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Comparative study of two predatory mites *Amblyseius swirskii* Athias-Henriot and *Transeius montdorensis* (Schicha) by predator-prey models for improving biological control of greenhouse cucumber

M.M. Téllez[a], T. Cabello[b], M. Gámez[d,*], F.J. Burguillo[c], E. Rodríguez[a]

[a] IFAPA, La Mojonera- Centre, Almería, Spain
[b] Department of Biology and Geology, University of Almería, Almería, Spain
[c] Department of Physical Chemistry, University of Salamanca, Salamanca, Spain
[d] Department of Mathematics, University of Almería, Almería, Spain

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ABSTRACT

Suppression of the whitefly *Bemisia tabaci* (Gennadius) and the thrips *Frankliniella occidentalis* (Pergande) by the predatory mite *Amblyseius swirskii* Athias-Henriot on greenhouse cucumbers can be considerably affected by cooler conditions in winter. In this study, this well known mite was tested simultaneously with a more recent predatory mite *Transeius montdorensis* (Schicha), to find out which of them was better at controlling pests on cucumbers in winter in Mediterranean greenhouses. We developed a mathematical predator-prey model which involved releasing both predators with populations of the two naturally occurring pests in a greenhouse cucumber trial. T. *montdorensis* provided pest control that was similar to and as effective as that by *A. swirskii. T. montdorensis* exhibited higher populations than *A. swirskii*, specifically when climatic conditions were colder. However, as the weather became warmer, the *A. swirskii* population increased quickly. Therefore, releasing *T. montdorensis* in winter, followed with releases of *A. swirskii* in spring, may be a good pest control strategy for greenhouse cucumbers.

1. Introduction

Protected cultivations have rapidly expanded in many regions all over the world, particularly in those with mild winter conditions (Fernández et al., 2018). In this respect, the province of Almería (36°50´N 02°23´O) is a region of southern Spain with the biggest concentration of greenhouses in the Mediterranean Basin (>31,000 ha). Cucumber cultivation (*Cucumis sativus* L.) which is the third most abundant greenhouse vegetable in the region after tomato and sweet pepper (CAP, 2018) occupies around 5000 ha. The western flower thrips, *Frankliniella occidentalis* (Pergande) (Thysanoptera, Thripidae), and the whitefly, *Bemisia tabaci* Genn. (Hemiptera, Aleyrodidade), are the two most damaging pests in greenhouse production. Both not only cause direct damage to plants by feeding, but also inflict damage indirectly by transmitting viruses (Glass and González, 2012). Implementing Integrated Pest Management (IPM) techniques promotes the rational use of pesticides in greenhouses and uses a range of strategies, among which the use of augmentative biological control has successfully increased in Almería since 2007 (Ehler 2006; Pilkinton et al., 2012).

Use of commercially available predatory mites (Acari: Phytoseiidae) has gained in popularity within the context of IPM programmes as being one of the most environmentally safe and economically viable pest management methods in greenhouse crops (Calvo et al., 2014; Vila and Cabello, 2014; van Lenteren et al., 2018). Among predatory mites, the phytoseiid *Amblyseius swirskii* Athias-Henriot, is the primarily agent used in the biocontrol of whiteflies and thrips in a wide range of greenhouse crops, including cucumber. This predator attacks instars thrips larvae as well as whitefly eggs and crawlers, but not adults (Bolckmans et al., 2005; van Maanen and Janssen 2008; Calvo et al., 2012). Moreover, *Amblyseius swirskii* can also develop and reproduce on a variety of other food sources including pollen (Nguyen et al., 2013). In spring, biological pest control in greenhouses is a successful pest management strategy. However, during winter; using their natural enemies can be less effective since they may be affected by colder temperatures, shorter photoperiods and lower relative humidity (Shipp et al., 1996; 2009). This is especially true in cucumbers for thrips (Nomikou et al., 2002; Van Houten et al., 2010; Calvo et al., 2011;

* Corresponding author.
E-mail address: mgamez@ual.es (M. Gámez).

