

Elevated carbon dioxide reduces *Aphis gossypii* intrinsic increase rates without affecting *Aphidius colemani* parasitism rate

Ana Moreno-Delafuente^a, Alberto Fereres^{b,c}, Elisa Viñuela^{a,c}, Pilar Medina^{a,c,*}

^a Unidad de Protección de Cultivos, Departamento de Producción Agraria, Escuela Técnica Superior de Ingeniería Agronómica, Alimentaria y de Biosistemas, Universidad Politécnica de Madrid, Avenida Puerta de Hierro 2, 28040 Madrid, Spain

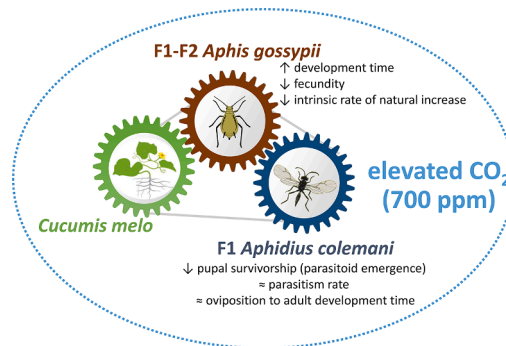
^b Insectos Vectores de Patógenos de Plantas, Departamento de Protección Vegetal, Instituto de Ciencias Agrarias, Consejo Superior de Investigaciones Científicas, Calle Serrano 115 bis, 28006 Madrid, Spain

^c Associate Unit IVAS (CSIC-UPM): Control of Insect Vectors of Viruses in Horticultural Sustainable Systems, Madrid, Spain

HIGHLIGHTS

- Elevated CO₂ induces changes in plant-aphid-parasitoid interactions.
- Elevated CO₂ is detrimental to *Aphis gossypii* by decreasing its growth rate.
- *Aphidius colemani* parasitism rate remained unchanged under elevated CO₂.
- Parasitoid survival can be compromised if aphid population drops under elevated CO₂.

GRAPHICAL ABSTRACT



ARTICLE INFO

Keywords:

Climate change
CO₂
Melon (*Cucumis melo*)
Melon-cotton aphid
Parasitoids
Tritrophic interactions

ABSTRACT

Climate change could increase pest pressure, due to changes in insect distribution and phenology, affecting herbivore-parasitoids interactions and consequently, the efficacy of biological control of crop pests. Elevated carbon dioxide (eCO₂) concentration directly affects plant growth, physiology and biochemistry, subsequently influencing herbivore insects and higher trophic levels. In this study, we analysed the effect of eCO₂ on the individual fitness of *Aphis gossypii* Glover (Hemiptera: Aphididae) and on the performance of its parasitoid *Aphidius colemani* Vierick (Hymenoptera: Braconidae), both reared for two successive generations on melon plants (*Cucumis melo* L., Cucurbitaceae), previous acclimated during two or six weeks to different CO₂ levels, eCO₂ (700 ppm) or ambient CO₂ (400 ppm). There was a significant impact of eCO₂ on aphid performance, prolonging its development time and decreasing its fecundity, negatively affecting the intrinsic rate of natural increase of the two generations of *A. gossypii* grown under eCO₂. There was lower proportion of parasitoids emergence (i.e. pupal survivorship) in first generation *A. colemani* grown under eCO₂. However, parasitism rate, emergence percentages by sex and the overall development time, remained unchanged due to the effect of eCO₂ for the two generations of *A. colemani*. Our research suggests that the effects of eCO₂ on the tritrophic interaction studied could reduce herbivore infestation by decreasing its growth rate. Despite the parasitism efficiency is

* Corresponding author.

E-mail address: pilar.medina@upm.es (P. Medina).

<https://doi.org/10.1016/j.biocontrol.2021.104741>

Received 22 January 2021; Received in revised form 9 May 2021; Accepted 8 August 2021

Available online 10 August 2021

1049-9644/© 2021 The Author(s). Published by Elsevier Inc. This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>).

maintained, in the long term *A. colemani* will not easily find *A. gossypii* for parasitism and their populations could be compromised, although less than other parasitoids with greater host specificity.

1. Introduction

Increasing concentrations of atmospheric carbon dioxide (CO₂), which could reach 670 ppm by the year 2100 (IPCC, 2013), cause land warming and changes in weather patterns, altering biological functions and ecosystems balance (Facey et al., 2014; FAO, 2016). Climate change compromises agriculture, due to its impact on crop production, water supply, yield stability and consequently, undermines global food security (FAO, 2016; IPCC, 2018; Myers et al., 2017).

Biotic factors, such as pests and diseases, also generate important losses in agriculture annually (Flood, 2010; Oerke, 2006). Climate change could increase pest pressure, due to changes in insects distribution, from equator to polar latitudes, and from lower to higher altitudes, that could also break out potential invasive/exotic species. Furthermore, climate change could alter insects phenology, generating winter survival of pests and temporal mismatches among host plants, pests and their natural enemies, finally jeopardizing biological control of many food crops (Lamichhane et al., 2015; Myers et al., 2017). To face these challenges, it is important to understand the effects of climate change on the interactions between the different biological components in agricultural systems (Bélanger and Pilling, 2019).

Elevated CO₂ (eCO₂) principally impacts plant growth, physiology and biochemistry, subsequently affecting herbivore insects and higher trophic levels (Boullis et al., 2015; Facey et al., 2014; Ode et al., 2014; Rosenblatt and Schmitz, 2016; Sun et al., 2011b). Under eCO₂, plants usually increase carbon (C) to nitrogen (N) ratio, due to a dilution in N content, thereby decreasing amino acids and proteins content, altering the food nutrition quality for herbivore insects and therefore, insect performance (Chen et al., 2019; Coviella and Trumble, 1999; Dáder et al., 2016; Moreno-Delafuente et al., 2021). Some insect guilds, such as chewing insects, usually show compensatory feeding under eCO₂; however, sap-feeding insects may perform differently and positive, negative or no responses to eCO₂ could be underscored (Bezemer and Jones, 1998; Hughes and Bazzaz, 2001; Moreno-Delafuente et al., 2021; Oehme et al., 2013; Stiling and Cornelissen, 2007; Trębicki et al., 2017). Bezemer and Jones (1998) suggest that the effects of eCO₂ on sucking insects could be only exhibited under a long-term exposure and biological parameters of sap-sucking insects could change differently over some generations (Chen et al., 2005; Klaiber et al., 2013a). Among sap-feeding insects, eCO₂ impacts aphids' individual behaviour and fitness (Chen et al., 2019; Johnson et al., 2014; Oehme et al., 2013; Ryalls et al., 2017; Ryan et al., 2015, 2014; Sun et al., 2009b; Vassiliadis et al., 2016; Wilkinson and Douglas, 2003). Thus, eCO₂ affects aphid longevity, development and fecundity, finally interfering growth rates and consequently, population dynamics and interspecific competition (Newman et al., 2003; Ryan et al., 2015; Sun et al., 2009a), which leads to change the interactions with their natural enemies (Boullis et al., 2015; Chen et al., 2007; Klaiber et al., 2013b; Sun et al., 2011a). Consequently, under eCO₂ the biological control of aphids by predators and parasitoids could enhance (Chen et al., 2005; Chen et al., 2007), decline (Klaiber et al., 2013b) or being unaffected (Bezemer et al., 1998; Stacey and Fellowes, 2002; Sun et al., 2011a). Furthermore, eCO₂ may induce changes in emissions of volatile organic compounds, that can be used as a defence either to deter herbivores or to attract parasitoids and predators, and also influences pests-natural enemies' interactions under future climate (Himanen et al., 2009; Ode et al., 2014; Sun et al., 2011b).

As no general conclusion can be drawn from literature review, we focused to examine a specific tritrophic system not studied before. Not much research has been undertaken on the effects of eCO₂ on horticulture plants. Therefore, melon (*Cucumis melo* L., Cucurbitaceae) was selected due to its importance as a fruit crop all over the globe, with a

production of 2.7×10^7 tons and an area of 1.05×10^6 ha in 2018 (FAO, 2021). *Aphis gossypii* Glover (Hemiptera: Aphididae) is one of its main pests, considered an agroecosystem threat due to its ability to transmit plant viruses (Blackman and Eastop, 2017; CABI, 2019). The chosen parasitoid was *Aphidius colemani* Vierick (Hymenoptera: Braconidae), a biocontrol agent widely applied to control adults and nymphs of aphids in protected crops (EPPO, 2020; Starý, 1975). Thus, our research aims to analyze the effects of eCO₂ on the individual fitness of *A. gossypii* and on the performance of its natural enemy *A. colemani*, when they reared over two generations on melon plants previous acclimated to different periods to CO₂, to provide more insights about the influence of exposure time to eCO₂ on the performance of both insects.

