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Pollen supply promotes, but high temperatures demote, predatory mite abundance in avocado orchards

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ABSTRACT

A biological control strategy aimed at enhancing the performance in the field of two predatory mites that are natural enemies of an invasive pest of avocado orchards was applied during two consecutive years. Unexpected poor pest control led us to analyse additional factors involved in the dynamics of the community such as summer environmental conditions, which were very hot and dry. Non-linear regressions considering biotic and abiotic effects suggested that high temperatures during the second half of the summer were central to predator/prey population dynamics. Harsh environmental conditions were therefore a direct cause of biological control disruption. We believe that, in the future, the success of biological control strategies will be limited by the natural enemy's capacity to respond adaptatively to rapid climate changes, and that research aimed at evaluating the evolutionary potential of natural enemies to rapid climate change should be the focus of future investigations.

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1. Introduction

Several studies have documented recent vertebrate, invertebrate, and plant range shifts that are associated with global warming (Letcher, 2009). However, fewer studies have addressed the potential effects of climate change on the relationships within communities such as those represented in biological pest control. Scenarios where organisms will be increasingly exposed to extreme high temperatures may become relevant for the future of biological pest control, as most of the natural enemies presently used as biocontrol agents are arthropods, in which many of the key processes related to their survival, reproduction and other fitness-related parameters depend on environmental conditions (Beveridge et al., 2010). Furthermore, sensitivity to temperature increase intensifies with trophic level (Voigt et al., 2003), a bottom-up amplification effect that often occurs to trophic webs subjected to disturbances of any kind (Cagnolo et al., 2002). Therefore, natural enemy efficiency or life-history negatively affected by environmental warming may dampen trophic cascades and profoundly affect pest control. Indeed, recent works have attributed herbivore control disruption in some agro-ecosystems to severe environmental conditions (Montserrat et al., 2012; Stavrinides et al., 2010).

In the coastal areas of the provinces of Malaga and Granada, in the region of Andalusia (Spain), avocado (*Persea americana* Mill., Lauraceae) is the third most abundant non-citrus fruit tree, after almond and olives (MARM, 2011). Avocado orchards in Andalusia had been free from most of the pests present in other avocado producing areas in the world until the detection, in 2004, of the persea mite, *Oligonychus perseae* Tuttle, Baker and Abatiello (Acari: Tetranychidae) (Vela et al., 2007). All the developmental stages of this tetranychid species inhabit dense silken nests built at the underside of avocado leaves, and their feeding activity causes necrotic spots on leaves. Nests protect mites against attack from some species of natural enemies, and also against adverse environmental conditions (Montserrat et al., 2008). In these coastal avocado orchards two species of phytoseiid mites have been found co-occurring with this herbivorous mite (González-Fernández et al., 2009): *Euseius stipulatus* (Athias-Henriot), an omnivore that attacks animal prey and also forages on pollen (Ferragut et al., 1987); and *Neoseiulus californicus* (McGregor), a specialist predator of tetranychid mites. Both species prey on the persea mite (González-Fernández et al., 2009; Montserrat et al., 2008) and are strong candidates to be considered in biological control programs against this pest, although only *N. californicus* is commercially available.

The population dynamics of phytoseiid mites in avocado trees in Spain typically show two population maxima, the first in spring and the second in summer. In spring, when the populations of persea mites are still small, phytoseiid populations increase as individuals feed on pollen deposited on the surface of leaves, whereas, in summer, phytoseiid populations respond numerically to the abundance of the persea mite (González-Fernández et al., 2009; Montserrat et al., 2012). Based on the mite community dynamics,

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a strategy to control this pest consisting of making maize pollen available in avocado orchards, which was released from intercropped maize plants, was recently tested (González-Fernández et al., 2009). Results obtained were very encouraging, as those trees that were closer to the maize pollen sources harboured more phytoseiid and less herbivore mites (González-Fernández et al., 2009). However, differences between treatments diluted with time because maize plants ceased to release pollen.

This study was aimed at enhancing the performance in the field of the two predatory mites, *E. stipulatus* and *N. californicus*, which behave as natural enemies of the persea mite. In the case of the non-commercial omnivore *E. stipulatus*, it was intended to indirectly increase its populations by prolonging the presence of pollen in the system through direct supply of stored pollen to the trees, at the beginning of summer. Enhancement of *N. californicus* performance would be achieved through releases of these mites during mid-summer, when populations of the persea mite start to increase. It was expected that the combination of both predator species supplied with an alternative food source would intensify the effects observed in a previous work (González-Fernández et al., 2009). However, due to unexpected results, we also attempted to identify whether environmental conditions in summer, which were very hot and dry, could have been an important driver of the observed dynamics of the community.

2. Methods

Cultures of the predatory mite *E. stipulatus* started in 2007 from ca. 300 individuals collected from avocado trees located in the area of study. *Euseius stipulatus* was cultured in a climate chamber at 25 ± 1 °C, 65 ± 5% HR and 16:8 L:D. Rearing units consisted of sponges (30 cm × 20 cm × 5 cm, approx.) covered with cotton wool, with a plastic sheet (27 cm × 17 cm, approx.) on top, and placed inside water-containing trays (8 l, 42.5 cm × 26 cm × 7.5 cm). Three bean (*Phaseolus vulgaris*) plants (6–10 leaves old) were positioned vertically with the stems in contact with three of the walls of the sponge, the roots in contact with the water of the trays, and the aerial parts touching each other forming a tent-like three-dimensional structure, where mites could easily walk from one plant to the other. Some of the leaves contained cotton threads that served as oviposition sites for the mites. Mites were fed ad libitum twice a week with pollen of *Carpobrotus edulis* that was spread on leaves with a fine brush. When required, new rearings were made either by transferring the cotton threads filled with eggs or by transferring adult females (ca. 150), to a new unit. Pollen of *C. edulis* was obtained from male flowers that were dried in a stove at 37 °C for 48 h, and then sieved (350 μm). *Neoseiulus californicus* used in the field experiments were kindly supplied by Koppert Biological Systems, The Netherlands.