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Téllez 2015). Firstly, in Almería, thrips populations start to increase in greenhouse crops during winter (Rodríguez et al., 2018). Secondly, the inability of *A. swirskii* to build-up high populations during cooler conditions restricts their establishment (Shipp et al., 2009: Lee and Gillespie, 2011). Finally, for *A. swirskii* (Messelink et al., 2006) the lack of pollen in cucumber greenhouse varieties, which produce only female flowers, implies a shortage of non-prey food. Therefore, additional commercially available natural enemies need to be found that perform better in winter on greenhouse cucumbers.

Recently, the new predatory mite, *Transeius montdorensis* (Schicha) (Mesostigmata: Phytoseiidae), was identified as a suitable predator of thrips and whitefly in greenhouse crops (Steiner et al., 2003). This phytoseiid is native to the Neotropical region (Schicha, 1979) and has recently come onto the biocontrol market. In particular, it has been commercially available in Europe since 2004 and in Spain since 2017 (van Lenteren et al., 2018). *T. montdorensis* can consume more thrips per day than *A. swirskii*, and high oviposition has been achieved under low temperature and low light conditions (Steiner et al., 2003; Hatherly et al., 2004). Recent evidence of its efficacy in suppressing thrips in cucumber greenhouse at northern latitudes has been provided (Labbé et al., 2019). However, no comparative studies have been published yet on how both predatory mites (*A. swirskii* vs *T. montdorensis*) control pests under Mediterranean greenhouse conditions. Therefore, there has been growing interest in the performance of new biological control agents' like *T. montdorensis* under such conditions.

Mathematical models can be a useful tool for evaluating the effectiveness of multiple factors in biological control in an IPM strategy (Tang and Cheke, 2008; Tian et al., 2019). Several studies have been carried out in greenhouses which focused on modeling dynamic population of pests and their natural enemies (Lloret-Climent et al., 2014), or involved tri-trophic interactions (Sánchez et al., 2018). Here, we use the simple three-species Lotka-Volterra model, which seemed to be a good option in the simplified environmental conditions of a greenhouse (Varga et al., 2010; Molnár et al., 2016). New applications of these models have also been used in a biological control context in a variety of different situations. For instance, instead of a simple proportional conversion of prey- predator, numerical responses could be calculated from appropriate functional responses. However, in our case, the interaction coefficients in the classical Lotka-Volterra model we use could be considered as being the average slopes of the functional and numerical responses, respectively. However, to develop a more accurate model which includes functional and numerical responses, further trials will be needed so that we can design better fits for these responses. Furthermore, in one-predator, two-prey models the optimal foraging approach may also provide a more precise model (see e.g. Stephens and Krebs (1986)).

Therefore, the objective of this study was to make comparisons by modeling the populations of two pests, whitefly and thrips, and two predators, *T. montdorensis* and *A. swirskii*, in order to determine which predator was more efficient in winter for cucumbers in Mediterranean greenhouses.

2. Material and methods

2.1. Experiment design

The trial was conducted from mid-November 2016 to end-March 2017 in an experimental greenhouse with a surface area of 960 m^2 at the IFAPA Research Institute "La Mojonera" (Almería, Spain, latitude 36° 45′N, longitude 2° 42′W). Cucumber seedlings (*Cucumis sativus* L.) from the variety Cosaco® (Fitó, Spain) were planted on 17th November 2016 in perlite bags with a density of 2 plants m^{-2}, in a type of semi-closed hydroponic system.