In a previous study, we observed a significant decrease in *A. gossypii* colony growth and weight under eCO₂, probably due to a reduction in melon plant nutritional quality (Moreno-Delafuente et al., 2021). To get further in-depth knowledge, in this manuscript we intend to explore the importance of host plant mediated effects on the individual fitness (fecundity, development and growth rates) of *A. gossypii* and how this bottom-up effect of CO₂ could influence the host parasitism success of *A. colemani*. This research is essential to enhance the understanding about tritrophic interactions on melon crops and how will be affected by the increasing levels of CO₂.

2. Materials and methods

2.1. Biological material: plants and insects

Seedlings of melon cv. Sancho (Syngenta Seeds B.V., Enkhuizen, The Netherlands) were transplanted to 11 × 11 × 12 cm pots at 7 days old (cotyledon stage) with a mixture 1:1 of vermiculite (No. 3, Asfaltex S.A., Barcelona, Spain) and soil substrate (GoV4, Jiffy International, A.S. Norway). Plants were maintained inside plant growth chamber at 24:20 °C light:dark (L:D) temperature, 60:100 % (L:D) relative humidity (RH) and 16:8 h (L:D) photoperiod until acclimation period. Plants were watered on alternate days with 195 ml/plant adding 20–20–20 (N-P-K) fertilizer (Miller Chemical & Fertilizer Corp., Pennsylvania, USA) at a dose of 1 g/L.

To initiate the laboratory culture of *A. gossypii*, a single virginiparous apterae was originally collected from melon in El Ejido, Spain, in 1998. Laboratory colonies were reared on melon plants for generations inside rearing cages in environmental growth chamber at 23:18 °C (L:D) temperature, 60–80 % RH and 14:10 h (L:D) photoperiod. Aphids were synchronized prior the bioassays to guarantee age homogeneity (10–11 days old) when experiments began. Plants and aphid colonies were reared adapting the methodology of Carmo-Sousa et al. (2014).

Mummies of *A. colemani* were supplied by Koppert Biological Systems (Berkel en Rodenrijs, The Netherlands) and maintained in a refrigerator at 4 °C. In order to obtain ≤ 24-hour-old adults the day of mating, mummies were transferred to the growth chamber at 25:20 °C (L:D) temperature, 65 ± 10 % RH and 16:8 h (L:D) photoperiod, the previous three days. All adults emerged were removed the day prior mating, and only adults emerged in the final 24 h were sexed and matched. Pairs were transferred to glass cylinders (5 cm high and 3 cm in diameter) with honey drops on a mesh lid and left to mate 24 h. These pairs were used for the experiment.

2.2. CO₂ acclimation procedure

Plant acclimation to CO₂ was performed in two walk-in climate chambers with same conditions of temperature (24:20 °C (L:D)), RH (65 ± 5 %) and photoperiod (14:10 h (L:D)) with $310 \pm 3 \mu\text{mol m}^{-2} \text{s}^{-1}$ light

intensity at canopy level (GreenPower LED production dr/b/fr 120, Philips, Eindhoven, The Netherlands). CO₂ atmospheric concentration differed between chambers, one chamber with elevated CO₂ (eCO₂) (700 ppm) and the other with ambient CO₂ (aCO₂) (400 ppm). The aCO₂ chamber conditions were monitored with a portable humidity, temperature and CO₂ datalogger device (Rotronic AG CP11, Bassersdorf, Switzerland), while eCO₂ chamber was monitored with the incorporated system that automatically regulates the chamber gas concentration. Furthermore, two data loggers (Tinytag Ultra 2, Gemini Data Loggers, UK) were displayed in each chamber to record temperature and humidity data every hour. Plants were exposed to different period of acclimation and different CO₂ concentration (Klaiber et al., 2013b). Plants were seeded and grown under general ambient conditions (see 2.1.) and then, in order to acclimatize to CO₂, transferred to either eCO₂ or aCO₂, for 6 or 2 weeks (four treatments in total) before the beginning of the bioassays (Moreno-Delafuente et al., 2021). All plants were 7-weeks-old when experiments with *A. gossypii* started and insect experimental units were maintained in the eCO₂ or aCO₂ chamber, depending on the bioassay.

2.3. Effects of CO₂ on *Aphis gossypii* individual fitness

Daily monitoring of two successive generations of *A. gossypii* was carried out on melon plants acclimated to different CO₂ concentration and period of acclimation in order to assess individual aphid performance. A single aptera adult female (11 days old) was placed on the adaxial side of the third true leaf of 7-week-old plants. Each adult female was confined in a 3 cm diameter clip-cage with a mesh at the top to allow transpiration, and laid 24 h to generate offspring. Afterwards, surplus nymphs as well as the adult were removed leaving two neonates per clip-cage, which were monitored until adulthood (first generation: F1). From the two aphids inside the clip-cage, the first newly emerged adult that generated offspring was maintained and the other removed (Fereses et al., 1989). F1 adult offspring were counted by removing nymphs daily until adults completed a period equal to the pre-reproductive period (number of days from birth to the onset of its reproduction). To evaluate the second generation (F2), the first nymph generated by the newly-emerged adult from F1 was placed in another clip-cage on the adaxial side of the same leaf than previous generation, repeating the process described above. Duration of each nymphal instar, pre-reproductive period (*d*), effective fecundity (offspring produced during a period equal to the pre-reproductive period) (*Md*), intrinsic rate of natural increase ($rm = 0.738 (\ln Md)/d$) (Wyatt and White, 1977), mean generation time ($Td = d/0.738$), and mean relative growth rate ($RGR = rm/0.86$) (Dixon, 1987) were calculated for both generations. Plants within each climate chamber were randomized every day to avoid any positional effects. Experiments were performed twice and 32–38 replicates per treatment were obtained.

2.4. Effects of CO₂ on the performance of first generation *Aphidius colemani*

We tested *A. colemani* performance when developed on aphids grown under aCO₂ or eCO₂ on melon plants previously acclimated two or six weeks to the different CO₂ concentrations. The day that CO₂-acclimation of melon plants concluded, one adult apterous female of *A. gossypii*, 10–11 days old, was placed on the adaxial surface of the 13–14th leaf of each plant in 3 cm diameter clip-cages. Twenty-four hours later, adults and first instar nymphs except five per clip-cage, were removed. Nymphs developed to adults in seven days and started to generate offspring. One day after first adults appeared, they were removed to get synchronized second generation nymphs. Progeny was kept until third instar nymphs arose (4 days after adult generated first offspring) in 12 cm cylindrical cages, with a hole with foam between the lid and the cage to allow the petiole insertion. Twenty nymphs were left on the cage and a pair of *A. colemani* parasitoids (male and female previously mated for 24 h) was

introduced and left 24 h for oviposition and then removed. Plants with parasitized nymphs were maintained in its correspondent acclimation chambers (aCO₂ or eCO₂) and monitored every day until mummies appeared. Aphid mummies were collected and transferred to 12 cm diameter cylindrical cages with wet filter paper and a cotton layer and maintained in the appropriate CO₂ chamber, and checked daily for adult parasitoid emergence. Adults of *A. colemani* were sexed using a zoom stereomicroscope (Nikon SMZ 1500, Kanagawa 210-0005, Japan). The parameters evaluated were the parasitism rate, understood as the proportion of the total number of exposed aphids that mummifies (Martinou and Wright, 2007); the pupal survivorship, i.e. the percentage of *A. colemani* emerged over the total number of mummies; the percentage of males and females emerged; and the individual development time (d) from oviposition to mummy, from oviposition to adult and from mummy to adult emergence, this last parameter also for females and males separately (Zamani et al., 2007). One plant with one cage was one replicate. During acclimation and bioassay, plants in each climate chamber were randomized every day to avoid any positional effects. Experiments were performed twice. We obtained 24 replicates in total for each treatment, except for 2 weeks of acclimation period to aCO₂, with *n* = 28.

