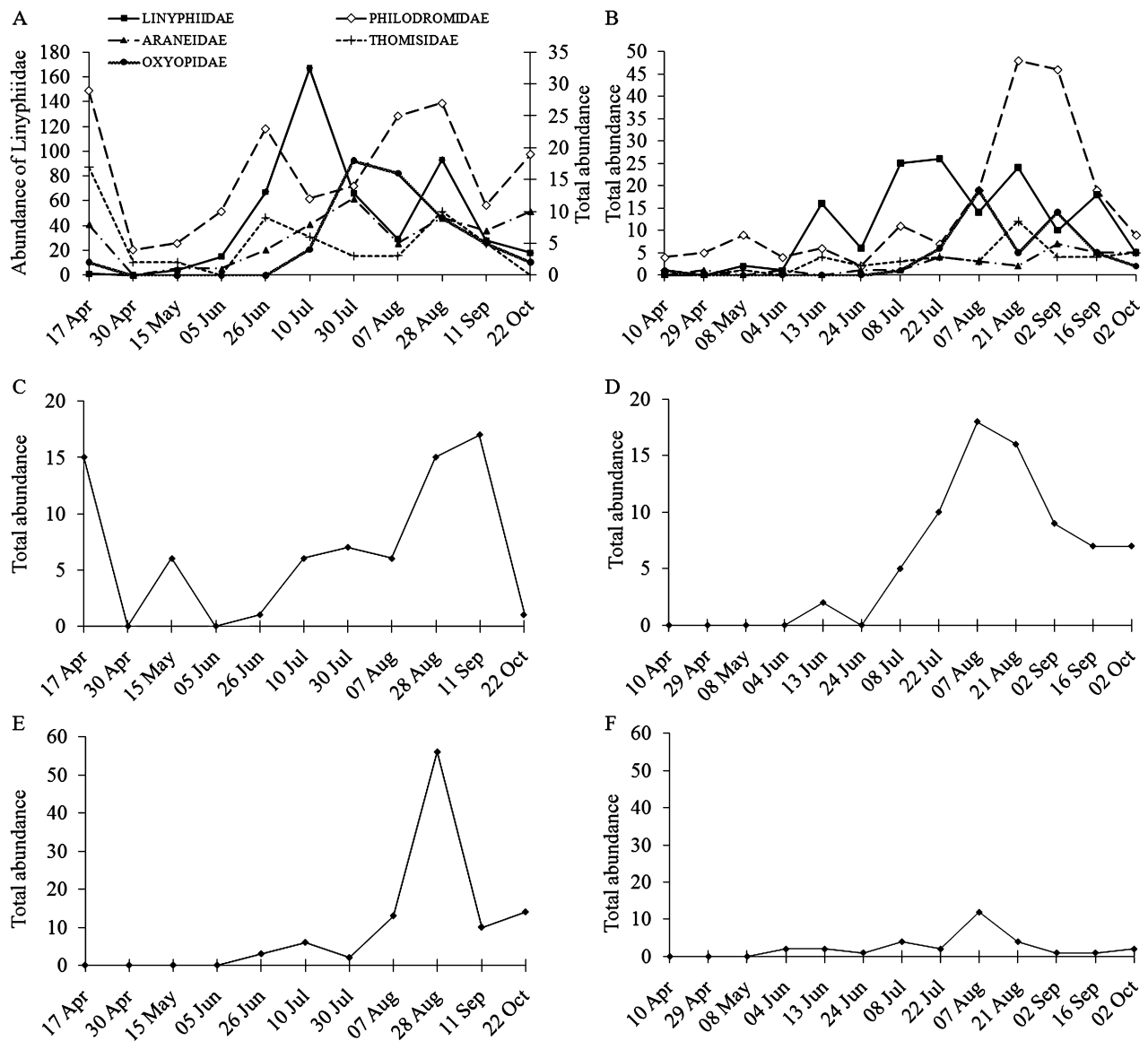


Figure 1 Total abundance of (A) Araneae in 2007, (B) Araneae in 2008 considering the five most abundant families, (C) Coccinellidae in 2007, (D) Coccinellidae in 2008, (E) Formicidae in 2007, and (F) Formicidae in 2008 collected in the almond tree canopy in each sampling date.