The predatory mites were released 6 weeks after planting, on 27th December 2016. The mites, *A. swirskii* and *T. montdorensis,* were supplied by Bioline AgroSciences Ltd as a commercial product consisting in sachet-based controlled-release systems containing 250 mites (all stages). One sachet per 2 plants (doses 125 ind/m2) were hung at an average height and protected from direct sunlight. The experiment had a randomized block design with two replications and one factor (predator species) as treatment (with 2 levels, *A. swirskii* and *T. montdorensis*). The replicate plots were four 15 m rows, spaced 150 cm apart. This distance was reported to be enough to limit *A. swirskii* dispersal when plants were not in contact (Buitenhuis et al., 2009; López et al., 2017). Naturally occurring pest populations could migrate between plots during this experiment in which no chemicals treatments against pests were used.

2.2. Sampling

Sampling of pests and predatory mites was initiated 7 days after the predators were released. Six fully grown leaves were sampled from six interspersed plants per treatment at 7 day intervals for 13 consecutive weeks, until 30th March. The predators and pests were assessed in the laboratory using a stereo microscope (Zeiss Stemi 2000-C, Carlzeiss Germany). All stages of predatory mites, including eggs, juveniles or adults, were counted in each treatment. As for the pests, only the eggs and larvae of the whiteflies, and those of the thrips were included in the analysis because they were the susceptible stages to predation by the predatory mites. In addition, the natural occurrence of adult stages of whitefly and thrips was monitored throughout the trial (eight weekly samples) by counting captures on fourteen 25 × 10 cm yellow sticky traps (average = 15 traps/ ha) (Agrobio S.L. La Mojonera, Almería, Spain) distributed uniformly and placed at the same height as the crop, and these were raised in tandem with the crop growth.

2.3. Data analysis

The numbers of pests and predatory mites were expressed as insect-day accumulated values (IDA). This index, proposed by Ruppel (1983), was applied to evaluate the total pest impact over a given time period. It was also used to evaluate the effect of biological pest control (e.g.: Sánchez and Lacasa, 2008; Cabello et al., 2012). Due to the non-random design, IDA and mean number of eggs per leaf laid by both predatory mites were subject to statistical analysis with generalized linear models (e.g. see Millar and Anderson, 2004; Semenov et al., 2013). For the statistical analyses, the models were fitted using maximum quasi-likelihood estimation (IBM, 2017) with the GenLin procedure with gamma errors and the log link function for IDA and Poisson errors and the log link function for the egg number per leaf using the IBM SPSS version 25.0 statistical software package. The significance of the model was assessed by an Omnibus test (to test whether the explained variance in a dataset is significantly greater than the unexplained variance, overall).

2.4. Mathematical model

Among the non-stage-structured multispecies models, in the first study we decided to apply the simplest classical Lotka-Volterra one in which each single-species dynamics is Malthusian (meaning an increase in prey populations and decrease in predators). A more precise model would be obtained with logistic rather than Malthusian dynamics (see e.g. Scudo and Ziegler, 2013). However, here, predator-prey interaction was just proportional to the product of densities, as in the original Lotka-Volterra model.

Previous results based on thrip surveys carried out in Almería greenhouses show that *F. occidentalis* is particularly active in greenhouse crops throughout the winter season, from October to April (Rodríguez et al., 2018). Moreover, whitefly populations remain low in winter (Rodríguez et al., 2018). Therefore, the number of *F. occidentalis* captured by the yellow sticky traps was included in the model. Fig. 1 shows the network interactions used in our model according to the nomenclature used by Mills (2006), whose equations are shown below:

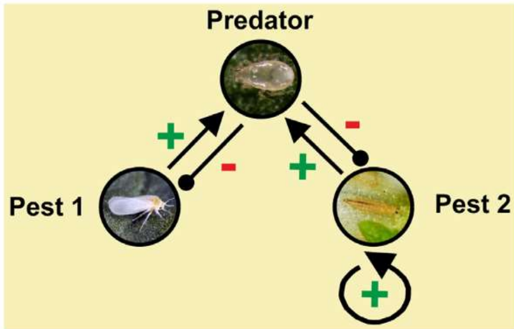

Fig. 1. Network of interactions considered in the mathematical model, the linking arrows and clubs show benefits (+) and losses (-). Predators species = *A. swirskii* or *T. montdorensis;* pests species: 1 for *B. tabaci* and 2 for *F. occidentalis*.

