



Does the Lewis spider mite constitute a threat to agricultural crops in Europe? New data on occurrence, host plants and damage in the invaded areas in Portugal

P. Naves^{1,2} · M. Santos¹ · A. Aguiar³ · A. Migeon⁴ · D. Navia⁴ · P. Auger⁴

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Abstract

The Lewis spider mite (LSM), *Eotetranychus lewisi*, is an important pest of poinsettias (*Euphorbia pulcherrima*) and agricultural crops worldwide. In Europe it is a quarantine-regulated pest with established outdoor populations in Portugal. We investigated the pest status and host plants of the LSM in Madeira and the Algarve, collecting and inspecting plants and crops. The LSM was mainly found on naturalized poinsettias and castor-bean. Additionally, live mites were collected in very low numbers from plants of vine, cherimoya, mango, papaya, cherry plum, wild tobacco, and bladder vine, in direct contact or close to LSM-infested poinsettias, strongly suggesting these plants would not be breeding hosts. LSM abundance was low and no damage was observed. Mites were absent from strawberry, bramble and citrus, important hosts worldwide. Field surveys were complemented with a citizen-science experiment in outdoor conditions to test whether proximity to mite-infested poinsettias affected subsequent detections on poinsettia, strawberry, or grapevine. After two months of exposure to potentially dispersing LSM individuals and a further two months under controlled conditions to allow the mite to develop, LSM was detected on the three-recipient species but at low infestation rates, with a higher number of infested leaves and abundance on poinsettias, and absence of damages to crops. Overall, we conclude that the primary reservoirs for LSM populations in southwestern Europe are poinsettia and castor-bean plants, and our results suggest that the LSM is not causing damages to crops and may not be as harmful as expected, which can have implications to the current quarantine-regulation of this mite in Europe.

Keywords *Eotetranychus lewisi* · Invasive mite pest · *Euphorbia pulcherrima* · Plant hosts · Citizen-science

✉ P. Naves
pedro.naves@iniav.pt

¹ Instituto Nacional de Investigação Agrária e Veterinária, I.P. (INIAV), Oeiras, Portugal

² GREEN-IT Bioresources for Sustainability, ITQB NOVA, Oeiras, Portugal

³ Secretaria Regional de Agricultura, Pescas e Ambiente (SRAPA), Direção Regional de Agricultura e Desenvolvimento Rural, Direção de Serviços dos Laboratórios Agrícolas e Agroalimentares (DSLAA), Laboratório de Qualidade Agrícola (LQA), Madeira, Portugal

⁴ CBGP, INRAE, CIRAD, Institut Agro, IRD, Univ Montpellier, Montpellier, France

Introduction

The Lewis spider mite (LSM), *Eotetranychus lewisi* (McGregor 1943), is a polyphagous arthropod which has been recorded on more than 79 herbaceous and woody plant species belonging to 28 different families (Migeon and Dorkeld 2024), being considered a pest of economically important agricultural crops such as peach (*Prunus persica*), papaya (*Carica papaya*), strawberry (*Fragaria × ananassa*) and apple (*Malus domestica*) in different regions of the world. In Mexico, attacks reduce commercial quality and value of chayote (*Sechium edule*) fruits (Cadena-Iñiguez et al. 2024), while in Ecuador the LSM affects white carrot, *Arracacia xanthorrhiza* (Vásquez et al. 2017) and peach (Miño et al. 2022), among other local crops (Vásquez and Dávila 2018; Caiza 2021). In Northern Mexico, it is the most important pest of peach orchards (Pérez-Santiago et al. 2007), while in some regions of Chile it causes significant damage to vineyards, *Vitis vinifera* (Sazo et al. 2003). In El Salvador it affects papaya (Andrews and Poe 1980), while in coastal California it is considered a problematic pest in commercially cultivated organic strawberries and raspberries (*Rubus* spp.) (Howell and Daugovich 2013).

A probable native of tropical Central America (Doucette 1962), the LSM has been dispersed most likely through the international trade of poinsettia plants (*Euphorbia pulcherrima* Willd. ex Klotzsch), being now found in several countries of North, Central and South America, Africa, Asia and Europe (EFSA PLH Panel 2017). On the European continent, the mite is a quarantine-regulated pest listed in the Annex IIA of the EU Regulation (EU) 2019/2072 and in the A1 EPPO list. Nevertheless, despite sanitary regulations, outbreaks in greenhouses and nurseries (usually associated with imported potted plants of *E. pulcherrima*) have been reported from several European countries (EFSA PLH Panel 2017), which lead to subsequent destruction of infested and suspicious plants and treatments with phytosanitary products to eradicate the pest (Labanowski 2009; EFSA PLH Panel 2017; NPPO The Netherlands 2021). There are, however, established populations in outdoor conditions in Europe since 1988, when it was found in Madeira Island on plants of poinsettia and *Vitis* sp. (Carmona 1992). Subsequently, in 2019 the mite was also detected in the Algarve (southern mainland Portugal) (Naves et al. 2021) and, more recently, in the Lisbon District (Santos et al. 2023), always associated with outdoor poinsettia plants. Overall, in southwestern Europe the LSM can be considered an invasive non-native species (sensu Soto et al. 2024), as it is establishing new populations extending beyond the initial introduction point.

