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Article

# First Record of *Eotetranychus libocedri* (McGregor, 1936) and *Eotetranychus thujae* (McGregor, 1950) (Acari: Tetranychidae) from Serbia

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## Abstract

During phytosanitary monitoring of ornamental conifers conducted across multiple regions of Serbia, two species of spider mite, *Eotetranychus libocedri* (McGregor, 1936) and *Eotetranychus thujae* (McGregor, 1950), were recorded for the first time. *E. libocedri* was found on *Platycladus orientalis* L. (Franco), *Thuja occidentalis* L., and *Cupressus × leylandii* A.B. Jacks & Dallim, while *E. thujae* was detected on *T. occidentalis*. These records have extended the distribution area of these two allochthonous species in Europe. Together with previous findings, the number of registered species of the Tetranychidae family in Serbia now totals 47 species across 10 genera.

**Keywords:** alien species; spider mites; cypress family; Europe

## 1. Introduction

Spider mites represent one of the most significant groups of pest mites in agriculture, forestry, and horticulture. The family Tetranychidae comprises 1,364 species of obligate phytophages that feed on more than 4,100 plant species [1]. Due to differences in host specificity, several spider mite species are closely associated with conifers; nearly 50 species from 12 spider mite genera feed on conifers of the cypress family (Cupressaceae). Five species of the genus *Eotetranychus* have been recorded on Cupressaceae plants, including *Eotetranychus cunninghamiae* Wang, *Eotetranychus juniperus* Tuttle & Baker, *Eotetranychus libocedri* (McGregor), *Eotetranychus pueblensis* (Tuttle) and *Eotetranychus thujae* (McGregor) [1]. *E. libocedri* and *E. thujae* are considered potential pests of coniferous plants. In contrast, *Oligonychus ununguis* (Jacobi) is regarded as the most harmful conifer-feeding spider mite, causing serious economic damage to ornamental conifers grown under the most unfavourable conditions of anthropogenic pressure, due to its high fecundity and rapid development. *O. ununguis* is a common species on conifers, but it is important to note that *E. libocedri* and *E. thujae* can also co-exist with it [2–6].

*E. libocedri* and *E. thujae* belong to the same guild of spider mites, exhibiting a strong trophic relationship with conifers, primarily members of the Cupressaceae family. According to available data, they are mostly found on conifers in urban environments and nurseries. Both species are native to North America, from where they have been introduced into Europe and Asia [1].

The species *E. libocedri* has been recorded worldwide on 16 coniferous plants across three families: Cupressaceae (*Calocedrus*, *Cupressus*, *Juniperus*, *Platycladus* and *Thuja*), Pinaceae (*Abies* and *Pinus*), and Tamaricaceae (*Tamarix*) [1]. Exceptionally, it was also found on one broad-leaved evergreen plant, *Euonymus fortunei*, family Celastraceae [7]. This species has been reported from

seven countries worldwide: United States, Mexico, Turkey, Hungary, Russian Federation, Kyrgyzstan and Uzbekistan. In Europe, its presence has been confirmed only in Hungary and the European parts of Turkey and Russia [7–17].

The species *E. thujae* has been recorded worldwide on 11 coniferous host plants, all belonging to the family Cupressaceae (*Chamaecyparis*, *Cupressus*, *Juniperus*, *Platyclusus*, and *Thuja* spp.) [1]. To date, the species has been reported from seven countries globally: United States, Canada, Georgia, Hungary, Kazakhstan, Kyrgyzstan and Russian Federation. In Europe, its presence has been confirmed only in Georgia, Hungary, and the European part of Russia [6,9,14,16,18–27].

Data on spider mites associated with conifers in Serbia are limited to a small number of published works [28–31] and relate solely to urban environments, while no data have been published on spider mites inhabiting conifers in natural habitats. Due to insufficient taxonomic research on spider mites associate with conifers in Serbia, there is no relevant data on the significance of existing and potentially harmful spider mite species on conifers.

This study contributes to the knowledge of the Serbian acarofauna by reporting spider mite species newly recorded on coniferous plants. These findings provide valuable insights into the distribution of alien species of spider mites on ornamental conifers in Europe and may serve as a basis for future assessment of their potential impact on conifers in Serbia and neighboring countries.