proportion of fauna was registered (89.73% in 2007, 79.62% in 2008); however at the species level, coccinellids and ants showed medium values for this parameter (35.48% in 2007, 67.16% in 2008 for coccinellids; 57.38% in 2007 for ants). In the case of coccinellids, extremely high values of minimum sampling effort ($n = 49$ samples) were achieved in 2008. In the other groups and years, the number of samples was between 18 (for spiders in 2007) and 23 (for ants in 2007).

The accumulation taxa curves for spiders, coccinellids and ants, which were calculated in the richest sam-

pling date, are shown in Figure 2. For spiders, accumulation taxa curves were calculated based on data collected on 10 July 2007 and 21 August 2008; for coccinellids they were calculated on 11 September 2007 and 21 August 2008; and for ants, on 28 August 2007. Due to the low abundance and richness of ants collected per sampling period in 2008, it was not possible to model the species accumulation curve. In 2007, 10 families of spiders, six species of coccinellids and five species of ants were achieved, whereas in 2008, 11 families of spiders and nine species of coccinellids were

Table 3 Total number of Coccinellidae species, percentage abundance, number of samples where the species occurred and occurrence observed in total samples collected in the almond tree canopy during the two years studied (Mogadouro, 2007–2008)

Subfamily/Species	2007 (<i>n</i> = 255)				2008 (<i>n</i> = 325)			
	<i>N</i>	%	<i>f</i>	O (%)	<i>N</i>	%	<i>f</i>	O (%)
Subfamily Chilocorinae								
<i>Exochomus nigromaculatus</i> (Goeze)	–	–	–	–	1	1.3	1	0.3
Subtotal	–	–	–	–	1	1.3	1	0.3
Subfamily Scymninae								
<i>Stethorus punctillum</i> Weise	12	16.2	11	4.0	9	12.2	9	2.8
<i>Scymnus mediterraneus</i> Iablokoff-Khnzorian	7	9.5	7	2.5	5	6.8	5	1.5
<i>Scymnus subvillosus</i> (Goeze)	–	–	–	–	5	6.8	5	1.5
<i>Scymnus interruptus</i> (Goeze)	2	2.7	2	0.7	16	21.6	15	4.6
<i>Scymnus apetzi</i> Mulsant	–	–	–	–	12	16.2	8	2.5
<i>Nephus helgae</i> Fürsch	–	–	–	–	1	1.3	1	0.3
Subtotal	21	28.4	16	6.3	48	64.9	36	11.1
Subfamily Coccidulinae								
<i>Rhyzobius chrysomeloides</i> (Herbst)	14	18.9	14	5.1	6	8.1	6	1.8
Subtotal	14	18.9	14	5.1	6	8.1	6	1.8
Subfamily Coccinellinae								
<i>Oenopia conglobata</i> (L.)	15	20.3	13	4.7	10	13.6	9	2.8
<i>Oenopia</i> sp.	4	5.4	4	1.5	–	–	–	–
<i>Adalia decempunctata</i> (L.)	1	1.4	1	0.4	–	–	–	–
<i>Adalia bipunctata</i> (L.)	9	12.2	6	2.2	6	8.1	6	1.8
<i>Adalia</i> sp.	2	2.7	2	0.7	–	–	–	–
<i>Hippodamia variegata</i> (Goeze)	7	9.5	7	2.5	1	1.3	1	0.3
<i>Coccinella septempunctata</i> L.	1	1.4	1	0.4	2	2.7	2	0.6
Subtotal	39	52.7	28	11.0	19	25.7	17	5.2
Total	74				74			

–, zero abundance; *f*, number of samples where the species occurred; *n*, number of samples; *N*, total number of Coccinellidae species; O, occurrence.

collected. Only in the case of spiders (in 2007), the family richness was very close to the asymptotic value (10 families); thus, the inventory was almost completed. In all cases, the percentage of explained variance was higher than 97%.