2.5. Assessment of the indirect effect of CO₂ on second generation *Aphidius colemani*

In the day of maximum emergency of parasitoids from first generation, adults (<24 h old) were sexed and matched. Under the respective CO₂ conditions, they were let to mate during one day. Then, from 4-week-old melon plants maintained in the plant growth chamber at environmental conditions, expanded leaves (7–8 cm approximately) were excised and inserted in Eppendorfs (2 ml) with agar-agar 2% to help maintaining the turgidity during the experiment. Leaves were placed in ventilated cages (12 cm diameter) with wet filter paper and cotton layer in the bottom. Twenty third-instar *A. gossypii* nymphs (four days old), previously synchronized and kept in the environmental growth chamber, were transferred to the leaves, left one hour to settle in, and then, a pair of *A. colemani* with the mated female was introduced. In order to separate the effect that eCO₂ exerts directly on the parasitoid from the mediated effect via its aphid host, these experimental units from all treatments were transferred to aCO₂ chamber. Thus, in this second generation, we evaluated the indirect effect of CO₂ on the parasitoid. After 24 h of oviposition, parasitoids were removed. Aphids were checked every day. The same *A. colemani* biological parameters as described in the previous section were determined. Experiment was repeated twice and 19–27 replicates per treatment were obtained.

2.6. Indirect effect of CO₂ on the survivorship of *Aphidius colemani*

The survivorship of *A. colemani* mated pairs that oviposited aphid nymphs in the parasitism bioassay described above (see 2.5) was evaluated. In order to test the indirect effect of CO₂ on *A. colemani* survival, males and females matched were reared under aCO₂, since its previous parasitoid life cycle (egg-larvae-pupae-adult emergence) was on aphids grown under aCO₂ and eCO₂, on melon plants previously acclimated two or six weeks to the different CO₂ concentration. The wasps, after parasitizing *A. gossypii* nymphs, were transferred in pairs to glass cylinders (5 cm high and 3 cm in diameter) with honey drops on the mesh lid as food supply, and checked every day until death.

2.7. Statistical analysis

Aphis gossypii individual fitness and *A. colemani* performance bioassays were conducted twice in time to increase number of repetitions per treatment and to decrease potential chamber effects. Differences between the two trials were analyzed by Student T-test or by non-parametric U Mann-Whitney test when the assumption of homogeneity of variances was

not met. As no significant effect due to trial was detected in the majority of cases ($P \geq 0.05$), data were pooled to continue the statistical analysis.

Afterwards, two-way ANOVA statistical test, conducted by a General Linear Model, was performed to analyse the effects of CO₂ concentration (aCO₂-400 ppm or eCO₂-700 ppm), acclimation period to CO₂ (two or six weeks) and their possible interaction, on *A. gossypii* individual fitness and *A. colemani* performance reared on acclimated melon plants. Some parameters needed data transformation ($\log(x + 1)$, or $\arcsin(\sqrt{x/100})$) in case of percentages) to fit normality and homogeneity of variance assumptions (level of significance, $\alpha = 0.05$). Pairwise comparisons for main effects were corrected using a Bonferroni adjustment. Parasitoid survivorship data were analyzed by Kaplan–Meier survival analysis with overall comparisons. As no statistically significant differences were observed (Log-Rank test, $P > 0.05$), it was not necessary to perform a pairwise comparison procedure. All statistical test were conducted using SPSS software (IBM SPSS Statistics 26.0.0.0 for Windows, Chicago, USA).

3. Results

3.1. Effects of CO₂ on *Aphis gossypii* individual fitness

Aphis gossypii biological parameters were significantly affected by CO₂ concentration but not by acclimation period (Fig. 1, Supplementary Table S1). The pre-reproductive period (d), as well as the mean generation time (Td), were significantly longer for aphids developed on plants under eCO₂ compared to aCO₂, both for first generation (F1) (d : $F_{1,144} = 4.448$, $P = 0.037$; Td : $F_{1,144} = 4.432$, $P = 0.037$) and for second generation (F2) (d : $F_{1,134} = 7.409$, $P = 0.007$; Td : $F_{1,134} = 7.409$, $P = 0.007$) (Fig. 1, Supplementary Table S1). Development time of each nymphal instar was only significantly different for the first nymphal stage (N1) of F1 *A. gossypii* and the third nymphal stage (N3) of F2 *A. gossypii*, both nymphal periods were longer on melon plants acclimated to eCO₂ compared to aCO₂ (N1 – F1: $F_{1,144} = 8.552$, $P = 0.004$; N3 – F2: $F_{1,134} = 6.681$, $P = 0.011$). In the remaining nymphal stages, as well as the arrival to adulthood (when aphids reached sexual maturity and started to generate offspring), no differences were observed among treatments in any of the two generations (Supplementary Table S2).

There was a significant decrease in the effective fecundity (Md) in *A. gossypii* both for first and for second generation under eCO₂ compared to aCO₂ (Md – F1: $F_{1,144} = 11.133$, $P = 0.001$; F2: $F_{1,134} = 9.265$, $P = 0.003$) (Fig. 1, Supplementary Table S1).

For both *A. gossypii* generations, there was a significant reduction in the intrinsic rate of natural increase (r_m) when aphids grown under eCO₂ compared to aCO₂. Because of the mean relative growth rate (RGR) is a related parameter of r_m , the same trend was observed for the two *A. gossypii* generations (r_m and RGR – F1: $F_{1,144} = 16.380$, $P \leq 0.001$; F2: $F_{1,134} = 19.367$, $P \leq 0.001$) (Fig. 1, Supplementary Table S1).

3.2. Effects of CO₂ on the performance of first generation *Aphidius colemani*

Pupal survivorship (percentage of emergence) significantly decreased in *A. colemani* grown under eCO₂, independently of the acclimation period, compared to parasitoids reared under aCO₂ ($F_{1,96} = 8.573$; $P = 0.004$) (Table 1).

The interaction between CO₂ concentration and the previous acclimation period was significantly different on the development time from oviposition to mummy (acclimation period \times CO₂: $F_{1,96} = 4.574$, $P = 0.035$), but the interaction effect was not conclusive (Table 1). The development time from mummy to adult emergence and the overall development time from oviposition to adult emergence (i.e., the sum of development time from oviposition to mummy and from mummy to adult emergence) were not influenced by eCO₂ or by the acclimation period (Table 1). The rest of F1 *A. colemani* biological parameters (parasitism rate and percentage of females or males emerged) were not

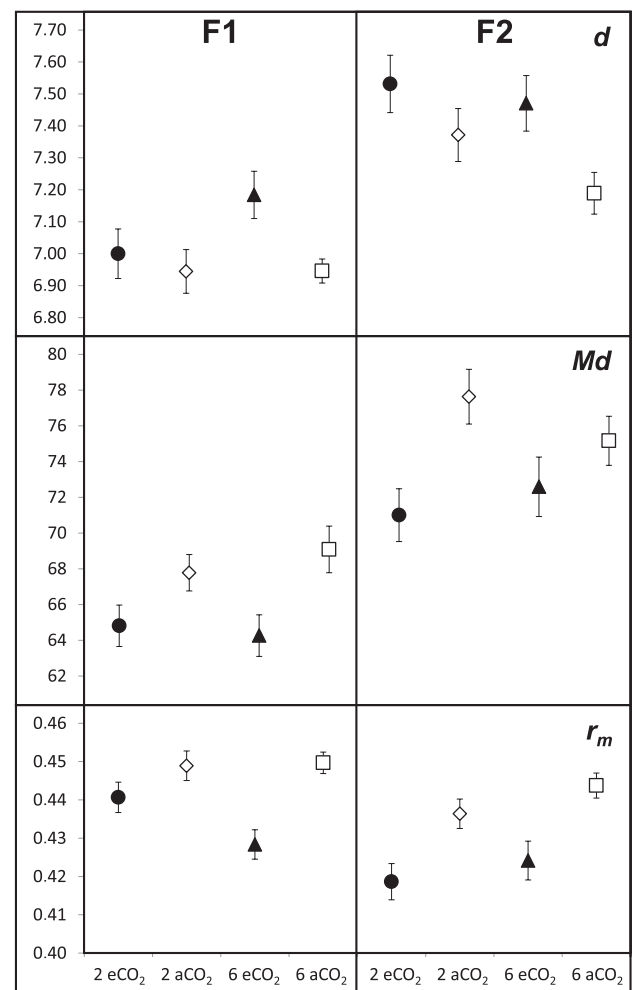


Fig. 1. Life history parameters of two successive generations of *Aphis gossypii*. Mean and standard error bars of the pre-reproductive period “ d ” (days), the effective fecundity “ Md ” and the intrinsic rate of natural increase “ r_m ” of first (F1) and second (F2) generation of aphids reared under ambient (aCO₂, 400 ppm) or elevated (eCO₂, 700 ppm) CO₂, on melon plants previously acclimated 2 or 6 weeks to the respective CO₂ concentration. The CO₂ concentration, but not the acclimation period or the interaction between factors, was statistically significant in all parameters ($P < 0.05$, Two-way ANOVA test and Bonferroni correction for pairwise comparison).

affected by CO₂ neither by the acclimation period to the different CO₂ concentration (Table 1).