Although the LSM has been present for more than 36 years in Madeira Island, there are no reports of damages to agricultural crops (EFSA PLH Panel 2017; EPPO 2020), a situation that requires clarification. Likewise, nothing is known on the impact and pest status of this invasive non-native mite in the Algarve, where suitable climatic conditions and susceptible plant hosts coexist.

In the current study, we investigated the pest status of the invasive LSM in Southwestern Europe, both in the region where it was first detected almost four decades ago (Madeira Island), and in the Algarve where it was detected only a few years ago. We evaluated the adaptation and pest relevance of the LSM to agricultural crops of importance to Southern Europe, by sampling various plants that are likely hosts for the mite's detection according to EPPO (2006) and EFSA PLH Panel (2017). Studies included two trials, with field surveys in Madeira and in the Algarve to identify host plants (including non-cultivated and agricultural plants), and a complementary

experiment in outdoor conditions to evaluate the natural infestation of sentinel-plants by mite-infested donor poinsettias, within a collaborative citizen-science approach.

Materials and methods

Eotetranychus lewisi samplings

To evaluate the host plant range of the LSM, field surveys were carried out on two periods in May and October 2022. The first sampling focused on plants close to known LSM-infested poinsettias, with the second sampling focusing on plants in direct contact or immediately adjacent to infested poinsettias.

In May 2022, the first sampling focused on plants close to known LSM-infested poinsettias in Madeira Island and in the Algarve, based on locations provided by the national NPPO (DGAV) and on previous field observations. We collected leaf samples from the infested poinsettias and from other plants within a (maximum) 100-m radius, sampling cultivated and non-cultivated plants. In both regions, samples were collected in urban, peri-urban and rural environments, and included cultivated terrain, forest edges, settlements, roadsides, vacant lots and public and private gardens. Whenever possible, locations where phytosanitary chemical treatments were minimal or inexistent were selected. If present, plants with symptoms of mite attacks were selected. Surveyed plants included native and exotic species and agricultural crops, listed in Table 1. Whenever present in the sampled locations, plants of lemon (*Citrus limon*), sweet orange (*Citrus sinensis*), strawberry, peach, bramble (*Rubus* sp.) and vine were thoroughly sampled, as these are considered the most relevant host species for detection surveys of the LSM in the EU (EFSA 2023). Grasses (Poaceae) are common hosts for several species of Tetranychidae but were not sampled because the LSM has not been recorded on this plant family until now (Migeon and Dorkeld 2024). Samplings consisted of collecting 50 to 150 leaves from each plant species on each location.

A second sampling took place on Madeira Island in October 2022. As before, samplings centred on mite-infested poinsettias but this time we prioritized plants in direct contact or immediately adjacent (≈ 1 m apart) to infested poinsettias, to maximise the detection of the mite and determine if plants in close vicinity of LSM-infested poinsettias were at higher risk. Other nearby and distant plants (within a few meters to several kilometres) from infested poinsettias were also sampled. Plant collection focused on agricultural crops [particularly grapevines (Carmona 1992)] and uncultivated plants. This was possible because agriculture in Madeira has a small-scale, self-sustaining component where crops are frequently planted in gardens alongside poinsettias.

Mite extraction and identification

When mites were present in sufficient numbers, adults of both sexes were collected directly from leaves with a fine brush under a stereomicroscope. Otherwise, samples were processed by the method described in Boller (1984), with leaves placed in a bucket with water and a few drops of detergent. Buckets were closed and stirred then liquid contents were filtered (using various mesh sizes) after 24 h. Filter suspensions were observed under a stereomicroscope and mites collected.

Table 1 Host plants sampled for *Eotetranychus lewisi* in the Algarve and Madeira Island

Plant Family	Genus/species	Region (District/Autonomic region)		Proximity to LSM-infested <i>E. pulcherrima</i>	Nb of samples with <i>E. lewisi</i> /total number of samples
		Algarve	Madeira		
Amaranthaceae	<i>Amaranthus</i> sp.		X	Close to	0/1
	<i>Salsola vermiculata</i>	X		Far from	0/1
	<i>Sarcocornia perennis</i>	X		Far from	0/1
Anacardiaceae	<i>Suaeda vera</i>	X		Far from	0/1
	<i>Mangifera indica</i>		X	In touch with	1/1
	<i>Persea americana</i>		X	Close to	0/3
	<i>Pistacia lentiscus</i>	X		Far from	0/1
Annonaceae	<i>Annona cherimola</i>		X	In touch with	2/3
Apiaceae	<i>Eryngium</i> sp.	X		Close to	0/1
Apocynaceae	<i>Araujia sericifera</i>		X	In touch with	1/1
	<i>Nerium oleander</i>	X		Close to	0/1
Asparagaceae	<i>Plumeria rubra</i>	X		Close to	0/1
	<i>Asparagus albus</i>	X		Far from	0/1
Asteraceae	<i>Bidens pilosa</i>		X	Close to	0/1
	<i>Helichrysum stoechas</i>	X		Close to	0/1
	<i>Sonchus oleraceus</i>		X	Close to	0/1
	<i>Campsis radicans</i>	X		Close to	0/1
	<i>Abelia x grandiflora</i>	X		Close to	0/1
Caricaceae	<i>Carica papaya</i>		X	Close to	1/1
Clethraceae	<i>Clethra arborea</i>		X	Far from	0/1
Convolvulaceae	<i>Ipomoea batatas</i>		X	Close to	0/1
Ebenaceae	<i>Diospyros kaki</i>	X		Far from	0/1