## 2. Materials and Methods

The research was conducted in August and September 2024 at 15 localities across different regions of Serbia as part of small-scale phytosanitary monitoring of ornamental conifers. A total of 35 samples were collected from 11 conifer species: *Chamaecyparis lawsoniana* (A. Murray bis) Parl., *Cupressus arizonica* Greene, *Cupressus* × *leylandii* A.B. Jacks & Dallim., *Juniperus chinensis* L., *J. horizontalis* Moech, *J. virginiana* L., *Platyclusus orientalis* L. (Franco), *Thuja occidentalis* L., *Picea abies* (L.) H. Karst., *P. glauca* (Moench) Voss, and *Taxus baccata* L.

Two spider mite species of the genus *Eotetranychus* were recorded on three conifer species (*Cupressus* × *leylandii* A.B. Jacks & Dallim., *Platyclusus orientalis* L. (Franco), and *Thuja occidentalis* L.) in seven samples collected from six locations in urban areas (Table 1).

**Table 1.** Sampling locations and plant species.

Sample code	Plant species	Locality		Date	Sample origin
		Geographical coordinates			
1	<i>Platyclusus orientalis</i>	Rušanj, Belgrade area 44°40'57.1"N, 20°26'21.5"E		22/08/2024	school yard
2		Dobanovci, Belgrade area 44°49'52.8"N, 20°13'58.5"E		22/08/2024	city street, ornamental row
3		Ušće, Belgrade area 44°48'54.3"N, 20°26'44.7"E		22/08/2024	city park
4		Niš area 43°19'45.6"N, 21°53'20.5"E		13/09/2024	hotel park
5	<i>Thuja occidentalis</i>	Dobanovci, Belgrade area		22/08/2024	city street, ornamental row

		44°49'52.8"N, 20°13'58.5"E Badanj, Raška Mt. Kopaonik		
6		43°16'26.7"N, 20°47'02.3"E Bubanj, Niš area	05/09/2024	home yard
7	<i>Cupressus × leylandii</i>	43°23'00.7"N, 21°48'32.7"E	13/09/2024	school yard

Plant samples were collected by cutting off branches, which were subsequently packed in 5-litre nylon bags, and transported to the laboratory. All samples were etherised with ethyl acetate for 30 minutes before examination to immobilise the mites. Quantitative assessment of scales was performed on each sample (25 twigs, each about 25–30 cm long) under a stereomicroscope, by counting active mite stages and eggs. During this examination, mites were photographed directly on the plant scales under the stereomicroscope. Qualitative examination was performed by beating the branches over sheets of white satin paper (60 × 40 cm), enabling the collection of additional individuals that may have remained undetected during the quantitative. During qualitative examination, isolated mites placed in Petri dishes were photographed under the stereomicroscope.

Preliminary examination of mites was conducted using a Leica Wild M3Z stereomicroscope, allowing the collection of isolated mites in Petri dishes. All collected specimens were subsequently placed in a clearing fixative, consisting of a 1:5 mixture of lactic acid (88%) and ethanol (70%). Permanent mounts were prepared using Heinze's medium [32].

Mounted mites were examined using a phase-contrast microscope Olympus BX53, equipped with cellSens Entry 2 (CS-EN-V2) imaging software for measurement and photography. Species identification was carried out using several literature sources for the family Tetranychidae [8,9,11,15,18,19,21,23,24,35–37]. The catalogue and worldwide database of spider mites were also consulted to verify the species distribution [1,38]. Permanent mounts are deposited in the collection of the Department of Entomology and Agricultural Zoology, Faculty of Agriculture, University of Belgrade.

### 3. Results

Two spider mites species, *Eotetranychus libocedri* (McGregor, 1936) and *Eotetranychus thujae* (McGregor, 1950) are reported for the first time in the fauna of Serbia on conifers. *E. libocedri* was recorded on three species, while *E. thujae* has been found on a single conifer species (Table 2).

**Table 2.** *Eotetranychus libocedri* and *Eotetranychus thujae* on plants and number of counted and examined individuals.

Sample code	Spider mite species	Plant species	Number of counted individuals		Number of examined individuals		
			active stages	eggs	female	male	nymph
1	<i>Eotetranychus libocedri</i>	<i>Platycladus orientalis</i>	156	188	11	6	2
2			2	10	5	2	1
3			3	16	1	2	4

4			23	193	8	2	/
5		<i>Thuja occidentalis</i>	1	7	5	1	2
7		<i>Cupressus × leylandii</i>	2	2	11	4	/
6	<i>Eotetranychus thujae</i>	<i>Thuja occidentalis</i>	18	13	10	2	1