3.3. Assessment of the indirect effect of CO₂ on second generation *Aphidius colemani*

When evaluated the indirect effect of CO₂, the development time from mummy to female adult emergence was significantly shorter in *A. colemani* grown on plants and aphids previous acclimated to eCO₂, compared to those grown on plants and aphids developed under aCO₂ ($F_{1,94} = 5.439$; $P = 0.022$). The development time from mummy to pooled adults was significantly shorter in parasitoids grown on plants and aphids grown under eCO₂, compared to those grown on plants and aphids developed under aCO₂ ($F_{1,96} = 4.070$; $P = 0.046$). None of the other biological parameters (parasitism rate, pupal survivorship, development time from oviposition to mummy, and from oviposition to adult emergence, among others) showed significant differences when the indirect effect of eCO₂ on the second generation of *A. colemani* was evaluated (Table 2).

Table 1

Biological parameters of *Aphidius colemani* first generation developed under ambient (400 ppm) or elevated (700 ppm) CO₂, on aphids grown on melon plants previously acclimated for 2 or 6 weeks to the respective CO₂ concentration.

First generation parasitoid parameter	Acclimation period	[CO ₂]	Mean ± SE		Acclimation period		[CO ₂]		Acclimation period × [CO ₂]			
			elevated	ambient	F	P	F	P	F	P		
Parasitism and emergence percentages (%)	Parasitism rate	2 weeks	77.85 ± 4.87	86.13 ± 2.42	81.99 ± 2.84	1.87	0.18	1.37	0.25	0.73	0.40	
		6 weeks	75.74 ± 4.41	77.04 ± 4.61	76.39 ± 2.95							
	Pupal survivorship	2 weeks	85.78 ± 2.77	93.07 ± 1.61	89.42 ± 1.48	2.14	0.15	8.57	0.004**	0.72	0.40	
		6 weeks	90.53 ± 2.48	94.34 ± 1.50	92.43 ± 1.53							
	Percentage of females emerged	2 weeks	31.75 ± 6.11	19.79 ± 4.95	25.77 ± 4.12	1.74	0.19	2.14	0.15	0.31	0.58	
		6 weeks	36.30 ± 7.29	30.90 ± 5.31	33.60 ± 4.27							
	Percentage of males emerged	2 weeks	34.03 ± 4.27	25.34 ± 4.12	29.68 ± 4.12							
		6 weeks	68.25 ± 6.11	80.21 ± 4.95	74.23 ± 4.12							
	Development time (days)	Oviposition to mummy	2 weeks	8.08 ± 0.10 bB	8.47 ± 0.10 a	8.27 ± 0.07	0.57	0.45	3.41	0.07	4.57	0.035*
			6 weeks	8.36 ± 0.12 A	8.34 ± 0.08	8.35 ± 0.07						
		Mummy to adult (♀♀)	2 weeks	4.82 ± 0.14	4.69 ± 0.18	4.76 ± 0.14	1.38	0.25	0.00	0.99	0.46	0.50
			6 weeks	4.92 ± 0.11	5.05 ± 0.28	4.98 ± 0.13						
Mummy to adult (♂♂)		2 weeks	4.70 ± 0.09	4.71 ± 0.08	4.70 ± 0.06	0.08	0.78	1.33	0.25	0.95	0.33	
		6 weeks	4.58 ± 0.10	4.77 ± 0.09	4.68 ± 0.07							
Mummy to adult (♀+♂)		2 weeks	4.73 ± 0.10	4.72 ± 0.08	4.72 ± 0.06	0.13	0.72	0.33	0.57	0.45	0.50	
		6 weeks	4.70 ± 0.09	4.81 ± 0.08	4.75 ± 0.06							
Oviposition to adult (♀+♂)		2 weeks	12.80 ± 0.17	13.19 ± 0.15	12.99 ± 0.11	0.45	0.50	2.15	0.15	0.94	0.34	
		6 weeks	13.06 ± 0.18	13.14 ± 0.15	13.10 ± 0.12							
			Mean ± SE	12.93 ± 0.12	13.17 ± 0.11							

[CO₂]: CO₂ concentration. SE: Standard error. P-values (P) according to Two-way ANOVA test and Bonferroni correction for pairwise comparisons. Significant differences represented by asterisks: * ($P \leq 0.05$) and ** ($P \leq 0.01$). Different upper case letters within columns indicate differences among acclimation period and different lower case letters within rows indicate differences among CO₂ concentration. Statistic values of "Percentage of females emerged" and "Percentage of males emerged" are the same as these parameters are complementary, thereby they are displayed once.

3.4. Indirect effect of CO₂ on the survivorship of *Aphidius colemani*

There was not an indirect effect of eCO₂ on the survivorship of *A. colemani* mated adults. Female and male survival curves, analyzed separately, did not significantly differ among treatments (Log Rank test, female: $\chi^2 = 3.055$, df = 3, $P = 0.383$; male: $\chi^2 = 0.750$, df = 3, $P = 0.861$) (Fig. 2).

4. Discussion

Nowadays, several studies confirm that the increasing levels of atmospheric CO₂ alter trophic interactions among crops, their phytophagous insects and their related natural enemies (Boullis et al., 2015; Chen et al., 2005; Klaiber et al., 2013b; Li et al., 2016; Sun et al., 2011b). Our findings indicate a noticeable negative impact of eCO₂ on *A. gossypii* fitness but practically no effect on *A. colemani* biological parameters when these insects were grown on melon plants.

4.1. Effects of eCO₂ on aphid individual fitness

Plant N content has a direct effect on herbivores growth, reproduction and survival; being N a limiting nutrient for many herbivorous insects, such as aphids (Douglas, 2003; Mattson, 1980). In a previous study we observed a decrease in foliar N content and an increase in C:N ratio in melon plants exposed to eCO₂ compared to aCO₂, independently of acclimation period, significantly reducing the content of some essential amino acids for aphid nutrition (Moreno-Delafuente et al., 2021). This change in host plant nutritional quality could presumably have caused the observed changes in *A. gossypii* life history under eCO₂ in our study. All main *A. gossypii* individual fitness parameters were influenced by

eCO₂ compared to aCO₂, independently of acclimation period, for the two successive generations of *A. gossypii*, verifying a negative response to eCO₂, by decreasing its reproduction and increasing its development time, leading to a significant decrease in the intrinsic rate of natural increase as well as in the mean relative growth rate. According to Dixon (1987), the latter aphid growth rate is principally determined by food quality and temperature, consequently a decrease in N content due to eCO₂, could lead to a poor food quality and produce the decrease in the mean relative growth rate of *A. gossypii* in our experimental conditions. The performance of other aphid species has also been negatively impacted under eCO₂ due to a dilution in N content in their host plants, as *Myzus persicae* Sulzer on pepper (*Capsicum annum* L.) or *Rhopalosiphum padi* L. on wheat (*Triticum aestivum* L.) (Dáder et al., 2016; Trębicki et al., 2016).

The increase in development time of herbivores could be a measure to compensate the reduction in nutrient content on plant tissues (Boullis et al., 2015; Coviella and Trumble, 1999; Stiling and Cornelissen, 2007), as observed in the present study with longer development time in both generations of *A. gossypii* reared under eCO₂. When aphids exposed to eCO₂ undergo compensatory feeding to satisfy their nutrient requirements, this could induce a greater damage. On the one hand, directly, weakening the plant by sucking phloem sap and increasing the production of honeydew that generates the growth of sooty mold and subsequently reducing the photosynthesis. On the other hand, indirectly, increasing the risk of virus transmission under climate change conditions (Sun et al., 2009b; Trębicki, 2020; Van Emden and Harrington, 2017).

Elevated CO₂ affects more markedly multivoltine insects, such as aphids, which have a short adult lifespan, the immature stages develop quickly and multiple generations occur in one season (Bezemer and

Table 2

Biological parameters to evaluate the indirect effect of CO₂ on *Aphidius colemani* second generation developed under ambient conditions, after its first generation was developed on aphids grown on melon plants previously acclimated for 2 or 6 weeks to ambient (400 ppm) or elevated (700 ppm) CO₂ concentration.