2.1. Commercial pollen as alternative food

The aim of this experiment was to determine if commercial bee pollen dissolved in water is a suitable food source for the omnivore mite, *E. stipulatus*. Commercial bee pollen was chosen because it is inexpensive and easily available. Commercial pollen in pellet form was obtained from a local shop, and comprised 60–80% *Cistus* spp. (Cistaceae) pollen; the remaining pollen was from various species of Boraginaceae, Rosaceae and Fabaceae.

Experiments were carried out in a climate chamber (600 l) at 25 ± 1 °C, 65 ± 5% HR and 16:8 L:D. The experimental set-up consisted of plastic arenas (3.5 cm Ø) placed on a layer of water-soaked cotton wool inside plastic containers (100 ml, 6.7 cm high, lower diameter 5.1 cm, upper 6.5 cm). The experimental arenas were allocated to the following treatments: (i) arenas supplied with commercial bee pollen ($N = 12$): 40 (approx.) pellets of commercial bee pollen were dissolved in 100 ml of tap water, and one drop of the solution was applied on the experimental arenas; (ii) arenas supplied with maize pollen ($N = 10$) as positive control, because, in previous field experiments (González-Fernández et al., 2009), predatory mites showed a numerical response to the presence of maize pollen that resulted in a decrease on the abundance of the pest; and iii) arenas with no food, as a negative control ($N = 12$). One gravid *E. stipulatus* female (10–15 d old since egg stage) was placed in each of the arenas using a fine brush. After 24, 48, and 78 h the number of eggs laid by the female was counted. To ensure that only the effect of the treatment was measured, only the average number of eggs laid during the second and third day was used in the analyses. The number of eggs/*d* was analysed with a one-factor ANOVA, with type of pollen as main factor. Means were separated with the Tukey Unequal N HSD test.

2.2. Pollen application and predator release

This experiment was designed to evaluate whether the combination of alternative food supply at the beginning of summer with artificial releases of *N. californicus* in mid-summer would result in a better control of the persea mite, *O. perseae*, through higher total numbers of predatory mites.

Preliminary experiments carried out in the laboratory revealed that, on the one hand, patches of commercial pollen were easily created on the leaves when avocado leaves were sprayed with 2 g/l of commercial pollen dissolved in water, from a distance of 1.5 m (Fig. A, supplementary material). On the other hand, patches contained pollen consumable by mites – who pierce individual grains with their mouthparts and suck their contents – as most of the sprayed pollen grains did not explode (Fig. B, supplementary material).

Field experiments were carried out in a 2 ha 'Hass' avocado orchard located at the IHSM La Mayora (Malaga, Spain) during two consecutive years (2009 and 2010) from the beginning of June to the end of September. The experimental field was divided East-to-West into five blocks, each containing five groups of three adjacent trees, which were separated by several additional avocado trees. Each of the groups per block was randomly assigned to one of the following treatments: (a) Pollen: 14 g of pollen (acquired at the same shop and from the same stock as the one described above) dissolved on 7 l of water were applied to the tree canopy using a spray gun trolley equipped with a piston pump; (b) water: 7 l of water were applied to the canopies using the same device as in (a), as control treatment of pollen application; (c) *N. californicus* release: 1000–3500 individuals of *N. californicus* were released per tree (see below); (d) Pollen + *N. californicus*: Trees were supplied with the same amount of pollen and number of *N. californicus* as in the treatments with either only pollen supply (a) or only predator release (c); (e) control: trees with no treatment applied. For each treatment, only the tree located in the middle of the group of three trees was used for samplings.

Prior to the start of the experiments, the abundance of phytoseiids and persea mites was assessed in May, when pollen counts are at their maximum, to ensure that phytoseiid abundance was high, and herbivore abundance low, as expected. At the end of May of 2009 there was an average of 1.16 ± 0.08 phytoseiid mites/leaf, and 0.048 ± 0.017 nests of persea mites/leaf. In 2010 these values averaged 1.42 ± 0.99 and 0.336 ± 0.172, respectively. Because abundance of pollen starts decreasing at the end of May, the first treatments involving pollen applications and the control were done within the first week of June. Subsequent treatments with pollen were decided upon actual abundances of phytoseiids recorded in the field. As a rule of thumb, the treatments were made when populations of predators decreased considerably during two consecutive

sampling dates. In 2009 and 2010 there was one additional pollen application in June, whereas in 2010 another application was made in July. Predatory mites were released in the field by means of paper bags containing ca. 250 individuals that were hung from the branches of trees. Decisions on releasing times were made upon actual densities of persea mites in the field. In 2009, we released ca. 1000 individuals per tree at the end of June. Based on sub-optimal results obtained in 2009, we decided to release higher amounts in 2010, and perform two applications in different dates: ca. 1500 individuals in the second week of June, and ca. 3500 individuals three weeks later.