Pest 1(*B. tabaci*) $x'_1 = x_1(m_1 - \gamma_1 \cdot x_4)$
Pest 2(*F. occidentalis*) $x'_2 = x_2(m_2 - \gamma_2 \cdot x_4)$
Pest 3(*F. occidentalis* on yellow sticky traps) $x'_3 = x_3(m_3 - \gamma_3 \cdot x_4)$
Predatory species $x'_4 = x_4(-m_4 + \bar{\gamma}_1 \cdot x_1 + \bar{\gamma}_2 \cdot x_2 + \bar{\gamma}_3 \cdot x_3)$ (1)

where x_1, x_2, x_3 and x_4 are the densities (number / leaf) of pests and predator species, respectively. According to the terminology of Abrams (2012), m_1, m_2 and m_3 are the intrinsic growth rate of the pests; m_4 is the death rate of the predator in the absence of the prey; γ_1, γ_2 and γ_3 are the slopes of the predator's functional response on killing the pest species respectively; and$\bar{\gamma}_1$, $\bar{\gamma}_2$and $\bar{\gamma}_3$are the slopes of the predator's numerical response on killing and eating the pest species respectively. Using the statistical software SIMFIT version 2017 Bardsley, 2017), the system of equations ((1) was fitted to the data corresponding to the number of leaves.

3. Results

3.1. Effects of predators on populations of whitefly and thrips

The temporal dynamics of whitefly and thrips were very similar in both mite treatments, as indicated by the IDA values monitored on the leaves throughout the trial (Fig. 2a,b). Moreover, the increase in mite population corresponded to reductions in those of the whiteflies and thrips, thereby showing that both predators, *T. montdorensis* and *A. swirskii*, were good pest controllers (Fig. 2a,b). In fact, the predator species factor observed to neither effect the mite's IDA (Chi-square likelihood ratio = 3.176; df = 1; P = 0,750); nor the whitefly's (Chi-square likelihood ratio = 0.469; df = 1; P = 0.494); nor the thrip´s (Chi-square likelihood ratio = 3.082; df = 1; P = 0.790). In the MLGZ analysis, we found the mite species factor (Chi-square likelihood ratio = 15.041; df = 1; $P < 0.0001$) and the sampling factor (Chi-square likelihood ratio = 2104.335; df = 12; $P < 0.0001$) had significant effects. Thus, for the sampling period, the values of the number of eggs per leaf are shown in Fig. 3 for both predatory mite species; the mean values estimated by statistical analysis were 4.04 ± 0.29 egg/leaf for *T. montdorensis* which were significantly higher than the 2.54 ± 0.20 found for *A. swirskii*.

3.2. Predator response to prey abundance

The dynamic populations of both mites, *T. mondorensis* and *A. swirskii*, was well simulated by the models, with the predicted number of both predatory mites very close to those observed (R^2 prediction =0.919 and 0.926 for *T. mondorensis* and *A. swirskii*, respectively) (Fig. 4a,b) (Table 1). The models also provided a highly accurate simulation of the dynamics of the two pest species (whitefly and thrips) both over time and in terms of numbers in both treatments (Fig. 4a,b) (Table 1). The migrant adult thrips in the greenhouses, captured by the yellow sticky traps, was also well simulated (Fig. 4a,b) (Table 1). The model results showed that both predators controlled increases in whitefly and thrip populations, and eventually suppressed both pests. In the middle of the crop cycle, particularly in the period between 40 and 60 days when the weather was colder, *T. montdorensis* showed higher populations than *A. swirskii* (Fig. 4a) and the former actually had a lower death rate in the absence of prey (Table 1). However, as the weather became warmer, *A. swirskii* populations increased quickly (Fig. 4b). Overall, with the treatment with *A. swirskii* there was a lower growth rate in whitefly populations (Table 1). Similarly, the growth rate in thrips was slightly lower with the treatment with *A. swirskii* than that with *T. montdorensis* (Table 1).