Second generation parasitoid parameter		Acclimation period	[CO ₂]		Mean ± SE	Acclimation period		[CO ₂]		Acclimation period × [CO ₂]	
			elevated	ambient		F	P	F	P	F	P
Parasitism and emergence percentages (%)	Parasitism rate	2 weeks	86.10 ± 3.50	90.11 ± 2.16	88.10 ± 2.13	0.87	0.35	1.05	0.31	0.07	0.79
		6 weeks	84.00 ± 3.42	86.38 ± 3.05	85.19 ± 2.28						
		Mean ± SE	85.05 ± 2.13	88.24 ± 2.28							
	Pupal survivorship	2 weeks	94.28 ± 1.83	95.69 ± 1.10	94.99 ± 0.98	0.58	0.45	0.99	0.32	0.00	0.99
		6 weeks	95.35 ± 1.46	96.81 ± 0.96	96.08 ± 1.05						
		Mean ± SE	94.82 ± 0.98	96.25 ± 1.05							
	Percentage of females emerged	2 weeks	71.57 ± 4.47	79.91 ± 2.37	75.74 ± 2.52	0.40	0.53	0.01	0.92	2.52	0.12
		6 weeks	77.04 ± 2.25	70.08 ± 5.17	73.56 ± 2.69						
		Mean ± SE	74.30 ± 2.52	74.99 ± 2.69							
	Percentage of males emerged	2 weeks	28.43 ± 4.47	20.09 ± 2.37	24.26 ± 2.52						
		6 weeks	22.97 ± 2.25	29.92 ± 5.17	26.44 ± 2.69						
		Mean ± SE	25.70 ± 2.52	25.01 ± 2.69							
Development time (days)	Oviposition to mummy	2 weeks	8.10 ± 0.17	8.52 ± 0.20	8.31 ± 0.14	0.28	0.60	2.53	0.12	0.24	0.63
		6 weeks	8.30 ± 0.20	8.53 ± 0.25	8.42 ± 0.15						
		Mean ± SE	8.20 ± 0.14	8.52 ± 0.15							
	Mummy to adult (♀♀)	2 weeks	5.03 ± 0.06	5.08 ± 0.06	5.06 ± 0.04	1.28	0.26	5.44	0.022*	1.91	0.17
		6 weeks	4.89 ± 0.05	5.10 ± 0.05	4.99 ± 0.04						
		Mean ± SE	4.96 ± 0.04b	5.09 ± 0.04a							
	Mummy to adult (♂♂)	2 weeks	4.78 ± 0.09	4.87 ± 0.07	4.83 ± 0.06	0.13	0.72	0.59	0.45	0.11	0.74
		6 weeks	4.78 ± 0.09	4.81 ± 0.07	4.80 ± 0.06						
		Mean ± SE	4.78 ± 0.06	4.84 ± 0.06							
	Mummy to adult (♀+♂)	2 weeks	4.97 ± 0.06	5.04 ± 0.05	5.01 ± 0.04	1.11	0.29	4.07	0.046*	0.49	0.48
		6 weeks	4.88 ± 0.04	5.02 ± 0.05	4.95 ± 0.04						
		Mean ± SE	4.93 ± 0.04b	5.03 ± 0.04 a							
	Oviposition to adult (♀+♂)	2 weeks	13.07 ± 0.19	13.56 ± 0.23	13.32 ± 0.15	0.06	0.82	3.67	0.06	0.08	0.78
		6 weeks	13.19 ± 0.22	13.55 ± 0.26	13.37 ± 0.16						
		Mean ± SE	13.13 ± 0.15	13.56 ± 0.16							

[CO₂]: CO₂ concentration. SE: Standard error. P-values (P) according to Two-way ANOVA test and Bonferroni correction for pairwise comparisons. Significant differences represented by asterisk: * (P ≤ 0.05). Different lower case letters within rows indicate differences among CO₂ concentration (P ≤ 0.05). Statistic values of "Percentage of females emerged" and "Percentage of males emerged" are the same as these parameters are complementary, thereby they are displayed once.

Jones, 1998). In these cases, a small difference in development time is likely to cause inter-generation population effects, as we have observed in *A. gossypii* under eCO₂ with a significant decrease in its intrinsic growth rate, as well as with a decline in the colony population growth (Moreno-Delafuente et al., 2021).

However, different trends have been observed in other plant-aphid interactions under eCO₂ and the scientific community assume that it is not possible to generalize the effect of eCO₂ on phloem feeders (Bezemer and Jones, 1998; Boullis et al., 2015; Hoover and Newman, 2004; Hullé et al., 2010; Trębicki et al., 2017). The aphid response to eCO₂ is not only

particular of the plant or aphid species (Hughes and Bazzaz, 2001; Sun et al., 2011b), but also specific of the host plant genotype (Johnson et al., 2014; Sun et al., 2011a), or even the aphid clone, as we can observe with an opposed *A. gossypii* performance between our study and other research in which *A. gossypii* increased its fecundity, body weight and population abundance under eCO₂ (Chen et al., 2005; Jiang et al., 2016). A different response of aphids to eCO₂ could be also dependent on the time of exposure to eCO₂ or even on the generation evaluated (Chen et al., 2005; Klaiber et al., 2013a).

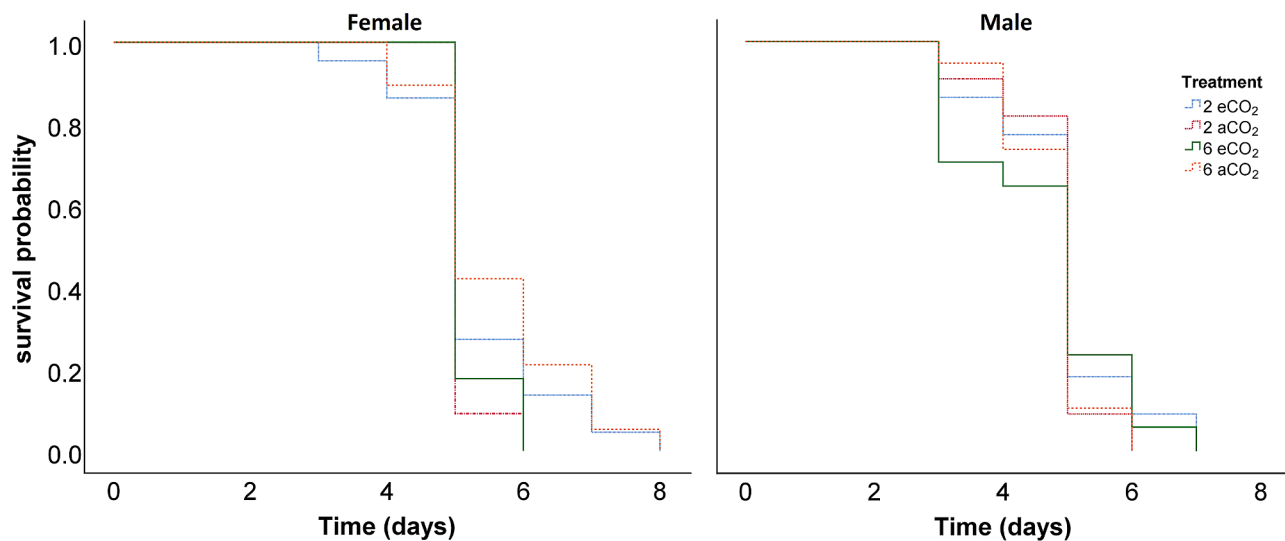


Fig. 2. Parasitoids survivorship. Survival curves of females and males of *Aphidius colemani* grown under ambient conditions, after their immature stages were developed under ambient CO₂ (400 ppm; aCO₂) or elevated CO₂ (700 ppm; eCO₂), on aphids grown on melon plants previously acclimated for 2 or 6 weeks to the respective CO₂ concentration. No significant differences were found among treatments (Log rank test, $P > 0.05$, females and males separately).

4.2. Effects of eCO₂ on parasitoid performance

Our study differentiated between interactive direct and host-mediated (indirect) effects of eCO₂, when parasitoids were grown under eCO₂ on plants and aphids previous acclimated to eCO₂; and the indirect effects of eCO₂, when parasitoids were grown under ambient CO₂ on plants and aphids previous acclimated to eCO₂ (De Paulo et al., 2020; Murray et al., 2013; King et al., 2003).

Modifications in plant quality and physiology due to eCO₂ will affect herbivorous insect chemical composition and consequently, higher trophic levels (Hoover and Newman, 2004; Roth and Lindroth, 1995; Sun et al., 2011b). Thus, parasitoid and predator performance will be altered due to changes in their diet (Chen et al., 2005; Klaiber et al., 2013b). In broad terms, eCO₂ could impact fitness, survival and population dynamics of herbivores and natural enemies, subsequently affecting the interactions with other plants and insects (Coviella and Trumble, 1999).