In each of the trees, the number of occupied nests of *O. perseae* on the upper margin of the second left vein of the underside of the leaf, UML2 hereafter, and the number of phytoseiid mites per leaf, were counted in ten leaves at ca. 150 cm height. In order to account for within-tree abundance variability, leaves were selected so that the whole perimeter of the canopy was covered. Counting was done in situ using a field magnifying glass (Ruper 8×). Monitoring was made every two weeks, starting the first week of June and ending the last week of September. The two species of predatory mites were not identified at each sampling date because *E. stipulatus* and *N. californicus* are morphologically very similar, and they can only be differentiated at a microscopic level.

The average number of nests (prey, hereafter) in the UML2 and the average number of phytoseiids (predators, hereafter) per leaf and tree ($N = 10$ leaves) was calculated for each sampling date of each year. Cumulative averages of predators and prey per tree and year were fitted to a non-linear logistic regression function, defined as:

$$N(t) = \frac{kN_0}{N_0 + (k - N_0)e^{-rt}}$$

where $N(t)$ was the cumulative number of predators or prey/leaf; k was defined as the maximum cumulative population size/leaf/tree of either predators or prey; r was the cumulative growth rate of each population; t was time (d); and N_0 was the initial number of either predators or prey, which was set to 0.1 and 0.001 for predators and prey, respectively, because samplings started at the beginning of June, when phytoseiid mites are present whereas abundance of the persea mite is negligible.

The inflexion point of the logistic curve, i.e. the time when the cumulative growth rate switches from positive to negative, was calculated as:

$$t_i = \frac{\ln(k - N_0/N_0)}{r}$$

There were, therefore, five estimated values of k, r and t_i of predators and prey, per treatment and year. The k-value was used as proxy for total population abundance of predators and prey throughout the season. The r-value was interpreted as how fast populations reached its upper cumulative limit along the season. The t_i-value was used as proxy for the time when the contribution of predator and prey to the cumulative numbers was at its maximum (the contribution to the cumulative number at time $t > t_i$ is smaller than that at $t < t_i$). The k, r, and t_i values were analysed with three-factor MANOVA, with year (2009 and 2010), species (predator or prey) and treatment (five treatments), as explanatory variables. When the MANOVAs were significant, each of the dependent variables was analysed separately with three-factor ANOVA to determine which one was causing the effect.

2.3. Biotic and abiotic effects on population dynamics

To determine whether environmental conditions had an effect on the population dynamics of predators and prey, we first calculated, for each tree and year, the so-called "interval growth rate" ($r_{i:t,t+\tau}$) of prey and predators, using the equation:

$$r_{t,t+\tau} = \frac{1}{\tau} \ln\left(\frac{N_{t+\tau}}{N_t}\right)$$

where τ is the time interval (d) between the first and the second sampling event and N_t and $N_{t+\tau}$ are the populations size at time t and $t+\tau$, respectively. Next, the interval growth rate/tree of predator and prey were averaged ($N = 25$) to obtain one single value of $r_{t,t+\tau}$ per interval. Daily records on average temperature and relative humidity at a height of ca. 2 m above the ground were obtained from an agro-climate station that belongs to the Government of Andalusia, located close to the area of study (ca. 15 km linear distance). For each single value of $r_{t,t+\tau}$ concurrent values of temperature and humidity were obtained averaging the daily records of temperature and relative humidity during the interval $(t,t+\tau)$. The variation of the average interval per capita growth rate of predators and prey during the sampling period was analysed by non-linear regression models (Proc NLR, SPSS Inc.). The predictor variables used in the analyses were the first and second order terms of (a) the interval average temperature ($T_{t,t+\tau}$), and (b) the interval average relative humidity ($RH_{t,t+\tau}$). First and second order terms were included because temperature and relative humidity usually have non-linear effects on life history of arthropods; (c) the interval average of the abundance of prey ($Prey_{t,t+\tau}$), to account for predation or for intra-specific competition when modelling the per capita growth rate of predators and prey, respectively; and (d) the interval average of the abundance of predators ($Pred_{t,t+\tau}$), to account for predation or for intra-specific competition when modelling the per capita growth rate of prey and predators, respectively. First, only biotic effects, i.e. predation, intra-specific competition, and their interaction, were included in the analyses to test how much of the variation in $r_{t,t+\tau}$ they could explain. Second, only abiotic effects, i.e. temperature, relative humidity, and their interaction, were included in the analyses; and last, the whole models including biotic and abiotic variables were analysed. All regression analyses were done with backwards stepwise procedure.

3. Results

3.1. Commercial pollen as alternative food

The number of eggs laid by predatory mites differed among treatments ($F_{2,31} = 25.63$, $P < 0.001$). Tukey Unequal N HSD post hoc tests revealed that oviposition rates of predators feeding on commercial bee pollen (0.96 eggs/d) were not statistically different from those feeding on maize pollen (1.15 eggs/d) ($P = 0.58$). Both treatments were significantly different ($P < 0.001$) to the control, where no food was provided and females did not oviposit.