Regarding taxonomic considerations, apart from their clear placement within the large genus *Eotetranychus* Oudemans, 1931 [39], both species share a number of distinctive morphological characters that have led to their previous assignment to smaller taxonomic groups. For instance, they have been included in the Libocedri Group within the genus *Eotetranychus* [9], the subgenus *Mononychus* Wainstein, 1960, and the genera *Platytetranychus* Oudemans, 1931, and *Mononychellus* Wainstein, 1971. According to Baker and Tuttle [21] their shared morphological characters are as follows: dorsal body setae are much shorter than the distance between their bases; prodorsal striae are longitudinal and opisthosomal striae are transverse; peritremes are distally bulbous; tibia II has five tactile setae; tarsus I with distal and adjacent duplex setae.

Family **Tetranychidae** Donnadieu, 1875

Subfamily **Tetranychinae** Berlese, 1913

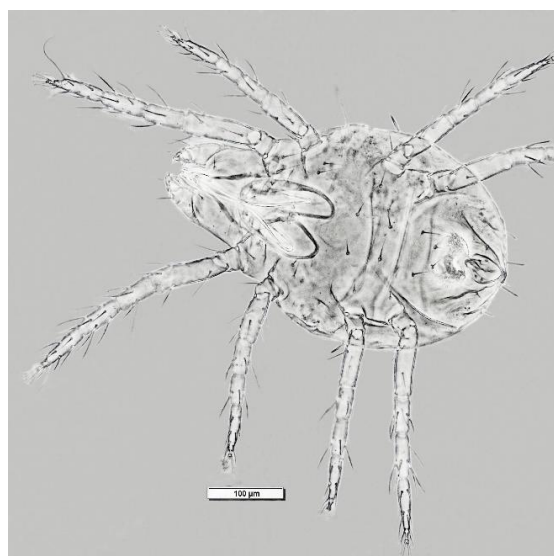
Tribe **Tetranychini** Reck, 1950

Genus ***Eotetranychus*** Oudemans, 1931

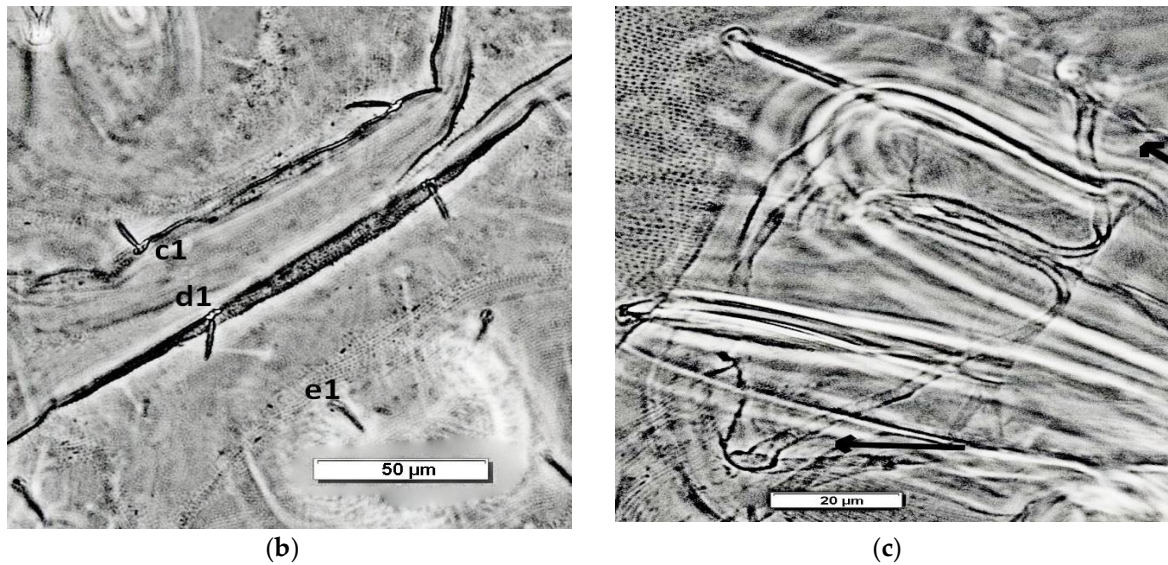
Species *Eotetranychus libocedri* (McGregor, 1936)

Material examined. Twenty-five females, twelve males and seven nymphs on *P. orientalis*. Five females, one male and two nymphs on *T. occidentalis*. Eleven females and four males on *Cupressus × leylandii* (Table 2)