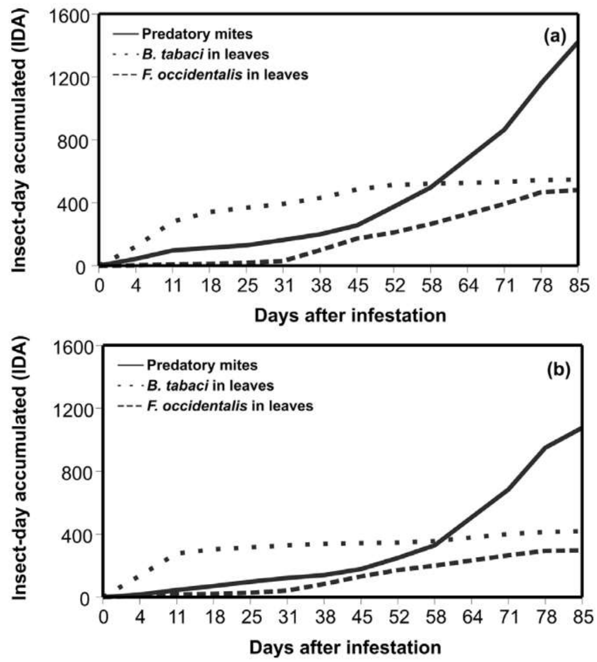

Fig. 2. Insect-day accumulated values (IDA) for the two pest species, whitefly and thrips, in greenhouse cucumber crop according to treatment: (a) *T. montdorensis* or (b) *A. swirskii* releases.

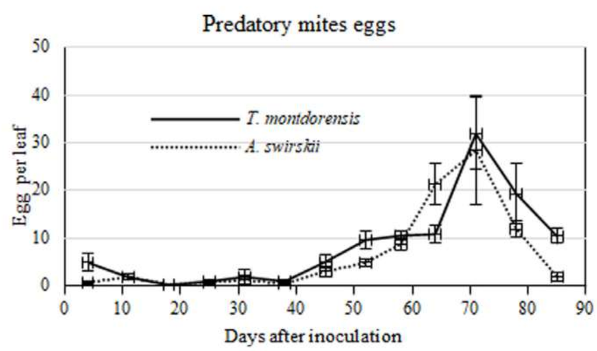

Fig. 3. Mean number of eggs per leaf laid by *T. montdorensis* and *A. swirskii* throughout the trial.

4. Discussion

In this research, we investigated whether the use of the predatory mite *Transeius montdorensis* in the biological control of two greenhouse pests, whitefly and thrips, resulted in better control than that carried out by the mite *Amblyseius swirskii*. Our results showed that both of them were equally effective predators on cucumbers in winter in