3.2. Pollen application and predator release

The MANOVA was significant only for the main factors Year and Species and for their interaction (Table 1). When k, r, and t_i were analysed separately to determine which variable was causing the effect, results revealed, on the one hand, that the maximum cumulative number of both predators and prey at the end of the season did not differ among treatments, although numbers of prey were much lower in 2010 than in 2009 (Table 2 – results for k). On the other hand, results revealed that predators reached their cumulative population maximum much earlier than prey (Table 2-results for t_i), i.e. 40.02 ± 1.57 and 70.06 ± 1.23 d after the beginning of the experiment, respectively. Last, results revealed that the cumulative growth rate of predators and prey had opposite trends depending on the treatment (Table 2, results for r, interaction term "species × treatment"). The graphical analysis of the interaction

Table 1
Results of three-factor MANOVA, with k (maximum cumulative population size), r (cumulative growth rate of populations), and t_i (time of maximum contribution to the cumulative number) as dependent variables, and year (2009 and 2010), species (predator or prey) and treatment (five treatments), as explanatory variables.

Effect	Wilks λ	F	d.f.	P
Intercept	0.0047	5546.81	3; 78	<0.0001
Year (1)	0.4747	28.79	3; 78	<0.0001
Species (2)	0.1418	157.31	3; 78	<0.0001
Treatment (3)	0.8671	0.95	12; 206.676	n.s.
(1) × (2)	0.7538	8.49	3; 78	<0.0001
(1) × (3)	0.9218	0.54	12; 206.676	n.s.
(2) × (3)	0.8369	1.20	12; 206.676	n.s.
(1) × (2) × (3)	0.8975	0.72	12; 206.676	n.s.

revealed that r was higher for predators, and lower for prey, when pollen was available, compared to that in the other treatments (Fig. 1).

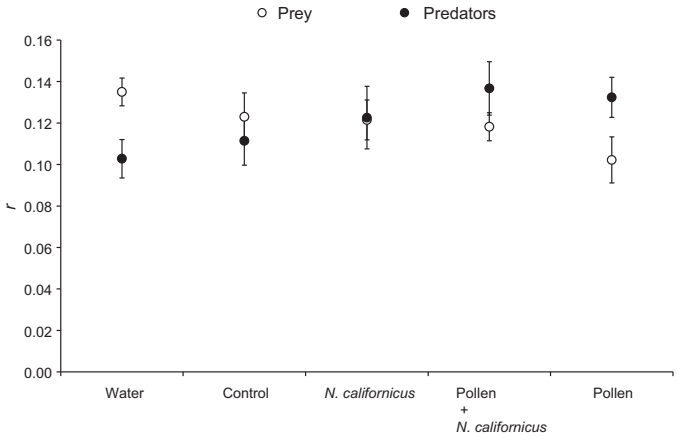

Fig. 1. Cumulative growth rate (r) of predators and prey, depending on treatment.

3.3. Biotic and abiotic effects on population dynamics

When only biotic effects were included in the regression analyses, only the abundance of prey significantly explained the variation of the average interval per capita growth rate of both prey and predators (Tables 3 and 4). The sign of the parameter estimate revealed that intra-specific competition had a negative effect on the growth rate of prey.

When only abiotic effects, and when all the effects, were included in the analyses, only the first and second order term of temperature significantly explained the variation of the average interval per capita growth rate in both prey and predators (Tables 3 and 4). Solving the equations of the regression analyses revealed that predator interval growth rate would be negative when the interval temperature is higher than 24.7 °C, whereas for the prey the interval temperature has to be higher than 25.6 °C. Fig. 2 shows that when the two-week interval average temperature was below these values, i.e. during the first half of summer, phytoseiid mites were abundant in the field and the population of the persea mite increased. However, when the two-week interval average temperatures were above their respective critical values, both predator and prey populations started to decrease dramatically.

4. Discussion

4.1. Pollen promoting predator abundance

Our results suggest that predatory mites responded numerically to the supply of pollen to the trees. This response was detected through a faster cumulative growth rate of predators inhabiting trees that were supplied with pollen (Fig. 1), indicating that the contribution of predator numbers at time $t+1$ to the cumulative number at time t was higher in trees with pollen applications than in trees with no pollen applications. Our results also suggest that such predator numerical response had an opposite effect on the cumulative growth rate of the persea mite populations (Fig. 1).

The use of an alternative food to improve pest control is aimed, on the one hand, at providing food for predators to promote the persistence of natural enemies in the field when pest populations are

Table 2
Results of the three univariate three-factor ANOVA, with either k (maximum cumulative population size), r (cumulative growth rate of populations), and t_i (time of maximum contribution to the cumulative number) as dependent variables, and year (2009 and 2010), species (predator or prey) and treatment (five treatments), as explanatory variables.

Dependent variables	R^2 model	F model	P model	d.f.	Predictor variables	F	P
k	0.691	9.407	<0.001	19; 80	Intercept	899.71	<0.001
					Year (1)	13.47	<0.001
					Species (2)	133.88	<0.001
					Treatment (3)	0.83	n.s.
					(1) × (2)	26.09	<0.001
					(1) × (3)	0.32	n.s.
					(2) × (3)	0.09	n.s.
					(1) × (2) × (3)	0.08	n.s.
r	0.325	2.027	0.016	19; 80	Intercept	1471.97	<0.001
					Year (1)	15.87	<0.001
					Species (2)	0.04	n.s.
					Treatment (3)	0.38	n.s.
					(1) × (2)	0.28	n.s.
					(1) × (3)	0.70	n.s.
					(2) × (3)	3.06	0.021
					(1) × (2) × (3)	1.44	n.s.
t_i	0.691	9.430	<0.001	19; 80	Intercept	3377.62	<0.001
					Year (1)	5.30	0.024
					Species (2)	159.96	<0.001
					Treatment (3)	1.24	
					(1) × (2)	0.003	
					(1) × (3)	0.95	
					(2) × (3)	0.89	
					(1) × (2) × (3)	0.39	

Table 3
Results of non-linear regression models on the biotic and/or abiotic effects on the variation of the average interval per capita growth rate of **prey**. First, only biotic variables and their interaction were included in the analyses; second, only abiotic variables and their interaction were included in the analyses; and last, the whole model including biotic and abiotic variables was analysed. All regression analyses were done with backwards stepwise procedure.