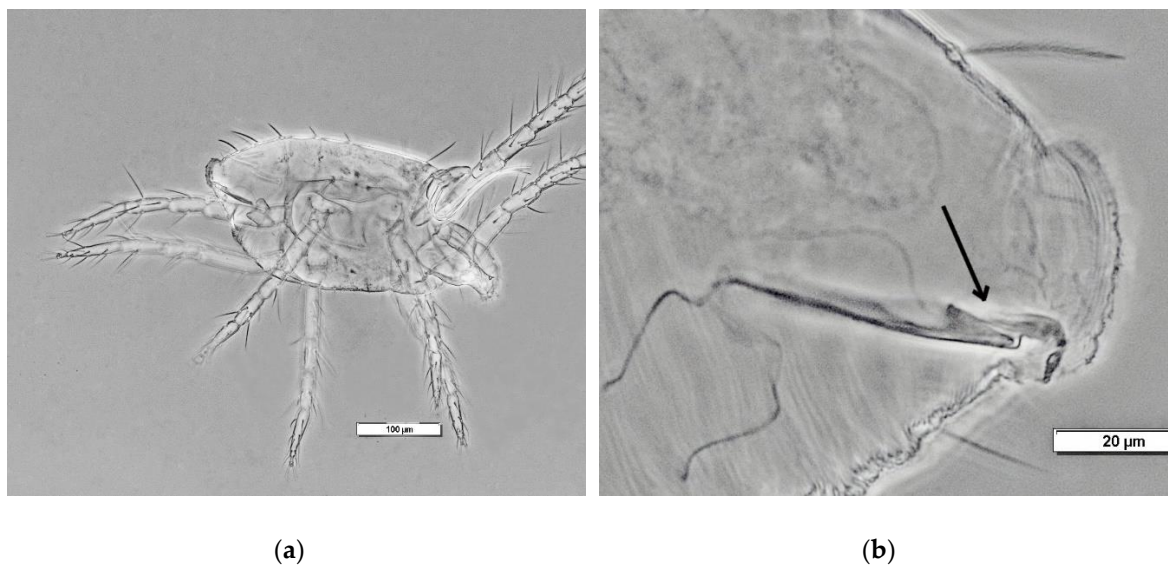
Remarks. The female *E. libocedri* (Figure 1a) can be recognized by the following characters: vertical setae (*v2*) are long, approximately equal to the distance between their bases; dorsocentral setae (*c1*, *d1*, *e1*, *f1*, *h1*) (Figure 1b) are smaller than other dorsal setae, very short, about half or less of the distance between their bases; dorsolateral setae (*c2*, *d2*, *e2*, *f2*) are slightly longer than dorsocentrals; both dorsocentral and dorsolateral setae are lanceolate and serrate; the spinneret of the palpal tarsus is very small, as long as it is wide; peritremes are slightly enlarged distally, not hooked (Figure 1c); pregenital striae are simple. The male (Figure 2a) can be recognised by the stout aedeagus, which narrows abruptly to the tip (Figure 2b).



(a)



**Figure 1.** *Eotetranychus libocedri* - Femal: (a) Dorsal view; (b) Dorsocentral setae: *c1*, *d1*, *e1*; (c) Peritremes.



**Figure 2.** *Eotetranychus libocedri* - Male: (a) Lateral view; (b) Aedeagus.

**Host plant.** This species was recorded on ornamental cypresses: *P. orientalis*, *T. occidentalis* and *Cupressus × leylandii*, (Table 2). As the research was conducted during a prolonged period of extremely high temperatures and drought, the population density was generally low (Table 2), and symptoms of scale damage were not clearly expressed. Only at one locality (Sample code 1), on *P. orientalis*, there were several isolated but well-developed colonies with visible webs (Figure 3a) and numerous eggs (Figure 3b), accompanied by localized discoloration of scales. During this research, *E. libocedri* was not found cohabiting with other spider mites species.