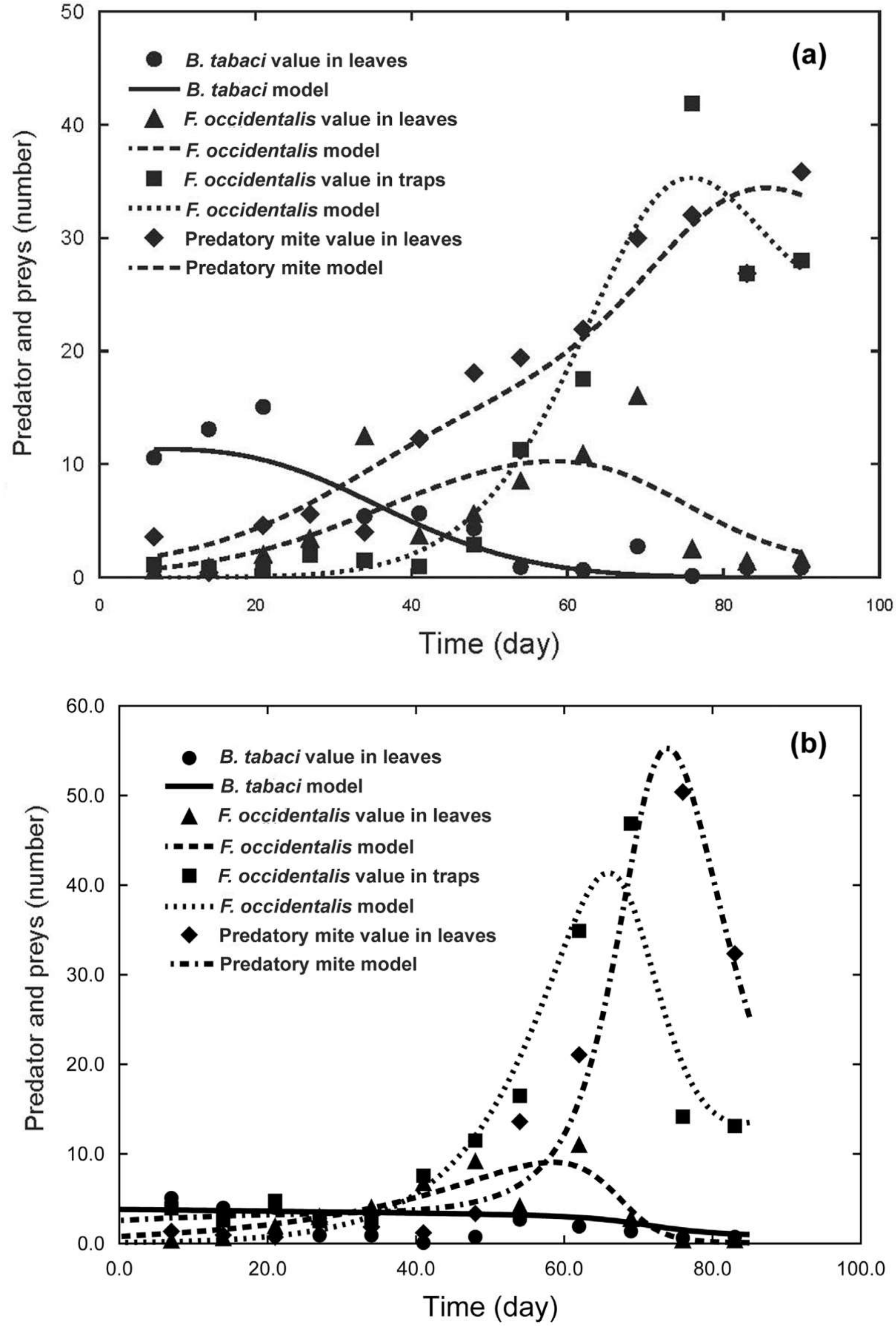

Fig. 4. Densities obtained from the fitted model for two pest species, whitefly and thrips, in greenhouse cucumber crops according to treatment: (a) *T. montdorensis* or (b) *A. swirskii* releases.

Mediterranean greenhouse conditions. There were no significant differences between the IDA value in the *T. montdorensis* and *A. swirskii* treatments. This was also true with the IDA values for whitefly and thrips between the two mite treatments. Moreover, the presence of these mites reduced whitefly and thrip abundance. Overall, these results indicate that each mite successfully controlled whitefly and thrip populations. Few studies on the density and predation of *T. montdorensis* on *B. tabaci* and *F. occidentalis* have been reported. For instance, our results confirmed previous findings by Labbé et al. (2019) in greenhouse cucumbers by demonstrating that this mite is a good predator of thrips in winter, similar to other phytoseiid mites such as *A. swirskii* and *Amblydromalus limonicus,* and even better than *Neoseiulus*

Table 1
Fitting and statistical parameters for fitted model for two pest species, whitefly and thrips, in greenhouse cucumber crops according to treatment: (a) *T. montdorensis* or (b) *A. swirskii* releases.