Table 1 (continued)

Plant Family	Genus/species	Region (District/Autonomic region)		Proximity to LSM-infested <i>E. pulcherrima</i>	Nb of samples with <i>E. lewisii</i> /total number of samples
		Algarve	Madeira		
Euphorbiaceae	<i>Codiaeum variegatum</i>		X	In touch with /Close to	0/2
	<i>Euphorbia piscatoria</i>		X	Far from	0/2
	<i>Euphorbia pulcherrima</i>	X		n/a	5/7
	<i>Euphorbia pulcherrima</i>		X	n/a	17/19
	<i>Euphorbia</i> sp.	X		Close to	0/2
	<i>Ricinus communis</i>		X	In touch with	1/2
	<i>Ricinus communis</i>		X	Close to	4/4
	<i>Ricinus communis</i>		X	Far from	1/4
	<i>Ceratonia siliqua</i>	X		Close to	0/1
	<i>Cytisus striatus</i>		X	Far from	0/3
Fabaceae	<i>Genista tenera</i>		X	Far from	0/2
	<i>Ononis</i> sp.	X		Close to	0/1
	<i>Vigna unguiculata</i>		X	Close to	0/1
	<i>Centaurium erythraea</i>	X		Close to	0/1
	<i>Lavandula stoechas</i>	X		Close to	0/1
	<i>Salvia</i> sp.	X		Close to	0/1
	<i>Laurus novocanariensis</i>		X	Close to/Far from	0/2
	<i>Abutilon theophrasti</i>	X		Close to	0/1
	<i>Hibiscus rosa-sinensis</i>	X		Close to	0/2
	<i>Malva</i> sp.		X	Close to	0/2
Meliaceae	<i>Tilia</i> sp.	X		Close to	0/1
	<i>Azadirachta indica</i>	X		Close to	0/1

Table 1 (continued)

Plant Family	Genus/species	Region (District/Autonomic region)		Proximity to LSM-infested <i>E. putcherrima</i>	Nb of samples with <i>E. lewisii</i> /total number of samples
		Algarve	Madeira		
Moraceae	<i>Ficus benjamina</i>		X	Close to	0/1
	<i>Ficus carica</i>		X	Close to	0/1
Nyctaginaceae	<i>Ficus carica</i>			Far from	0/1
	<i>Mirabilis jalapa</i>	X		Close to	0/1
Myricaceae	<i>Myrica faia</i>	X	X	Far from	0/1
	<i>Psidium guajava</i>	X		Far from	0/1
Oxalidaceae	<i>Oxalis</i> sp.		X	Close to	0/2
	<i>Oxalis</i> sp.	X		Far from	0/1
Oleaceae	<i>Olea europaea</i>	X		Close to	0/1
Pittosporaceae	<i>Pittosporum tobira</i>	X		Close to	0/1
Plumbaginaceae	<i>Limoniastrum monopetalum</i>	X		Far from	0/1
	<i>Arundo donax</i>	X		Close to	0/1
Poaceae	<i>Eriobotrya japonica</i>	X		Far from	0/1
	<i>Fragaria x ananassa</i>			Close to	0/7
Rosaceae	<i>Malus domestica</i>		X	Close to	0/1
	<i>Prunus amygdalus</i>	X		Close to	0/1
	<i>Prunus amygdalus</i>			Far from	0/1
	<i>Prunus cerasifera</i>		X	Close to	0/1
	<i>Prunus persica</i>		X	Close to	1/1
	<i>Rosa</i> sp.		X	Close to	((1)/2)
	<i>Rosa</i> sp.	X		In touch with	0/1
	<i>Rosa</i> sp.	X		In touch with	(1)/1
	<i>Rubus</i> sp.	X		Close to	0/1
			X	In touch with /Closeto	0/2

Table 1 (continued)