DISCUSSION

Abundance and richness of natural enemies

In this study, spiders dominated the community of natural enemies in the almond tree canopy where arthropods can take refuge or may be useful in biological control. Many studies have been reported from different kind of crops, showing that spiders can efficiently control pests (Riechert & Lockley 1984; Marc *et al.* 1999; Maloney *et al.* 2003; Ghavami 2008). In the two years studied, Linyphiidae, Philodromidae, Araneidae, Thomisidae and Salticidae were the most abundant families. Even considering slight differences in the relative abundance of each family, these results are consistent with those obtained by Bolu *et al.* (2008) in the

almond tree in Turkey, in other groves such as the olive tree by Cárdenas (2008) and Cárdenas *et al.* (2006) in Spain and the apple tree by Sackett *et al.* (2009) in Canada.

Although all taxa identified have a wide distribution and are common in the Iberian peninsula, it should be noted that according to Morano and Cardoso (2010) the species *Agalenatea redii* (Scopoli), *Araneus diadematus* Clerck, *Araniella opistographa* (Kulczynski), *Ebrechtella tricuspoidata* (Fabricius), *Kochiura aulica* (C.L. Koch), *Meioneta rurestris* (C.L. Koch), *P. praedatus* and *P. impressa* were recorded for the first time in the district of Bragança.

The maximum abundance of spiders appeared from mid-July to early October, which is in agreement with the results obtained by Mansour *et al.* (1983) in apple orchards in Israel. One possible reason for the differences in the peak abundances found between the two years studied could be the variations of abiotic factors such as temperature (Daiqin & Jackson 1996; Finch *et al.* 2008).

In this work, the proportion of juveniles collected was extremely high and this finding was already observed in

Table 4 Total number of Formicidae species, percentage abundance, number of samples where the species occurred and occurrence observed in total samples collected in the almond tree canopy during the two years studied (Mogadouro, 2007–2008)

Subfamily/Species	2007 (<i>n</i> = 255)				2008 (<i>n</i> = 325)			
	<i>N</i>	%	<i>f</i>	O (%)	<i>N</i>	%	<i>f</i>	O (%)
Subfamily Dolichoderinae								
<i>Tapinoma nigerrimum</i> (Nylander)	5	4.8	4	1.5	5	16.1	4	1.2
Subtotal	5	4.8	4	1.5	5	16.1	4	1.2
Subfamily Formicinae								
<i>Camponotus aethiops</i> (Latreille)	3	2.8	2	0.7	–	–	–	–
<i>Camponotus cruentatus</i> (Latreille)	1	1.0	1	0.4	–	–	–	–
<i>Camponotus foreli</i> Emery	1	1.0	1	0.4	1	3.2	1	0.3
<i>Camponotus lateralis</i> (Olivier)	1	1.0	1	0.4	–	–	–	–
<i>Camponotus truncatus</i> (Spinola)	2	1.9	1	0.4	1	3.2	1	0.3
<i>Cataglyphis hispanicus</i> (Emery)	–	–	–	–	3	9.7	3	0.9
<i>Cataglyphis ibericus</i> (Emery)	–	–	–	–	3	9.7	3	0.9
<i>Plagiolepis pygmaea</i> (Latreille)	1	1.0	1	0.4	–	–	–	–
Subtotal	9	8.7	6	2.4	8	25.8	8	2.5
Subfamily Myrmicinae								
<i>Crematogaster auberti</i> Emery	57	54.7	18	6.5	1	3.2	1	0.3
<i>Crematogaster scutellaris</i> (Olivier)	31	29.8	4	1.5	1	3.2	1	0.3
<i>Leptothorax angustulus</i> (Nylander)	1	1.0	1	0.4	–	–	–	–
<i>Tetramorium semilaeve</i> Andre	1	1.0	1	0.4	16	51.7	8	2.5
Subtotal	90	86.5	23	9.0	18	58.1	10	3.1
Total	104				31			

–, zero abundance; *f*, number of samples where the species occurred; *n*, number of samples; *N*, total number of Formicidae species; O, occurrence.