The parasitism efficiency, i.e. the parasitism rate, together with offspring development time, percentage of males and females emerged, and longevity, may characterize parasitoid performance under eCO₂ (Klaiber et al., 2013b). There is no evidence of reduction in the parasitism efficiency of the two generations of *A. colemani* under the eCO₂ conditions proposed. Thus, parasitism rate remains unchanged when evaluated the interactive direct and indirect effect of eCO₂ on F1, but also when evaluated the indirect eCO₂-effect on F2. As observed in our tritrophic system, the parasitism rate of *Aphidius matricariae* Haliday was not affected by eCO₂ when parasitized *M. persicae* (Bezemer et al., 1998). However, *Diaeretiella rapae* McIntosh parasitism efficiency was reduced under eCO₂ when parasitized *Brevicoryne brassicae* L. grown on *Brassica oleracea* L. var. *gemmifera* previously acclimated ten weeks to eCO₂ (Klaiber et al., 2013b). Conversely, the parasitism efficiency could increase under eCO₂, as occurred to *Aphidius picipes* Nees parasitizing *Sitobion avenae* Fabricius (Chen et al., 2007).

Although the proportion of emerged adults separated by sex remained unchanged under eCO₂ compared to under aCO₂, we observed a slightly, but significant reduction in pupal survivorship of F1 *A. colemani*, decreasing the proportion of total emerged adults exposed to eCO₂ compared to those exposed to aCO₂. In agreement with our results, *A. picipes* emergence rate (i.e. pupal survivorship) was reduced under 750 ppm CO₂ concentration (Chen et al., 2007).

Elevated CO₂ could also affect natural enemies development time (Boullis et al., 2015; Klaiber et al., 2013b; Roth and Lindroth, 1995; Sun et al., 2011b). In our study, F1 *A. colemani* development time from oviposition to mummification was affected by eCO₂ depending on the acclimation period, and F2 parasitoid shortened its development time from mummification to adult emergence (in females and in both sexes pooled). However, the overall development time (from oviposition to adult emergence) of *A. colemani* was not influenced by eCO₂ in both generations. In agreement with our results, the development time and growth of *Cotesia melanoscela* Ratz. parasitizing *Lymantria dispar* L. were not affected by eCO₂ (Roth and Lindroth, 1995). However, eCO₂ shortened the overall development time of the parasitoid *Lysiphlebia japonica* Ashmead when parasitized *A. gossypii* reared on cotton, although finally did not influence the parasitism and emergence rates (Sun et al., 2011a).

Overall, the effects observed in *A. colemani* life parameters in our study could have presumably been caused by a reduction in the size of their aphid hosts grown under eCO₂ (Moreno-Delafuente et al., 2021).

Aphidius colemani survival was not indirectly influenced by CO₂ concentration neither by the acclimation period, contrary to *D. rapae* which lived shorter when parasitized *B. brassicae* grown on plants previous acclimated six and ten weeks to eCO₂ (Klaiber et al., 2013b).

Therefore, positive, negative and neutral effects of eCO₂ on the third trophic level may be underscored, so a clear effect of eCO₂ on parasitoids and other natural enemies could not be generalized (Boullis et al., 2015). The effect of eCO₂ on the plant-insect interaction studied is not only species-specific but also specific to the plant-insect system itself (Coviella and Trumble, 1999). The experimental conditions could have influenced our results, and the variation observed among studies are more than just species-specific relationships (Boullis et al., 2015; Martinou and Wright, 2007; Torres et al., 2007; Zamani et al., 2007).

4.3. Conclusions

Elevated CO₂ affects the tritrophic system studied, specially the herbivore level, presumably due to changes mediated by the melon plant (Moreno-Delafuente et al., 2021). Thus, eCO₂ could decrease the pest status of *A. gossypii* without practically affecting *A. colemani* individual performance. Overall, eCO₂ is detrimental to *A. gossypii* by reducing its intrinsic rate of natural increase and by maintaining *A. colemani*

parasitism efficiency, which will probably reduce faster the pest population growth. Under the eCO₂ conditions proposed, the individual parasitoid biology seems to be slightly affected by eCO₂ however, the decrease in *A. gossypii* population would imply changes in the natural enemy ecology. *Aphidius colemani*, as a polyphagous parasitoid (Jacas et al., 2006), could need to alternate hosts, even suboptimal species, to perpetuate under eCO₂, which indirectly would favour the intraguild competition among generalist aphid parasitoids. In a climate change context, some temporal host-parasitoid mismatches due to modifications in the phenology of host species induced by warmer environments (Tougeron et al., 2020) should be added to the potential alterations in host–parasitoid food-web composition. Both phenomena together may affect the efficiency of biological control.

Our research supports the notion that eCO₂ generates potential impacts on agricultural systems, with changes in trophic interactions among crops, phytophagous insects and their associated natural enemies. Other abiotic factors, such as elevated ozone, water deficit or heat stress, have also an impact on agriculture and complex studies evaluating the effect of different factors combined should be performed to better understand how climate change is affecting multitrophic interactions and therefore, the effectiveness of the biological control on agroecosystems.

CRediT authorship contribution statement

Ana Moreno-Delafuente: Conceptualization, Methodology, Formal analysis, Investigation, Data curation, Writing - original draft, Writing - review & editing, Visualization. **Alberto Fereres:** Resources, Writing - review & editing, Supervision, Funding acquisition. **Elisa Viñuela:** Resources, Writing - review & editing, Funding acquisition. **Pilar Medina:** Conceptualization, Methodology, Validation, Resources, Writing - original draft, Writing - review & editing, Visualization, Supervision, Project administration, Funding acquisition.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This work was supported by the Spanish Ministry of Science, Innovation and Universities with R&D project grants (Numbers AGL2013-47603-C2, AGL2017-83498-C2-2-R); and with a PhD grant to Ana Moreno-Delafuente (FPU2015-05173). We are very grateful to Celeste Azpiazu, Sergio Estébanez, Elisa Garzo, Ignacio Morales, Ana Murcia, Sandra Pla, Pablo Poveda, Inés Prieto, Gonzalo Sancho and Andrea Wanumen for technical assistance; and Miguel Ángel Ibáñez for his statistical advises.

Data statement

The datasets generated during the current study are available from the corresponding author on reasonable request.

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.biocontrol.2021.104741>.