Plant Family	Genus/species	Region (District/Autonomic region)		Proximity to LSM-infested <i>E. pulcherrima</i>	Nb of samples with <i>E. lewisii</i> /total number of samples
		Algarve	Madeira		
Rutaceae	<i>Citrus x aurantium</i>	X		Far from	0/1
	<i>Citrus limon</i>		X	Close to	0/1
	<i>Citrus medica</i>		X	Close to	0/1
	<i>Citrus sinensis</i>		X	Close to	0/1
	<i>Citrus</i> sp.		X	Close to	0/2
	<i>Citrus</i> sp.	X		Far from	0/1
Salicaceae	<i>Salix pedicellata</i>		X	Close to	0/2
Sapindaceae	<i>Cardiospermum grandiflorum</i>		X	Close to	0/1
Scrophulariaceae	<i>Myoporum tenuifolium</i>	X		Close to	0/1
Solanaceae	<i>Datura</i> sp.		X	Close to	0/1
	<i>Physalis</i> sp.	X		Far from	0/1
	<i>Solanum lycopersicum</i>		X	Close to	0/1
	<i>Solanum mauritanum</i>		X	Close to	0/1
	<i>Solanum mauritanum</i>		X	In touch with	1/1
	<i>Solanum nigrum</i>		X	Close to	0/2
Verbenaceae	<i>Lantana camara</i>	X		Close to	0/1
	<i>Verbena officinalis</i>		X	Close to	0/1
Viburnaceae	<i>Viburnum tinus</i>	X		Close to	(1)/2

Table 1 (continued)

Plant Family	Genus/species	Region (District/Autonomic region)		Proximity to LSM-infested <i>E. putcherrima</i>	Nb of samples with <i>E. lewisii</i> /total number of samples
		Algarve	Madeira		
Vitaceae	<i>Vitis vinifera</i>		X	In touch with	3/4
	<i>Vitis vinifera</i>		X	Close to	0/4
	<i>Vitis vinifera</i>	X		Close to	0/1
	<i>Vitis vinifera</i>		X	Far from	1/3
	<i>Vitis vinifera</i>	X		Far from	0/1

The terms “In touch with”, “Close to”, and “Far from” refer to plants either in direct contact (dark grey), within a few meters (light grey), or several kilometres distant from mite-infested poinsettias (no colour), respectively (see Materials and Methods). Numbers in single brackets refer to samples containing only dead *E. lewisii* mites, and double brackets to samples where no male individuals were collected. Species signalled in bold refer to the plant hosts most relevant for detection surveys in the EU according to EFSA (2023). *n/a* not applicable

Specimens were kept in 96% ethanol, subsequently cleared in 50% lactic acid for 24 h and slide-mounted in Hoyer's medium for phase contrast microscope observation (Leica DM750). Identification of mite species was based on morphological characters, using published descriptions of *E. lewisi* (original and re-descriptions) and those of morphologically closely related *Eotetranychus* species (McGregor 1943; Pritchard and Baker 1955; Meyer 1974; Baker and Tuttle 1994; EPPO 2006; Seeman et al. 2017; Gotoh and Arabuli 2019).

Plant colonization experiment

To study the possibility of *E. lewisi* movement from poinsettia to selected agricultural crops, we conducted an experiment in outdoor conditions during the summer of 2023 in Carcavelos and Oeiras parishes (Lisbon District), where the LSM had been previously detected (Santos et al. 2023). This trial involved the volunteer participation of several private garden owners within a collaborative citizen-engagement approach. We searched and detected LSM-infested poinsettia plants in outdoor conditions in urban and peri-urban areas, and contacted the garden owners to raise awareness for the study to be carried out, requesting their collaboration in its implementation. From a total of 36 garden owners identified, it was possible to collaborate with 24, and therefore a total of 24 mature poinsettia plants (over 1.5 m high) were selected as "donor" plants after confirming natural infestation with the LSM.

The young potted "sentinel" plants used for the experiment were purchased from certified private plant nurseries, and were analysed by direct observation using a hand magnifier (10x) to confirm they were free of mites and other pests. "Sentinel" plants were placed adjacent to the trunk of each mite-infested donor-poinsettia, directly under its canopy so that they were exposed to high potential colonisation by dispersing LSM. In each location (gardens) we installed one strawberry, one vine, and two poinsettias, totalling 24 strawberries, 24 vines and 48 poinsettias. Plants remained in the outdoor gardens for eight consecutive weeks in the summer months, from week 29 (mid-July) to week 37 (mid-September), being watered whenever necessary (typically daily), and were not subjected to any chemical treatment. Climatic data was retrieved from a nearby online-certified weather station located within a 3.5 km radius of all plants.

In September, LSM infestation level on donor poinsettias was evaluated. We randomly collected 15 leaves from each plant and observed them individually under a stereomicroscope, grading the mite population (adults only) in four levels on each leave as (i) no lewisi; (ii) 1–20 individuals; (iii) 20–100 individuals; (iv) more than 100 individuals.

Due to the low number of mites observed and the subsequent difficulty in counting them and ensuring the development of LSMs on vines and strawberries, the surviving sentinel-plants were isolated and transported to the INIAV's laboratories in Oeiras. There, they were kept isolated by species in a closed climatic chamber, with the pots separated by dishwashing liquid barriers to prevent movement between hosts, at 25 °C, 70% RH and 16:8 LD. After eight weeks, all leaves of the sentinel-plants were collected and individually observed under a stereomicroscope, and mite populations were graded as above for donor-plants. All tetranychid mites were collected, processed, and identified as described above.