Model	R^2 model	F model	P model	d.f.	Predictor variables	Parameter estimates	F/t	P
Only biotic effects	0.420	6.148	0.0098	2; 17	Intercept	0.039	2.548	0.021
					$Prey_{t,t+\tau}$ (1)	−0.087	−2.449	0.025
					$Predators_{t,t+\tau}$ (2)	Not included		
					(1) × (2)	0.070	2.173	0.044
Only abiotic effects	0.441	6.696	0.0072	2; 17	Intercept	−2.368	−2.142	0.047
					$T_{t,t+\tau}$	0.223	2.307	0.034
					$T_{t,t+\tau}{}^2$	−0.0051	−2.443	0.026
					$RH_{t,t+\tau}$	Not included		
					$RH_{t,t+\tau}{}^2$	Not included		
					$T_{t,t+\tau} \times RH_{t,t+\tau}$	Not included		
Whole model effects	0.441	6.696	0.0072	2; 17	Intercept	−2.368	−2.142	0.047
					$T_{t,t+\tau}$	0.223	2.307	0.034
					$T_{t,t+\tau}{}^2$	−0.0051	−2.443	0.026
					$RH_{t,t+\tau}$	Not included		
					$RH_{t,t+\tau}{}^2$	Not included		
					$T_{t,t+\tau} \times RH_{t,t+\tau}$	Not included		
					$Prey_{t,t+\tau}$ (1)	Not included		
					$Predators_{t,t+\tau}$ (2)	Not included		
					(1) × (2)	Not included		

still scarce. Persistence of predators due to the presence of alternative food when the target prey was not abundant has been studied in predatory mites (Nomikou et al., 2002; Pozzebon et al., 2009), insect predators such as bugs or beetles (Berkvens et al., 2008; Frank et al., 2011; Lucas et al., 2009; Sanchez et al., 2003), parasitoids (Vollhardt et al., 2010), and spiders (Kuusk and Ekbom, 2010). On the other hand, co-occurrence of alternative food and the target pest may promote predator abundance and pest control through apparent competition (Holt, 1977), as has been shown in some studies (Aguilar-Fenollosa et al., 2011; González-Fernández et al., 2009; Maoz et al., 2011; Messelink et al., 2008, 2010; Nomikou et al., 2010). However, no effect (Brown and Mathews, 2008) or a negative effect (Desneux and O'Neil, 2008; Musser and Shelton, 2003; Spellman et al., 2006) of alternative food/prey on biological pest control have also been reported. Additionally, when natural enemies can potentially engage in trophic interactions, the presence of alternative food may reduce the strength of interactions such as intraguild predation or cannibalism.

In our system, the numerical effect of the presence of pollen on predators occurred only during the first half of summer and did not translate into an increase of the total population abundance of predators throughout the season, nor did it in a better control of the persea mite populations. Indeed, the maximum cumulative number of both predators and prey at the end of the season did not differ among treatments in either of the two years (Table 2, results for k). Also, the maximum contribution of predator abundance to the cumulative population number occurred much earlier than that of prey ($t_i = 40.02 \pm 1.57$ and 70.06 ± 1.23 d after the beginning of the experiment, respectively), indicating that predator population did not increase when the target prey was present, i.e. during the second half of summer. This result contradicted previous observations of predators responding numerically to the abundance of the persea mite (González-Fernández et al., 2009), suggesting that, in these two years, factors other than the presence of alternative food, or the abundance of the prey, were probably driving predator-prey community dynamics.

Table 4
Results of the non-linear regression models on the biotic and/or abiotic effects on the variation of the average interval per capita growth rate of **predators**. First, only biotic variables and their interaction were included in the analyses; second, only abiotic variables and their interaction were included in the analyses; and last, the whole model including biotic and abiotic variables was analysed. All regression analyses were done with backwards stepwise procedure.

Model	R^2 model	F model	P model	d.f.	Predictor variables	Parameter estimates	F/t	P
Only biotic effects	0.255	6.152	0.023	1; 18	Intercept	0.039	2.548	0.021
					$Prey_{t,t+\tau}$ (1)	−0.0057	−2.480	0.023
					$Predators_{t,t+\tau}$ (2)	Not included		
					(1) × (2)	Not included		
Only abiotic effects	0.576	11.548	0.0007	2; 17	Intercept	−1.040	−1.932	0.070
					$T_{t,t+\tau}$	0.1014	2.153	0.046
					$T_{t,t+\tau}{}^2$	−0.0024	−2.361	0.030
					$RH_{t,t+\tau}$	Not included		
					$RH_{t,t+\tau}{}^2$	Not included		
					$T_{t,t+\tau} \times RH_{t,t+\tau}$	Not included		
Whole model effects	0.576	11.548	0.0007	2; 17	Intercept	−1.040	−1.932	0.070
					$T_{t,t+\tau}$	0.1014	2.153	0.046
					$T_{t,t+\tau}{}^2$	−0.0024	−2.361	0.030
					$RH_{t,t+\tau}$	Not included		
					$RH_{t,t+\tau}{}^2$	Not included		
					$T_{t,t+\tau} \times RH_{t,t+\tau}$	Not included		
					$Prey_{t,t+\tau}$ (1)	Not included		
					$Predators_{t,t+\tau}$ (2)	Not included		
					(1) × (2)	Not included		