References

- Bélanger, J., Pilling, D., 2019. The State of the World's Biodiversity for Food and Agriculture, FAO. 978-92-5-131270-4.
- Bezemer, T.M., Jones, T.H., Knight, K.J., 1998. Long-term effects of elevated CO₂ and temperature on populations of the peach potato aphid *Myzus persicae* and its parasitoid *Aphidius matricariae*. *Oecologia* 116 (1-2), 128–135. <https://doi.org/10.1007/s004420050571>.
- Bezemer, T.M., Jones, T.H., 1998. Plant-insect herbivore interactions in elevated atmospheric CO₂: quantitative analyses and guild effects. *Oikos* 82, 212–222. <https://doi.org/10.2307/3546961>.
- Blackman, R.L., Eastop, V.F., 2017. *Taxonomic Issues*. In: Van Emden, H.F., Harrington, R. (Eds.), *Aphids as Crop Pests*. CAB International, Oxfordshire, UK, pp. 1–36.
- Boullis, A., Francis, F., Verheggen, F.J., 2015. Climate change and tritrophic interactions: Will modifications to greenhouse gas emissions increase the vulnerability of herbivorous insects to natural enemies? *Environ. Entomol.* 44 (2), 277–286. <https://doi.org/10.1093/ee/nvu019>.
- CABI, 2019. *Aphis gossypii*. Invasive species Compend. URL <http://www.cabi.org/isc/datasheet/6204> (accessed 11.29.20).
- Carmo-Sousa, M., Moreno, A., Garzo, E., Fereres, A., 2014. A non-persistently transmitted-virus induces a pull-push strategy in its aphid vector to optimize transmission and spread. *Virus Res.* 186, 38–46. <https://doi.org/10.1016/j.virusres.2013.12.012>.
- Chen, F., Ge, F., Parajulee, M.N., 2005. Impact of Elevated CO₂ on Tri-Trophic Interaction of *Gossypium hirsutum*, *Aphis gossypii*, and *Leis axyridis*. *Environ. Entomol.* 34 (1), 37–46. <https://doi.org/10.1603/0046-225X-34.1.37>.
- Chen, Y., Serateyn, L., Wang, Z., He, K., Francis, F., 2019. Reduction of plant suitability for corn leaf aphid (Hemiptera: Aphididae) under elevated carbon dioxide condition. *Environ. Entomol.* 48, 935–944. <https://doi.org/10.1093/ee/nvz045>.
- Chen, F.J., Wu, G., Parajulee, M.N., Ge, F., 2007. Impact of elevated CO₂ on the third trophic level: A predator *Harmonia axyridis* and a parasitoid *Aphidius picipes*. *Biocontrol Sci. Technol.* 17 (3), 313–324. <https://doi.org/10.1080/09583150701211814>.
- Coviella, C.E., Trumble, J.T., 1999. Effects of elevated atmospheric carbon dioxide on insect-plant interactions. *Conserv. Biol.* 13 (4), 700–712. <https://doi.org/10.1046/j.1523-1739.1999.98267.x>.
- Dáder, B., Fereres, A., Moreno, A., Trębicki, P., 2016. Elevated CO₂ impacts bell pepper growth with consequences to *Myzus persicae* life history, feeding behaviour and virus transmission ability. *Sci. Rep.* 6, 19120. <https://doi.org/10.1038/srep19120>.
- De Paulo, P.D., Pereira, E.J.G., Oliveira, E.E., Fereres, A., Garzo, E., 2020. Indirect effect of elevated CO₂ concentration on *Bemisia tabaci* MEAM1 feeding on Bt soybean plants. *J. Appl. Entomol.* 1–11. <https://doi.org/10.1111/jen.12822>.
- Dixon, A.F.G., 1987. Parthenogenetic reproduction and the rate of increase in aphids. In: Minks, A.K., Harrewijn, P. (Eds.), *Aphids: Their Biology, Natural Enemies and Control*. Elsevier, Amsterdam, pp. 269–287.
- Douglas, A.E., 2003. The nutritional physiology of aphids. *Adv. Insect Phys.* 31, 73–140. [https://doi.org/10.1016/S0065-2806\(03\)31002-1](https://doi.org/10.1016/S0065-2806(03)31002-1).
- EPPO, 2020. Safe use of biological control. List of biological control agents widely used in the EPPO region. Standard PM6/3, 2020 Version. EPPO mBull. URL https://www.eppo.int/media/uploaded_images/RESOURCES/eppo_standards/pm6/pm6-03-2020-en.pdf (accessed 8.20.20).
- Facey, S.L., Ellsworth, D.S., Staley, J.T., Wright, D.J., Johnson, S.N., 2014. Upsetting the order: how climate and atmospheric change affects herbivore-enemy interactions. *Curr. Opin. Insect Sci.* 5, 66–74. <https://doi.org/10.1016/j.cois.2014.09.015>.
- FAO, 2016. *The State of Food and Agriculture. Climate change, agriculture and food security*. FAO, Rome, Italy.
- FAO, 2021. FAOSTAT. URL <http://www.fao.org/faostat/en/#data/QC> (accessed 2.6.21).
- Fereres, A., Lister, R.M., Araya, J.E., Foster, J.E., 1989. Development and reproduction of the English grain aphid (Homoptera: Aphididae) on wheat cultivars infected with *Barley yellow dwarf virus*. *Environ. Entomol.* 18, 388–393. <https://doi.org/10.1093/ee/18.3.388>.
- Flood, J., 2010. The importance of plant health to food security. *Food Secur.* 2 (3), 215–231. <https://doi.org/10.1007/s12571-010-0072-5>.
- Himanen, S.J., Nerg, A.M., Holopainen, J.K., 2009. Degree of herbivore feeding damage as an important contributor to multitrophic plant-parasitoid signaling under climate change. *Plant Signal. Behav.* 4 (3), 249–251. <https://doi.org/10.4161/psb.4.3.7958>.
- Hoover, J.K., Newman, J.A., 2004. Tritrophic interactions in the context of climate change: a model of grasses, cereal Aphids and their parasitoids. *Glob. Chang. Biol.* 10, 1197–1208. <https://doi.org/10.1111/j.1365-2486.2004.00796.x>.
- Hughes, L., Bazzaz, F.A., 2001. Effects of elevated CO₂ on five plant-aphid interactions. *Entomol. Exp. Appl.* 99 (1), 87–96.
- Hullé, M., Cœur d'Acier, A., Bankhead-Dronnet, S., Harrington, R., 2010. Aphids in the face of global changes. *Comptes Rendus - Biol.* 333 (6-7), 497–503. <https://doi.org/10.1016/j.crvi.2010.03.005>.
- IPCC, 2013. *Climate Change 2013: The Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change, Climate Change 2013: The Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate*