Data analysis

A Kruskal–Wallis non-parametric analysis of variance test ($\alpha=0.05$), and subsequent post-hoc tests, were used to compare frequency of infestation and abundance of the LSM on recipient sentinel-plants (plant colonization experiment). Statistical analyses were performed using Statistica 12 software (StatSoft Inc. 2014).

Results

LSM samplings

In Madeira, 27 sites covering the whole island were surveyed in May 2022, totalling 24 plant species (some of which sampled more than once) from 14 families. In the Algarve, we sampled 43 plant species from 30 families in seven locations. The second survey in Madeira Island comprised 23 sites throughout the Island, with the sampling of 23 plant species from 14 families.

Results of the field surveys to identify host plants of the LSM are summarised in Table 1. The LSM was detected in several locations in Madeira and in the Algarve, primarily on naturalised poinsettias (Madeira and Algarve) and on Castor-bean, *Ricinus communis* (Madeira). Most of the sampled poinsettia plants were infested with the LSM, with 90% infestation in Madeira (17 samples out of 19) and 70% in the Algarve (5 out of 7).

Apart from poinsettias, 75 other plant species were sampled for a total of 129 samples (45 in the Algarve and 84 in Madeira). From these, we recovered LSM live individuals from eight plant species: *R. communis* (six locations), *V. vinifera* (four locations), *Annona cherimola* (two locations), and *Mangifera indica*, *C. papaya*, *Solanum mauritianum*, *Prunus cerasifera* and *Araujia sericifera* with one location each (Table 1).

Castor-bean plants appear to be a favourable host to the LSM, since in more than half of the cases (6 out of 10 samples), large colonies (with obvious damages and silk) were found on it. This was true not only when the castor-bean were proximal to infested poinsettia, but also on plants located distant to infested poinsettia in Madeira. Another plant appearing potentially favourable for the LSM is *V. vinifera*, as the mite was found on four occasions out of 13 samples processed. However, if we exclude vine plants in direct contact with infested poinsettias (three positive cases out of four samples), the LSM was only found on one occasion (one sample out of nine) on a vine stock located far from an infested poinsettia, while no specimens were found on five other vine stocks located near infested poinsettias.

We found other plants that may potentially be infested by the LSM. It is important to note that LSM individuals were found on *M. indica*, *A. cherimola*, *A. sericifera* and *S. mauritianum* only when the plants were in direct contact with a LSM-infested poinsettia. Only *C. papaya* and *P. cerasifera* were not in direct contact with a poinsettia but were in close proximity. In all cases, the density of the LSM was very low, immature stages were rare or absent, and no symptoms of mite infestation were apparent.

Due to the low densities observed (only a few specimens) and the absence of males (necessary for species-identification), it was not possible to identify the species of *Eotetranychus* present in one *P. persica* tree located near an infested poinsettia. However,

the morphological characteristics of the females are consistent with those of *E. lewisi*. It is also noteworthy that on two additional plants, we observed the remains (dead individuals) of *Eotetranychus* females, with morphological characteristics also corresponding to those of the LSM. These observations were made on Rose (*Rosa* sp.) in direct contact with an infested poinsettia and on Laurustinus (*Viburnum tinus*), on a plant located near an infested poinsettia and without live specimens detected. Finally, despite the plants being in contact with an infested poinsettia, no infestation was detected in two samples of fire croton (*Codiaeum variegatum*), of *Rubus* sp., and in one sample of *Rosa* sp.

Considering the plants indicated by EFSA (2023) as the most relevant for the detection of the LSM, no specimens were found on strawberry (seven samples “close to”), *Rubus* sp. (one sample “in touch” and one “close to”) and *Citrus* (six samples including five “close to”) (Table 1).

Plant colonization experiment

In the first phase (outdoor) of the plant colonization experiment, we sampled 360 leaves from the 24 donor-poinsettias, with all plants confirmed to be infested with the LSM. Mites were present in most (55%) of the sampled leaves, but usually in low numbers, with one to 20 adult mites per leaf (Fig. 1).

We did not recover sentinel plants from all locations, as some plants died due to water stress. In six of the locations all plants were dead, and in the remaining locations we were able to recover one, two or three live sentinel-plant species (Fig. 2). Overall, we recovered and examined 15 of the 24 strawberries, 18 of the 24 vines and 32 of the 48 poinsettias.

Fig. 1 Classes of abundance of *Eotetranychus lewisi* adult mites (as %) on leaves of infested donor-poinsettias (outdoor phase)