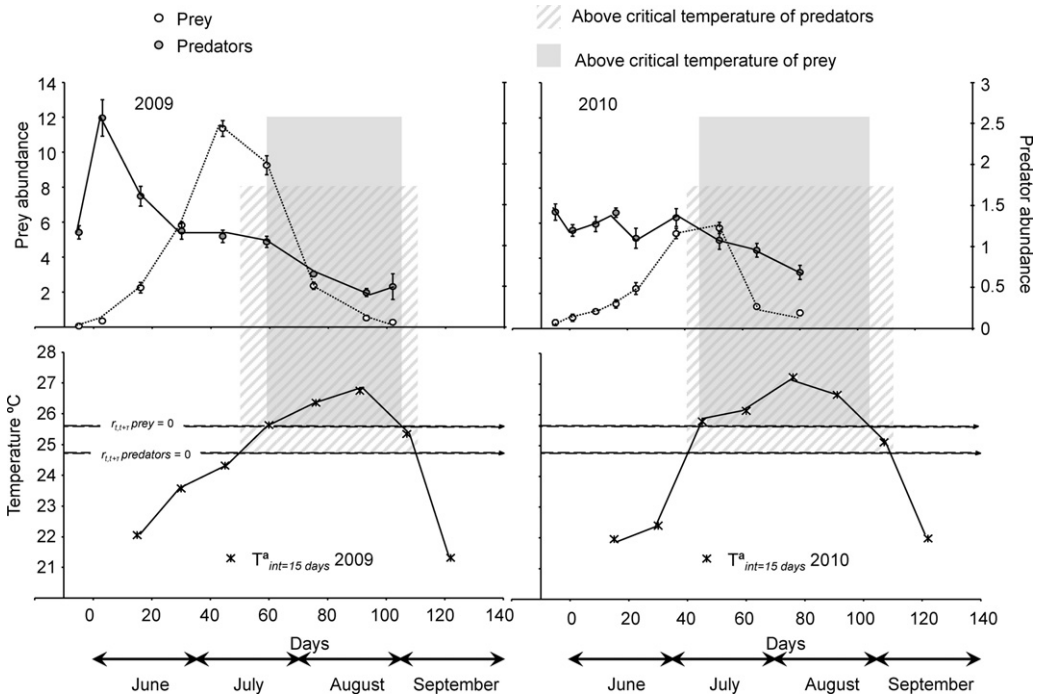

Fig. 2. Dynamics of predators (●) and prey (○) during 2009 (left panel) and 2010 (right panel). Interval average temperatures for the same periods are also depicted (lower panels). Horizontal lines in lower panels represent the critical temperature value (i.e. value above which population growth rate is expected to be negative), estimated from the non-linear regressions. Grey areas in above panels represent periods of time in which interval average temperatures were above critic values for predators (□) and prey (▤).

4.2. Climate demoting predator abundance

The analysis of the effect of environmental conditions on predator and prey population growth rates suggested that high temperatures during the second half of summer were central to predator/prey population dynamics. Indeed, first and second order term of temperature were the only significant parameters explaining the variability of the interval growth rates of predators and prey. Non-linear regressions allowed us to estimate the two-week interval average temperature values above which the growth rate of predators and prey would be negative. These values were 24.7 °C and 25.6 °C for predators and prey, respectively. Despite non-linear regression analyses helped to identify that environmental conditions were an important driver of the dynamics of the community during the two years of our study, the critical values should be taken cautiously as they are only meaningful as a descriptive tool, and lack of predictive power. The reason of this is the arbitrary nature of data they were obtained from. On the one hand, values of temperature and relative humidity used as explanatory variables in the regression analyses were averages of two-week intervals that were simply defined by the sampling dates. On the other hand, this study includes data of two years that were both hot and dry. If more benign environmental data, which probably would be associated to higher values of $r_{t,t+\tau}$ in predators and prey, would have been included in the analyses, it is probable that higher critical values would have been obtained, as the amplitude of the curve relating $r_{t,t+\tau}$ with temperature would be higher. Indeed, this was the case of a previous study that included four years of population dynamics, with summer conditions of two of the years being more benign than those of the other two (Montserrat et al., 2012). Yet, such analyses offer an excellent descriptive tool to evaluate, in specific systems, the effect of environmental conditions on the dynamics of whole communities. Indeed, Fig. 2 clearly shows that the populations of both predator and prey started to decrease when the interval average temperatures were above their respective critical values.