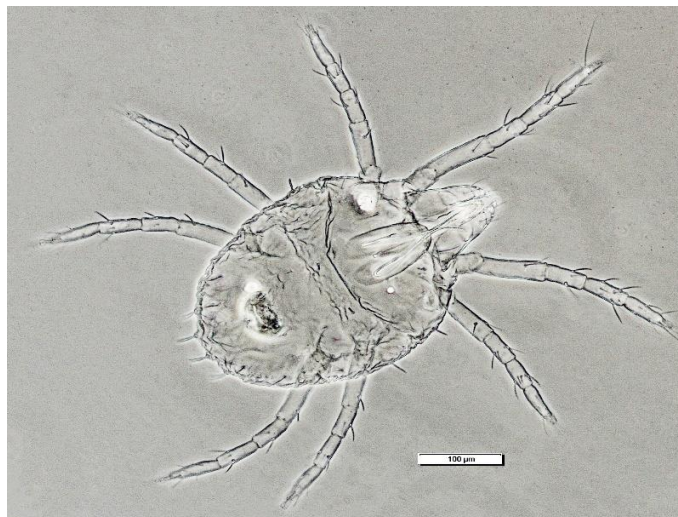


**Figure 3.** *Eotetranychus libocedri* on *Platicladus orientalis*: (a) Mite colony in web; (b) Mite eggs in scales.

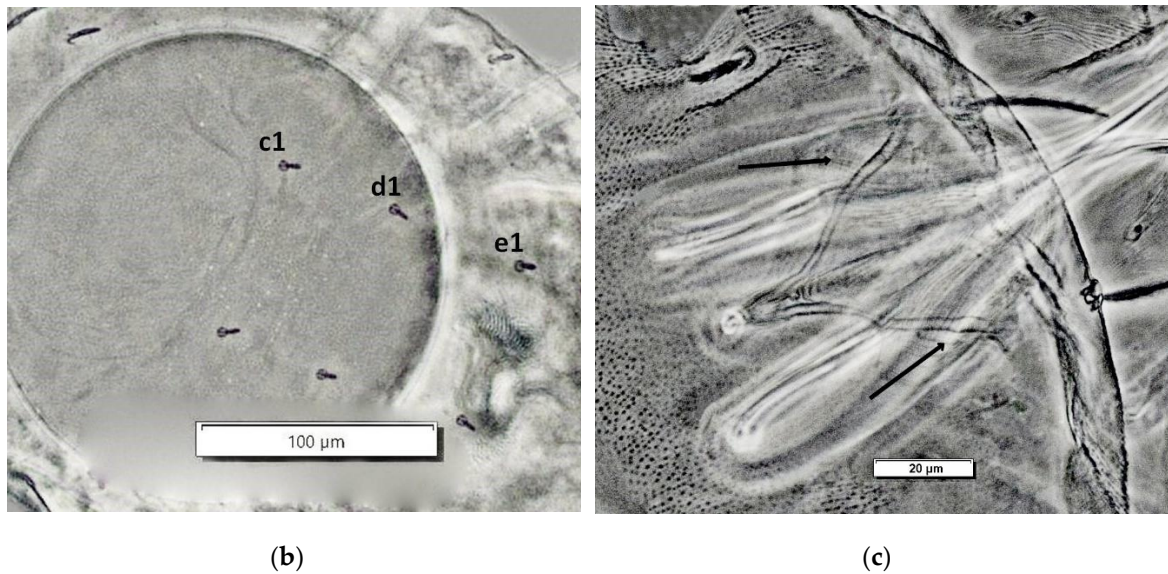
Species *Eotetranychus thujae* (McGregor, 1950)

Material examined. Ten females, two males, and one nymph on *Thuja occidentalis*.