Table 5 Characterization of the inventory for Araneae, Coccinellidae and Formicidae collected in the almond tree canopy in two consecutive years (Mogadouro, 2007–2008)

	Araneae		Coccinellidae		Formicidae
	2007	2008	2007	2008	2007
Parameter <i>a</i>	2.4778	2.0097	0.6728	0.6417	0.4072
Parameter <i>b</i>	0.2223	0.1454	0.0716	0.0310	0.0430
Percentage of registered fauna (F)	89.73	79.62	63.85	43.49	52.08
Quality of the inventory (<i>C_i</i>)	0.00	0.00	0.01	0.01	0.10
Percentage of explained variance (<i>R</i> ²)	97.27	98.89	99.98	99.99	99.82
Number of samples (<i>n</i>) for <i>C_i</i> = 0.1	18	23	22	49	23

different works developed in other ecosystems (Coddington *et al.* 1996; Cardoso *et al.* 2004; Jiménez-Valverde & Lobo 2006). The occurrence of the high number of juveniles can be related to specific characteristics of the biology of spiders: (i) the two biological peaks that usually occur during the year (spring and autumn peaks) cause an overlap of generations of spiderlings; (ii) depending on the circumstance and habitat, some spiders need 18 months to reach the adult stage, which implies spending a complete year in juvenile stages (Morse 2007; Cárdenas 2008); and (iii) the maternal care spent by many spider species (mainly belonging to Araneidae and Pisauridae families), which requires the agglomeration of the eggs in special web sacs. In the

last case, prior to dispersion, the juveniles usually molt at least once (Turnbull 1973) and the beating technique may involve the collection of the whole nest, thus greatly increasing the abundance of juveniles in samples collected from the canopy.

Predaceous coccinellid species were also present in the almond tree and both abundance and community composition may be influenced by prey availability, since they are specialists in a range of preys (Honěk 1985). *Stethorus punctillum*, for instance, is a specialist on spider mites from the family Tetranychidae and its importance as predator of *T. urticae* has been documented in Australia (Biddinger *et al.* 2009). Therefore, it is likely that the abundance of this coccinellid species in