- Change. Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.
- IPCC, 2018. Summary for Policymakers, in: Masson-Delmotte, V., Zhai, P., Pörtner, H.-O., Roberts, D., Skea, J., Shukla, P.R., Pirani, A., Moufouma-Okia, W., Péan, C., Pidcock, R., Connors, S., Matthews, J.B.R., Chen, Y., Zhou, X., Gomis, M.I., Lonnoy, E., Maycock, T., Tignor, M., Waterfiel, T. (Eds.), Global Warming of 1.5°C. An IPCC Special Report on the Impacts of Global Warming of 1.5°C above Pre-Industrial Levels and Related Global Greenhouse Gas Emission Pathways, in the Context of Strengthening the Global Response to the Threat of Climate Change., World Meteorological Organization, Geneva, Switzerland, p. 32.
- Jacas, J., Urbaneja, A., Viñuela, E., 2006. History and future of introduction of exotic arthropod biological control agents in Spain: a dilemma? *BioControl* 51 (1), 1–30. <https://doi.org/10.1007/s10526-005-5808-3>.
- Jiang, S., Liu, T., Yu, F., Li, T., Parajulee, M.N., Zhang, L., Chen, F., 2016. Feeding behavioral response of cotton aphid, *Aphis gossypii*, to elevated CO₂: EPG test with leaf microstructure and leaf chemistry. *Entomol. Exp. Appl.* 160 (3), 219–228. <https://doi.org/10.1111/eea.12475>.
- Johnson, S.N., Ryalls, J.M.W., Karley, A.J., 2014. Global climate change and crop resistance to aphids: contrasting responses of lucerne genotypes to elevated atmospheric carbon dioxide. *Ann. Appl. Biol.* 165 (1), 62–72. <https://doi.org/10.1111/aab.12115>.
- Klaiber, J., Dorn, S., Najar-Rodriguez, A.J., 2013a. Acclimation to elevated CO₂ increases constitutive glucosinolate levels of *Brassica* plants and affects the performance of specialized herbivores from contrasting feeding guilds. *J. Chem. Ecol.* 39 (5), 653–665. <https://doi.org/10.1007/s10886-013-0282-3>.
- Klaiber, J., Najar-Rodriguez, A.J., Dialer, E., Dorn, S., 2013b. Elevated carbon dioxide impairs the performance of a specialized parasitoid of an aphid host feeding on *Brassica* plants. *Biol. Control* 66 (1), 49–55. <https://doi.org/10.1016/j.biocontrol.2013.03.006>.
- Lamichhane, J.R., Barzman, M., Booiij, K., Boonekamp, P., Desneux, N., Huber, L., Kudsk, P., Langrell, S.R.H., Ratnadass, A., Ricci, P., Sarah, J.-L., Messéan, A., 2015. Robust cropping systems to tackle pests under climate change. A review. *Agron. Sustain. Dev.* 35 (2), 443–459. <https://doi.org/10.1007/s13593-014-0275-9>.
- Li, X., Meng, L., Xing, G., Li, B., 2016. Constitutive and induced resistance in soybean interact to affect the performance of a herbivore and its parasitoid. *Biol. Control* 101, 145–151. <https://doi.org/10.1016/j.biocontrol.2016.07.008>.
- Martinou, A.F., Wright, D.J., 2007. Host instar and host plant effects on *Aphidius colemani*. *J. Appl. Entomol.* 131 (9–10), 621–624. <https://doi.org/10.1111/j.1439-0418.2007.01220.x>.
- Mattson, W. J., 1980. Herbivory in relation to plant nitrogen content. *Annu. Rev. Ecol. Syst.* 11 (1), 119–161.
- Moreno-Delafuente, A., Morales, I., Garzo, E., Fereres, A., Viñuela, E., Medina, P., 2021. Changes in melon plant phytochemistry impair *Aphis gossypii* growth and weight under elevated CO₂. *Sci. Rep.* 11, 2186. <https://doi.org/10.1038/s41598-021-81167-x>.
- Murray, T.J., Ellsworth, D.S., Tissue, D.T., Riegler, M., 2013. Interactive direct and plant-mediated effects of elevated atmospheric [CO₂] and temperature on a eucalypt-feeding insect herbivore. *Glob. Chang. Biol.* 19 (5), 1407–1416. <https://doi.org/10.1111/gcb.12142>.
- Myers, S.S., Smith, M.R., Guth, S., Golden, C.D., Vaitla, B., Mueller, N.D., Dangour, A.D., Huybers, P., 2017. Climate change and global food systems: potential impacts on food security and undernutrition. *Annu. Rev. Public Health* 38 (1), 259–277. <https://doi.org/10.1146/annurev-publhealth-031816-044356>.
- Newman, J.A., Gibson, D.J., Parsons, A.J., Thornley, J.H.M., 2003. How predictable are aphid population responses to elevated CO₂? *J. Anim. Ecol.* 72, 556–566. <https://doi.org/10.1046/j.1365-2656.2003.00725.x>.
- Ode, P.J., Johnson, S.N., Moore, B.D., 2014. Atmospheric change and induced plant secondary metabolites - Are we reshaping the building blocks of multi-trophic interactions? *Curr. Opin. Insect Sci.* 5, 57–65. <https://doi.org/10.1016/j.cois.2014.09.006>.
- Oehme, V., Högy, P., Zebitz, C.P.W., Fangmeier, A., 2013. Effects of elevated atmospheric CO₂ concentrations on phloem sap composition of spring crops and aphid performance. *J. Plant Interact.* 8 (1), 74–84. <https://doi.org/10.1080/17429145.2012.736200>.
- Oerke, E.C., 2006. Crop losses to pests. *J. Agric. Sci.* 144 (1), 31–43. <https://doi.org/10.1017/S0021859605005708>.
- Rosenblatt, A.E., Schmitz, O.J., 2016. Climate change, nutrition, and bottom-up and top-down food web processes. *Trends Ecol. Evol.* 31 (12), 965–975. <https://doi.org/10.1016/j.tree.2016.09.009>.
- Roth, S.K., Lindroth, R.L., 1995. Elevated atmospheric CO₂: effects on phytochemistry, insect performance and insect-parasitoid interactions. *Glob. Chang. Biol.* 1, 173–182. <https://doi.org/10.1111/j.1365-2486.1995.tb00019.x>.
- Ryalls, J.M.W., Moore, B.D., Riegler, M., Bromfield, L.M., Hall, A.A.G., Johnson, S.N., Raubenheimer, D., 2017. Climate and atmospheric change impacts on sap-feeding herbivores: a mechanistic explanation based on functional groups of primary metabolites. *Funct. Ecol.* 31 (1), 161–171. <https://doi.org/10.1111/1365-2435.12715>.
- Ryan, G.D., Shukla, K., Rasmussen, S., Shelp, B.J., Newman, J.A., 2014. Phloem phytochemistry and aphid responses to elevated CO₂, nitrogen fertilization and endophyte infection. *Agric. For. Entomol.* 16 (3), 273–283. <https://doi.org/10.1111/afe.12055>.
- Ryan, G.D., Sylvester, E.V.A., Shelp, B.J., Newman, J.A., 2015. Towards an understanding of how phloem amino acid composition shapes elevated CO₂-induced changes in aphid population dynamics. *Ecol. Entomol.* 40 (3), 247–257. <https://doi.org/10.1111/een.12181>.
- Stacey, D.A., Fellowes, M.D.E., 2002. Influence of elevated CO₂ on interspecific interactions at higher trophic levels. *Glob. Chang. Biol.* 8, 668–678. <https://doi.org/10.1046/j.1365-2486.2002.00506.x>.
- Starý, P., 1975. *Aphidius colemani* Viereck: its taxonomy, distribution and host range (Hymenoptera: Aphididae). *Acta ent. Bohemoslov.* 72, 156–163.
- Stiling, P., Cornelissen, T., 2007. How does elevated carbon dioxide (CO₂) affect plant-herbivore interactions? A field experiment and meta-analysis of CO₂-mediated changes on plant chemistry and herbivore performance. *Glob. Chang. Biol.* 13 (9), 1823–1842. <https://doi.org/10.1111/j.1365-2486.2007.01392.x>.
- Sun, Y.C., Chen, F.J., Ge, F., 2009a. Elevated CO₂ changes interspecific competition among three species of wheat aphids: *Sitobion avenae*, *Rhopalosiphum padi*, and *Schizaphis graminum*. *Environ. Entomol.* 38, 26–34.
- Sun, Y.C., Jing, B.B., Ge, F., 2009b. Response of amino acid changes in *Aphis gossypii* (Glover) to elevated CO₂ levels. *J. Appl. Entomol.* 133, 189–197. <https://doi.org/10.1111/j.1439-0418.2008.01341.x>.
- Sun, Y.C., Feng, L., Gao, F., Ge, F., 2011a. Effects of elevated CO₂ and plant genotype on interactions among cotton, aphids and parasitoids. *Insect Sci.* 18, 451–461. <https://doi.org/10.1111/j.1744-7917.2010.01328.x>.
- Sun, Y.C., Yin, J., Chen, F.J., Wu, G., Ge, F., 2011b. How does atmospheric elevated CO₂ affect crop pests and their natural enemies? Case histories from China. *Insect Sci.* 18, 393–400. <https://doi.org/10.1111/j.1744-7917.2011.01434.x>.
- Torres, A.D.F., Bueno, V.H.P., Sampaio, M.V., de Conti, B.F., 2007. Tabela de Vida de Fertilidade de *Aphidius colemani* Viereck (Hymenoptera: Braconidae, Aphidiinae) em *Aphis gossypii* Glover (Hemiptera: Aphididae). *Neotropical Entomol.* 36 (4), 532–536. <https://doi.org/10.1590/S1519-566X2007000400009>.
- Tougeron, K., Brodeur, J., Le Lann, C., Baaren, ., 2020. How climate change affects the seasonal ecology of insect parasitoids. *Ecol. Entomol.* 45 (2), 167–181. <https://doi.org/10.1111/een.12792>.
- Trębicki, P., Vandegeer, R.K., Bosque-Pérez, N.A., Powell, K.S., Dader, B., Freeman, A.J., Yen, A.L., Fitzgerald, G.J., Luck, J.E., 2016. Virus infection mediates the effects of elevated CO₂ on plants and vectors. *Sci. Rep.* 6, 22785. <https://doi.org/10.1038/srep22785>.
- Trębicki, P., 2020. Climate change and plant virus epidemiology. *Virus Res.* 286, 198059. <https://doi.org/10.1016/j.virusres.2020.198059>.
- Trębicki, P., Däder, B., Vassiliadis, S., Fereres, A., 2017. Insect-plant-pathogen interactions as shaped by future climate: effects on biology, distribution and implications for agriculture. *Insect Sci.* 24, 975–989. <https://doi.org/10.1111/1744-7917.12531>.
- Van Emden, H.F., Harrington, R., 2017. *Aphids as crop pests*, 2nd ed. CAB International, Oxfordshire, UK.
- Vassiliadis, S., Plummer, K.M., Powell, K.S., Trębicki, P., Luck, J.E., Rochfort, S.J., 2016. The effect of elevated CO₂ and virus infection on the primary metabolism of wheat. *Funct. Plant Biol.* 43, 892–902. <https://doi.org/10.1071/FP15242>.
- Wilkinson, T.L., Douglas, A.E., 2003. Phloem amino acids and the host plant range of the polyphagous aphid, *Aphis fabae*. *Entomol. Exp. Appl.* 106, 103–113. <https://doi.org/10.1046/j.1570-7458.2003.00014.x>.
- Wyatt, L.J., White, P.F., 1977. Simple estimation of intrinsic increase rates for aphids and tetranychid mites. *J. Appl. Ecol.* 14 (3), 757. <https://doi.org/10.2307/2402807>.
- Xing, G., Zhang, J., Liu, J., Zhang, X., Wang, G., Wang, Y., 2003. Impacts of atmospheric CO₂ concentrations and soil water on the population dynamics, fecundity and development of the bird cherry-oat aphid *Rhopalosiphum padi*. *Phytoparasitica* 31 (5), 499–514. <https://doi.org/10.1007/BF02979743>.
- Zamani, A.A., Talebi, A., Fathipour, Y., Baniamiri, V., 2007. Effect of temperature on life history of *Aphidius colemani* and *Aphidius matricariae* (Hymenoptera: Braconidae), two parasitoids of *Aphis gossypii* and *Myzus persicae* (Homoptera: Aphididae). *Environ. Entomol.* 36, 263–271. <https://doi.org/10.1603/0046-225X-36.2.263>.