Our results support the hypothesis that higher trophic levels are typically less tolerant to high temperatures than lower trophic levels, and they provide an example of environmental conditions being a direct cause of biological control disruption. Negative effects of high temperatures on the effectiveness of natural enemies, and thereby on pest biological control, have been shown in other agricultural systems (Stavrinides et al., 2010). Furthermore, literature provides many examples of high temperatures negatively affecting fitness-related parameters in many species that are used as biological control agents, including the two species studied here. Life-history parameters of both *E. stipulatus* and *N. californicus* are negatively affected by high temperatures (Ferragut et al., 1987; El-Taj and Jung, 2012) as well as by low relative humidity (Ghazy et al., 2012; C. Guzmán and M. Montserrat, unpublished data), although in the case of *N. californicus* the effect of relative humidity differs among strains (Walzer et al., 2007).

Climate change predictions depict scenarios where natural enemies will be increasingly exposed to extreme high temperatures. In Mediterranean regions such scenarios might be worsen by periods of severe drought. The combination of high temperatures with low air humidity may be detrimental for predatory mite populations, as younger developmental stages of phytoseiid mites, such as eggs, are highly vulnerable to desiccation (DeCourcy-Williams et al., 2004; Ferrero et al., 2010). Therefore, it is expected that future climate change scenarios may provide herbivores with an increase chance to escape predator control, even if high temperatures may also negatively affect herbivores, as occurred in our system during 2010 (Fig. 2). Pest outbreaks will be expected when the range of environmental temperatures lie between predator and prey critical values.

5. Conclusions

This study showed that environmental conditions might be crucial for the success of biological pest control, particularly in areas likely to be affected by climate warming, such as

Mediterranean and temperate regions. Extinction of species, both at a global or local level, can be avoided if populations migrate to more favourable habitats, if organisms overcome stress conditions through phenotypic plasticity, such as behavioural, physiological and/or morphological changes, or if populations undergo evolutionary adaptation (Berg et al., 2010; Hoffmann and Sgrò, 2011). Because of the speed and magnitude of climate change, the response of many populations to counteract the negative effect of increasing temperatures may not be adequate. We believe that in the future the success of biological control strategies will be limited by the natural enemy's capacity to respond adaptatively to rapid changes of climate (Hoffmann and Sgrò, 2011).

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.agee.2012.09.014.

References

Aguilar-Fenollosa, E., Ibañez-Gual, M.V., Pascual-Ruiz, S., Hurtado, M., Jacas, J.A., 2011. Effect of ground-cover management on spider mites and their phytoseiid natural enemies in clementine mandarin orchards (I): top–down regulation mechanisms. Biol. Control 59 (2), 171–179.

Berg, M.P., Kiers, E.T., Driessen, G., van der Heijden, M., Kooi, B.W., Keunen, F., Liefting, M., Verhoef, H.A., Ellers, J., 2010. Adapt or disperse: understanding species persistence in a changing world. Glob. Change Biol. 16, 587–598.

Berkvens, N., Bonte, J., Berkvens, D., Deforce, K., Tirry, L., De Clercq, P., 2008. Pollen as an alternative food for *Harmonia axyridis*. BioControl 53, 201–210.

Beveridge, O.S., Humphries, S., Petchey, O.L., 2010. The interacting effects of temperature and food chain length on trophic abundance and ecosystem function. J. Anim. Ecol. 79, 693–700.

Brown, M., Mathews, S., 2008. Conservation biological control of spirea aphid, *Aphis spiraecola* (Hemiptera: Aphididae) on apple by providing natural alternative food resources. Eur. J. Entomol. 105 (3), 537–540.

Cagnolo, L., Molina, S.I., Valladares, G.R., 2002. Diversity and guild structure of insect assemblages under grazing and exclusion regimes in a montane grassland from Central Argentina. Biodivers. Conserv. 11, 407–420.

DeCourcy-Williams, M.E., Kravar-Garde, L., Fenlon, J.S., Sunderland, K.D., 2004. Phytoseiid mites in protected crops: the effect of humidity and food availability on egg hatch and adult life span of *Iphiseius degenerans*, *Neoseiulus cucumeris*, *N. californicus* and *Phytoseiulus persimilis* (Acari: Phytoseiidae). Exp. Appl. Acarol. 32, 1–13.

Desneux, N., O'Neil, R.J., 2008. Potential of an alternative prey to disrupt predation of the generalist predator, Orius insidiosus, on the pest aphid, *Aphis glycines*, via short-term indirect interactions. Bull. Entomol. Res. 98, 631–639.

El-Taj, H.F., Jung, C., 2012. Effect of temperature on life-history traits of *Neoseiulus californicus* (Acari: Phytoseiidae) fed on *Panonychus ulmi*. Exp. Appl. Acarol. 56, 247–260.

Ferragut, F., García-Mari, F., Costa-Comelles, J., Laborda, R., 1987. Influence of food and temperature on development and oviposition of *Euseius stipulatus* and *Typhlodromus phialatus* (Acari. Phytoseiidae). Exp. Appl. Acarol. 3, 317–329.

Ferrero, M., Gigot, C., Tixier, M.S., van Houten, Y.M., Kreiter, S., 2010. Egg hatching response to a range of air humidities for six species of predatory mites. Entomol. Exp. Appl. 135, 237–244.

Frank, S.D., Shrewsbur, P.M., Denno, R.F., 2011. Plant versus prey resources: influence on omnivore behaviour and herbivore suppression. Biol. Control 57, 229–235.

Ghazy, N.A., Suzuki, T., Shah, M., Amano, H., Ohyama, K., 2012. Using high relative humidity and low air temperature as a long-term storage strategy for the predatory mite *Neoseiulus californicus* (Gamasidae: Phytoseiidae). Biol. Control 60, 241–246.