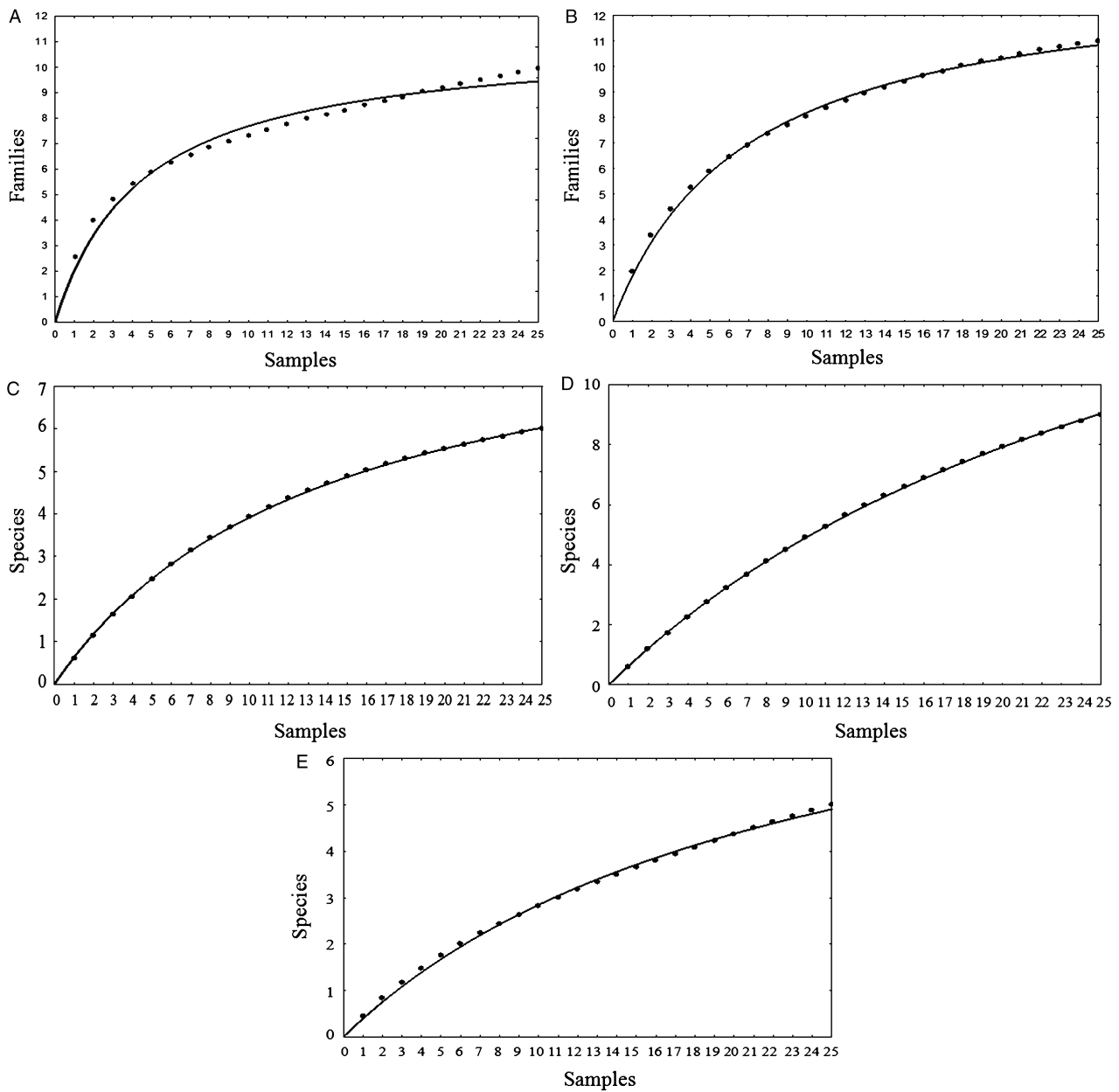


Figure 2 Species accumulation curves for each studied group: (A) Araneae in 2007, (B) Araneae in 2008, (C) Coccinellidae in 2007, (D) Coccinellidae in 2008, and (E) Formicidae in 2007. Each point represents the mean of 50 randomizations. Solid lines represent the estimated species accumulation.

the almond tree could be due to the presence of their prey on the leaves. Other aphidophagous species such as *Oenopia* spp., *Adalia* spp. and *Hippodamia variegata* (Goeze), and polyphagous species such as *R. chrysomeloides* and *S. mediterraneus* Iablokoff-Khnzorian could also be considered important for this crop. These are widespread species that were already found in many other crops (Biddinger *et al.* 2009; Giorgi *et al.* 2009).

Similarly to what was described for other agroecosystems, in the almond grove ants can play major ecological roles as predators, scavengers, mutualists and ecosystem engineers acting within the soil–nutrient and enrichment cycle (Redolfi *et al.* 1999; Pereira *et al.* 2002; Santos *et al.* 2007; Ivanov & Keiper 2009). The four most abundant ant species, *C. auberti*, *C. scutellaris*, *T. semilaeve* and *T. nigerrimum*, were already

found in other agroecosystems such as olive groves (Redolfi *et al.* 1999; Pereira *et al.* 2002; Santos *et al.* 2007) and citrus groves (Zina 2008). As generalist predators, ants are known to consume not only a broad range of arthropod prey but are also among the main consumers of honeydew secreted by aphids (Pereira *et al.* 2002), which could be the main reason for their presence in almond trees.