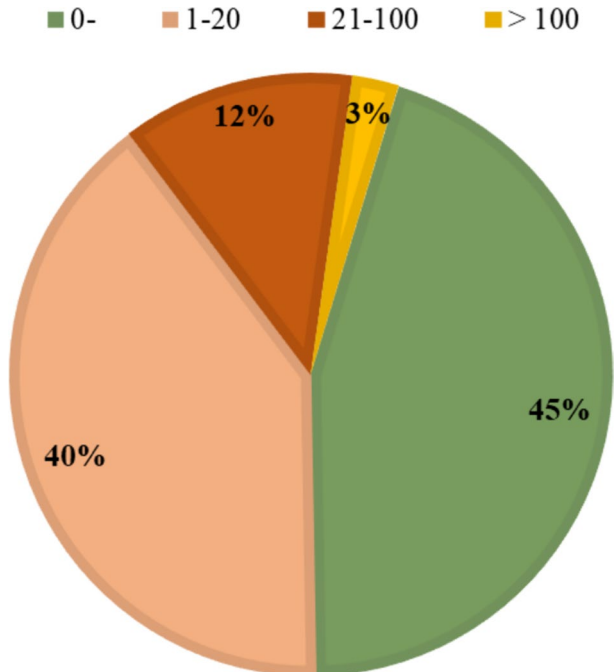
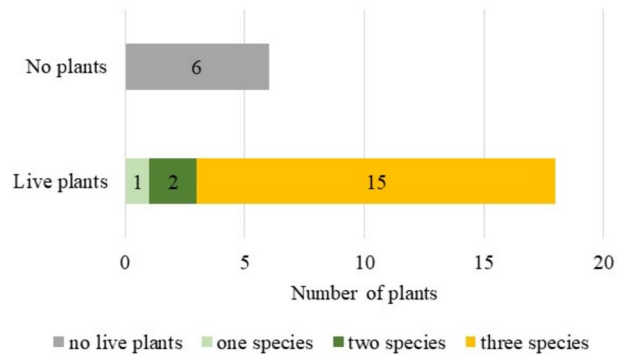


Fig. 2 Number of locations with no plants recovered (the three species were dead), and with one, two or three species of sentinel-plants recovered. N = 24 locations



During the outdoor phase, climatic conditions were suitable for mite development and breeding, with a mean temperature of 22 °C during the eight weeks (daily mean maximum of 27 °C and mean minimum of 19 °C), and with five days with precipitation above 1 mm, totalling 40 mm (Fig. 3).

After eight weeks of outdoor exposure followed by eight weeks in climatic room, the LSM was detected on all three species of sentinel-plants, with higher frequency in the poinsettias (10 out of 32 plants) and lower frequency in the strawberries and vines, where just four out of 15 plants and two out of 18 plants, respectively, were infested (Table 2), although the differences were not statistically significant (Kruskal–Wallis test: $\chi^2 = 2.562$; $df = 2$; $p = 0.278$). However, the number of infested leaves and the abundance of the LSM was significantly higher on poinsettias than on the crops (Kruskal–Wallis test: $\chi^2 = 88.580$; $df = 2$; $p = 0.001$). In general, mites were not frequent or abundant in the sentinel-plants

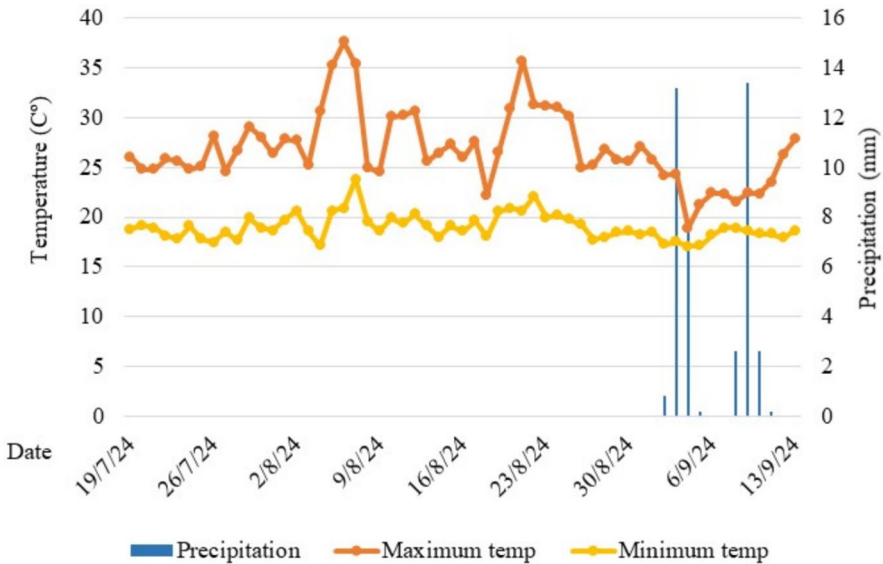


Fig. 3 Climatic conditions [daily maximum and minimum temperature (°C) and daily precipitation (mm)], of sentinel-plants subjected to natural infestation of *Eotetranychus lewisi* from mite-infested poinsettias (outdoor phase)

Table 2 Frequency of infestation and abundance of *Eotetranychus lewisi* and other arthropods on recipient sentinel-plant species after eight weeks of outdoor exposure and eight weeks of rearing in climatic room