González-Fernández, J.J., de la Peña, F., Hormaza, J.I., Boyero, J.R., Vela, J.M., Wong, E., Trigo, M.M., Montserrat, M., 2009. Alternative food improves the combined effect of an omnivore and a predator on biological pest control. A case study in avocado orchards. Bull. Entomol. Res. 99 (5), 433–444.

Hoffmann, A.A., Sgrò, C.M., 2011. Climate change and evolutionary adaptation. Nature 470, 479–485.

Holt, R.D., 1977. Predation, apparent competition, and the structure of prey communities. Theor. Popul. Biol. 12, 197–229.

Kuusk, A.K., Ekbom, B., 2010. Lycosid spiders and alternative food: feeding behavior and implications for biological control. Biol. Control 55, 20–26.

Letcher, T.M., 2009. Climate Change: Observed Impact on Planet Earth, 1st ed. Elsevier B.V., Amsterdam, The Netherlands.

Lucas, E., Frechette, B., Alomar, O., 2009. Resource quality, resource availability, and intraguild predation among omnivorous mirids. Biocontrol Sci. Techn. 19, 555–572.

Maoz, Y., Gal, S., Argov, Y., Coll, M., Palevski, E., 2011. Biocontrol of persea mite, Oligonychus perseae, with an exotic spider mite predator and an indigenous pollen feeder. Biol. Control 59, 147–157.

MARM, 2011. Anuario de Estadística del Ministerio de de Medio Ambiente y Medio Rural y Marino, Available via dialog: http://www.mapa.es/es/estadistica/pags/anuario/2009/indice.asp

Messelink, G.J., van Maanen, R., van Steenpaal, S.E.F., Janssen, A., 2008. Biological control of thrips and whiteflies by a shared predator: two pests are better than one. Biol. Control 44, 372–379.

Messelink, G.J., van Maanen, R., van Holstein-Saj, R., Sabelis, M.W., Janssen, A., 2010. Pest species diversity enhances control of spider mites and whiteflies by a generalist phytoseiid predator. BioControl 55, 387–398.

Montserrat, M., de la Peña, F., Hormaza, J.I., González-Fernández, J.J., 2008. How do *Neoseiulus californicus* (Acari: Phytoseiidae) females penetrate densely webbed spider mite nests? Exp. Appl. Acarol. 44, 101–106.

Montserrat, M., Sahún, R.M., Guzmán, C., 2012. Can climate change jeopardize predator control of herbivore invasive species? A case study in avocado agro-ecosystems in Spain. Exp. Appl. Acarol http://dx.doi.org/10.1007/s10493-012-9560-y, in press.

Musser, F.R., Shelton, A.M., 2003. Predation of *Ostrinia nubilalis* (Lepidoptera: Crambidae) eggs in sweet corn by generalist predators and the impact of alternative foods. Environ. Entomol. 32 (5), 1131–1138.

Nomikou, M., Janssen, A., Schraag, R., Sabelis, M.W., 2002. Phytoseiid predators suppress populations of *Bemisia tabaci* on cucumber plants with alternative food. Exp. Appl. Acarol. 27, 57–68.

Nomikou, M., Janssen, A., Sabelis, M.W., 2010. Pollen subsidies promote whitefly control through the numerical response of predatory mites. BioControl 55, 253–260.

Pozzebon, A., Loeb, G.M., Duso, C., 2009. Grape powdery mildew as a food source for generalist predatory mites occurring in vineyards: effects on life-history traits. Ann. Appl. Biol. 155, 81–89.

Sanchez, J.A., Guillespie, D.R., McGregor, R.R., 2003. The effects of mullein plants (*Verbascum thapsus*) on the population dynamics of *Dicyphus hesperus* (Heteroptera: Miridae) in tomato greenhouses. Biol. Control 28, 313–319.

Spellman, B., Brown, M.W., Mathews, C.R., 2006. Effect of floral and extrafloral resources on predation of *Aphis spiraecola* by *Harmonia axyridis* on apple. BioControl 51, 715–724.

Stavrinides, M.C., Daane, K.M., Lampinen, B.D., Mills, N.J., 2010. Plant water stress, leaf temperature, and spider mite (Acari: Tetranychidae) outbreaks in California vineyards. Environ. Entomol. 39 (4), 1232–1241.

Vela, J.M., González-Fernández, J., Wong, E., Montserrat, M., Farré, J.M., Boyero, J.R., 2007. El ácaro del aguacate (*Oligonychus perseae*): Estado actual del problema e investigación en Andalucía. Agrícola Vergel 306, 301–308.

Voigt, W., Perner, J., Davis, A.J., Eggers, T., Schumacher, J., Bahrmann, R., Fabian, B., Heinrich, W., Kohler, G., Lichter, D., Marstaller, R., Sander, F.W., 2003. Trophic levels are differentially sensitive to climate. Ecology 84 (9), 2444–2453.

Vollhardt, I.M.G., Bianchi, F.J.J.A., Wäckers, F.L., Thies, C., Tscharntke, T., 2010. Spatial distribution of flower vs. honeydew resources in cereal fields may affect aphid parasitism. Biol. Control 53, 204–213.

Walzer, A., Castagnoli, M., Simoni, S., Liguori, M., Palevsky, E., Schausberger, P., 2007. Intraspecific variation in humidity susceptibility of the predatory mite *Neoseiulus californicus*: survival, development and reproduction. Biol. Control 41, 42–52.