The other groups sampled in the almond tree canopy include Neuroptera, Heteroptera (mainly anthocorids) and Dermaptera, which can comprise generalist predators. These groups were found in less abundance but they can help the natural control of the main almond pests in the region.

Beyond the presence of potential prey in the almond tree canopy, the relative abundances of natural enemies found in this study can also be due to the existence of extrafloral nectar secretions that are common in *Prunus* spp. (e.g. almond, peach and cherry trees). These secretions can attract some spider species such as philodromids (Brown *et al.* 2003), ants (Limburg & Rosenheim 2001) and coccinellids (Lundgren 2009), which have been found to feed on them. Extrafloral nectar can provide supplemental food to natural enemies, thus enhancing their survival, fecundity and longevity (Jervis & Heimpel 2005).

Sampling protocol

The beating technique as a sampling method used in this study to collect arthropods was effective in obtaining good representations of the most abundant groups. In particular, this sampling technique was efficient for the spider families Linyphiidae, Philodromidae, Thomisidae and Araneidae. In the last two families, our results are in agreement with the results obtained by Jiménez-Valverde and Lobo (2006). However, the biology and distribution of the species in the field can bring several constraints to the optimization of a sampling protocol. For instance, the lowest density of individuals of Clubionidae, Anyphaenidae and Miturgidae collected in the two years of study using this technique may be due to their nocturnal hunting habits (Bristowe 1958; Jones 2004). Because of this, it would be advisable to use the combination of at least two sampling periods when the study aims to survey the biodiversity of spiders, one in the morning and the other in the evening, to avoid underestimation of the abundance of nocturnal species. On the other hand, due to the social way of life of ants, they are probably non-randomly distributed in the grove (Ivanov & Keiper 2009) and the abundance of each species could be influenced by the proximity of the nest, their aggression against other species and competition

for food. These characteristics should be considered when the abundance of ants is surveyed.

According to Jiménez-Valverde and Lobo (2006), the required field sampling effort should be optimized considering both the reduction of samples, and the reduction of seasonal coverage of samplings that would be limited to the richest season. In the first case, and based on results obtained using the accumulation curves for different groups (Fig. 2), it can be estimated that 18 to 22 samples would be sufficient to achieve a satisfactory quality inventory using the beating technique in the almond tree. However, reducing the sample size may be feasible for obtaining a rapid as well as a satisfactory representation of the most abundant groups sampled in this crop. As a result, the number of samples might be reduced to 10 or 11 per sampling date in studies with minimal sampling programs. Considering the reduction of the seasonal coverage of samplings and based on the annual richness distributions of Araneae, Coccinellidae and Formicidae in the almond grove studied, the optimal sampling period can be set from the beginning of July to the end of September. However, it is important to note that under Mediterranean conditions with very hot and dry summers, almond groves are usually non-irrigated and pest populations (mainly *T. urticae* and *M. uniconstata*) reach higher levels in August and September. To minimize the effects of dryness and pest damage, harvesting can start in the beginning of September, causing a reduction of the leaf coverage and, as a consequence, of the abundance of natural enemies. Because of that, the seasonal sampling period should be adjusted according to the management practices adopted by farmers in each Mediterranean country.

In conclusion, the knowledge of abundance and richness of natural enemies and their seasonality could help growers develop management plans to increase their action and maintain their population in the orchards. Moreover, the establishment of a sampling protocol for the almond tree canopy can minimize resource consumption while giving an accurate picture of biodiversity. As a result, 11 samples collected on a fortnightly basis from the beginning of July to the harvesting of almonds might be a useful method for exploring the patterns of biodiversity within this crop and understanding the effects of agroecosystem management in a community of natural enemies.

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