Trap-plant species	Nb of live plants sampled	Nb of plants with <i>E. lewisi</i>	Nb of plants without <i>E. lewisi</i>	% of infested leaves (in infested plants only)	<i>E. lewisi</i> abundance in infested plants (nb of leaves)			Order/Families of mites and insects on the leaves	
					Without	Class I	Class II		Class III
<i>Euphorbia pulcherrima</i>	32	10	22	29a	103	35	7	0	Acaridae, Phytoseiidae, Tenuipalpidae, Tetranychidae, Tydeidae (Acar), Aphididae, Cicadellidae, Coccidae, Thysanoptera (Insecta)
<i>Fragaria × ananassa</i>	15	4	11	9b	109	11	0	0	Acaridae, Phytoseiidae, Tenuipalpidae, other Tetranychidae, Tydeidae (Acar), Aphididae, Cicadellidae, Coccidae, Thysanoptera (Insecta)
<i>Vitis vinifera</i>	18	2	16	2b	360	7	0	0	Eriophyiidae, Phytoseiidae, Tarsonemidae, Tenuipalpidae, other Tetranychidae, Tydeidae (Acar), Aleyrodidae, Aphididae, Cicadellidae, Coccidae, Thysanoptera (Insecta)

Abundance of *E. lewisi* (adults only) on each leaf graded as *Without*, *Class I*: 1–20 individuals, *Class II*: 20–100 individuals, *Class III*: more than 100 individuals. Significant differences (post-hoc test) after a Kruskal–Wallis non-parametric analysis of variance test ($\alpha=0.05$) are signalled with different letters for the percentage of infested leaves in infested plants

(Table 2) and obvious damages to the leaves were observed only in a few poinsettias and never on the crops.

In addition to *E. lewisi*, a diversified community of phytophagous mites and insects colonised and developed on the three sentinel-plant species, generally with high frequency and abundance (Table 2).

Discussion

We found the LSM to be established and spread in outdoor locations in Madeira and the Algarve, frequently associated with poinsettia and castor-bean plants exhibiting high infestation levels, resulting in chlorosis and visible damage. These findings support previous observations in the Lisbon District (Carcavelos and Oeiras parishes), where the LSM was also found associated with the same two plant hosts (Santos et al. 2023). Similar observations were reported by Caiza (2021), who surveyed different plant species in Ecuador and found the highest incidence of the LSM on poinsettia plants. Attacks on castor-bean are also not surprising, as these two plants belong to the same family and the LSM is known to readily infest this plant (Guanilo et al. 2012), on which populations increase as rapidly as on poinsettia (Doucette 1962). Castor-bean is a shrub native to Eastern Africa and present in many tropical and subtropical regions around the world, often behaving as a highly invasive species that can impact composition, richness and diversity of natural plant communities (Silva and Fabricante 2022). It is easy to find and particularly abundant in disturbed environments such as waste heaps, roadsides, forest edges and urban vacant lots (Martins et al. 2011), being a suitable breeding host for several species of non-native invasive mites (e.g., Ho et al. 2004; Guanilo et al. 2012; Ragusa et al. 2019; Naves et al. 2023). Considering the frequent association of the LSM with *R. communis*, we recommend this plant to be included in the list of priority plants for detection-surveys of *E. lewisi* within the EU.

The LSM was absent from most of the sampled agricultural crops, including several important Mediterranean and sub-tropical plants such as avocado, tomato, sweet potato, kaki, fig-tree, olive-tree, strawberry and citrus, even when the crops were in the vicinity of mite-infested poinsettias. According to the EFSA PLH Panel (2017), strawberry and raspberry are important potential pathways for dissemination and establishment of the LSM in the EU. The absence on strawberry in Madeira (where seven locations were sampled) is particularly relevant, as the LSM can be an important pest of this crop (Howell and Daugovich 2013) and *Citrus* (Doucette 1962). The absence of the LSM on these crops, which are the most relevant host plants for detection of the LSM in the EU (EFSA 2023), suggests that the LSM is not behaving as an economically important pest of strawberry or citrus in Madeira or the Algarve.

The LSM was detected in a few crops in Madeira but, except for one vine location distant from infested poinsettias, most detections were from plants in direct contact (touching) mite-infested poinsettias, with two additional detections from plants located very close to poinsettias. Furthermore, in all detections from agricultural plants mite populations were very low with just a few adult specimens collected or non-viable populations (i.e., just males). Therefore, it is not clear if the mites were developing on the crops or if their presence is an artefact of proximity to LSM-infested poinsettias. Indeed, the report of a plant hosting the LSM does not necessarily mean that the mite can complete its life cycle or cause economic damage, therefore we refrain from considering these crops and other

plant species where the mite was detected with low incidence and abundance (Table 1) as breeding hosts until further testing. As for vine, except for one distant plant, all the positive samples were plants directly touching poinsettias (with an additional nine samples where the LSM was absent), so current observations do not indicate that the mite is a pest of vines in south-western Europe.

Field samplings were complemented by the colonization experiments on the movement from infested hosts to nearby non-infested hosts under natural conditions. In this case, we confirmed that the LSM can naturally infest, develop and breed on strawberry and vine (besides poinsettia), which was expected considering the reports of the LSM on these crops in other countries (e.g., Sazo et al. 2003; Howell and Daugovish 2013). Nevertheless, it is worth mentioning the low number of poinsettias infested by LSM (about 1/3) after eight weeks of exposure to possible dispersing LSM, with a similar pattern observed for strawberry and grapevine, with only about 1/4 and 1/9 of infested plants, respectively. This indicates that infestation from plants containing colonies to nearby non-infested hosts is relatively infrequent and/or slow, even for preferred species such as poinsettias.