Predator	Fitting parameters (average ± SE)										Statistical parameters		
	m_1	m_2	m_3	m_4	γ_1	γ_2	γ_3	$\tilde{\gamma}_1$	$\tilde{\gamma}_2$	$\tilde{\gamma}_3$	d.f.	R^2	P
(a)	0.0138	0.0977	0.2224	0.0508	0.0066	0.0051	0.0072	1.4848	0.9216	0.1667	10	0.9191	<0.05
	(0.005)	(0.009)	(0.011)	(0.016)	(0.002)	(0.001)	(0.0007)	(0.002)	(0.002)	(0.0002)			
(b)	0.0018	0.0679	0.1129	0.2392	0.0012	0.0067	0.0039	0.0669	000,002	0.0056	10	0.9257	<0.05
	(0.004)	(0.036)	(0.004)	(0.161)	(0.001)	(0.008)	(0.0005)	(0.044)	(0.0006)	(0.0018)			

cucumeris. There have been similar findings in ornamental crops, in which it was one of the natural enemies analysed for controlling thrips and seen to be one of the best pest controllers (Manners et al., 2013). As for controlling the whiteflies species (*B. tabaci* and *Trialurodes vaporarium* (Westwood)) in poinsettia plants, it showed it was similarly effective as the parasitic wasp *Encarsia formosa*, and more so than *A. limonicus* (Richter, 2017).

Moreover, the model outcomes showed that it coped with winter environmental conditions the best. In fact, we recorded significant differences in the number of eggs laid by both predators which depending on the sampling period and these differences were higher between 40- 60 days into the trial, which directly corresponded to the dates 7 −27 February. This period of time was characterised by low relative humidity (RH). To be specific, we recorded over ten hours per day with a RH below 70% (data not shown). Furthermore, in the warmer conditions at the end of the trial, *A. swirskii* performed much better. In fact, their population did not grow in colder crop conditions, but showed high and fast growth in warmer weather. These results closely matched those reported by Clymans et al. (2017) in which seven predatory mite species were evaluated under different climatic conditions in strawberries. They showed that the warmest regime was that most adequate for populations of *A. swirskii* to grow. In addition, and as reported in other studies on greenhouse pests in Almería (Rodríguez et al., 2018), the outcomes of the models showed that whitefly abundance tended to be low in winter whereas thrips gradually increased in abundance in this period with a more marked population increase in spring. The model results (Table 1) showed that whiteflies exhibited lower population growth when *A. swirskii* was present, suggesting that, in general, it was the optimum predator for reducing the whitefly population, albeit it had a stagnant population in colder conditions. In conclusion, in winter, *T. montdorensis* was the only mite whose population grew significantly, but in warmer weather, *A. swirskii* was the most adequate predatory mite. These findings led to significant practical considerations since, it is likely that, seasonal and consecutive releases of the two predatory mite species (first *T. montdorensis* in autumn-winter and then *A. swirskii* in spring) will suppress both pests on cucumbers. Therefore, studies need to be made to determine whether seasonal alternation of the two predatory mites within the Mediterranean winter crop season could lead to enhanced pest control in cucumbers overall.

5. Conclusion

The two predatory phytoseiid mites, *Amblyseius swirskii* and *Transeius montdorensis*, were, generally speaking, good biological agents for whitefly and thrip control under Mediterranean greenhouse conditions. Nevertheless, *T. montdorensis* showed better growth capacity in the winter than did *A. swirskii*. However, as spring approached, *A. swirskii* was seen to be the best predator. Therefore, greenhouse pest control in the winter crop season may be greatly enhanced by combining seasonal and consecutive releases of *T. montdorensis* (in the autumn-winter) and *A. swirskii* (afterwards in spring) rather than releasing them individually.

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.

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