Furthermore, no damage to the sentinel-crops was observed and mite populations were low even after the prolonged period at favourable conditions (eight weeks at a mean temperature of 22 °C and eight weeks at 25 °C). According to Lai and Lin (2005), the LSM requires 19.8 days at 20 °C, 16.7 days at 24 °C and 13.2 days at 28 °C to complete one generation on poinsettia, which means that several consecutive generations could be completed on the three plant hosts during the 16-week experiment. The low infestation rate and population levels of LSM on the poinsettia and on the other sentinel plants raises questions about the biology, dispersal, colonization and development capabilities of this mite population, and on the suitability of climatic conditions in Southern Europe to promote LSM populations.

It is not clear why *E. lewisi* is not causing damage to agricultural crops in Madeira and in mainland Portugal, while economic damages are regularly reported from crops in other locations with comparable climatic conditions including Northern Mexico, California and Chile (e.g., Sazo et al. 2003; Pérez-Santiago et al. 2007; Howell and Daugovish 2013). Interspecific competition with the two spotted spider mite *T. urticae* may be locally preventing the LSM from developing high populations in Portugal, considering the abundance and widespread of *T. urticae* in local crops (Naves et al. 2021). According to Howell and Daugovish (2013), when *E. lewisi* and *T. urticae* occupy the same resource, *T. urticae* outcompetes and displaces *E. lewisi*. Likewise, a competitive displacement between the two spider mite species (with similar advantage to *T. urticae*) has also been suggested to occur in Mexico (Abato-Zárate 2011; Abato-Zárate et al. 2014). Moreover, it is also possible that the colonization of the sentinel-plants by a diverse and abundant community of competitive phytophagous insects, including aphids, whiteflies, leafhoppers, thrips and scale-insects, may deter establishment and development of *E. lewisi*, which deserves to be investigated with further detail. Another possibility is the effective control of Lewis mite populations by phytoseiid (Phytoseiidae) predators (Howell and Daugovish 2013), though on poinsettias and castor-oil plants with high populations of the LSM there was no sign of efficient control by natural enemies.

Another factor that could explain the apparent low invasiveness of *E. lewisi* that we report here could be linked to the genetic pool of the population at the origin of the settlement in Madeira and Portugal. The role of the genetic variability on the invasiveness of spider mites has already been put into evidence with the invasive tomato red mite *Tetranychus evansi* Baker & Pritchard, 1960. Indeed, using genetic markers (ITS and COI), Boubou et al. (2011) showed that *T. evansi* populations found in several parts of the world

belonged to two main genetic groups (clades) resulting from multiple complex invasion histories (Boubou et al. 2012). While the individuals of clade I (present in Africa, Asia, Europe, South America and USA) appears to have a higher invasive potential and are responsible for serious outbreaks on solanaceous crops (e.g. Oatman et al. 1967; Qureshi et al. 1969; Migeon 2007; Migeon et al. 2009), individuals of clade II are less dispersed (being present only in Europe and South America) and usually do not cause damages to solanaceous crops. One of the reasons for the invasive success of individuals from clade I that has been put into evidence is their greater tolerance and adaptability to variable habitats (Meynard et al. 2013), particularly in terms of increased cold tolerance (Migeon et al. 2015). Future molecular and biological studies comparing LSM American and European populations could clarify this.

Overall, our results suggest that the host range, dispersal, host colonization and population development of the LSM in its current European range is not as harmful as expected. Even though the LSM can attack and cause economic damage to different agricultural crops worldwide, we found that in outdoor conditions in south-western Europe, both in mainland Portugal and in Madeira Island, the LSM is almost exclusively found on poinsettia and castor-oil plants, and rarely on agricultural plants. This is particularly relevant for Madeira, where the LSM has been present for almost four decades with the potential to infest several crops that are widespread and important in southern Europe. When present on crops, such as grapevine in Madeira, the LSM exhibits very low population levels and does not cause significant damage. Several possible mechanisms could explain this pattern including genetics, presence of competitors, climate conditions, and predation, all of which merit further study. Furthermore, we show that under natural conditions in southern Portugal, the LSM does not appear to rapidly colonize or build significant populations in new host plants, even when favourable hosts are present. The apparent non-pest status of the LSM in Portugal is relevant to management programs throughout Europe, considering the quarantine-regulated classification of the Lewis mite and its status as a significant pest of several important agricultural crops worldwide.

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Data availability We declare all experimental data is being provided within this manuscript. The mite slides obtained during the current study are available in the INIAV Acarological collection (INIAV, Quinta do

Marquês, Oeiras, Portugal), and INRAE Acarological collections (Centre de Biologie pour la Gestion des Populations, 2018, CBGP—Continental Arthropod Collection, <https://doi.org/10.15454/D6XAKL>, France). Mite slides and non-treated experimental data can be made available on reasonable request.

Declarations

Competing interests The authors have no relevant financial or non-financial interests to disclose.

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