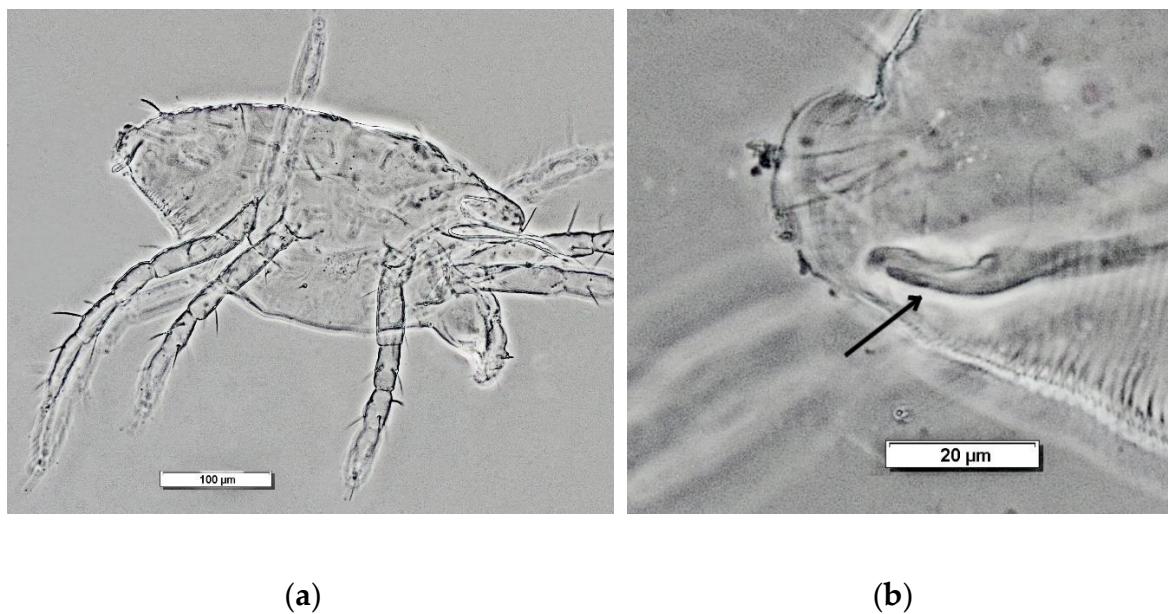
Remarks. The female *E. thujae* (Figure 4a) can be recognized by the following characters: vertical setae (*v2*), outer sacral (*f2*), and clunals (*h1*) are slightly longer than the other setae; other dorsal setae are small, lanceolate, and serrate (Figure 4b); the spinneret of the palpal tarsus is three times as long as it is wide; peritremes are slightly enlarged and distally bulbous (Figure 4c); pregenital striae are lobed. The male (Figure 5a) can be recognized by the elongate aedeagus, which gradually tapers to a rounded tip (Figure 5b).



(a)

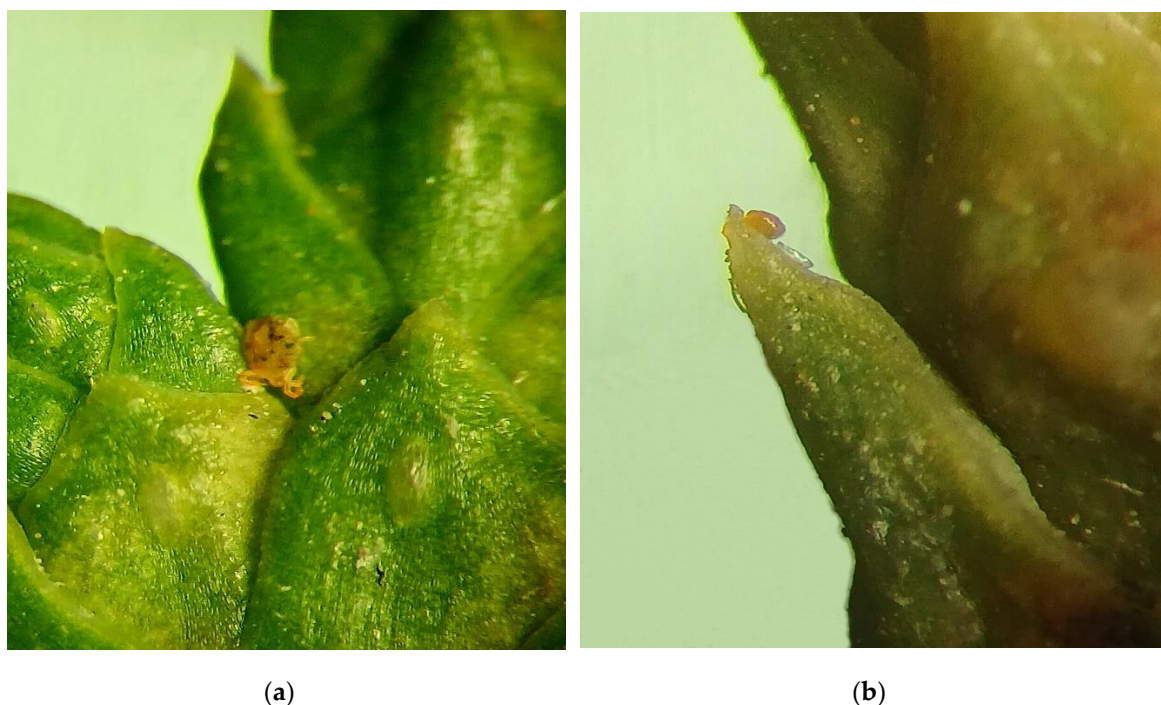


**Figure 4.** *Eotetranychus thujae* – Female: (a) Dorsal view; (b) Dorsocentral setae: *c1*, *d1*, *e1*; (c) Peritremes.



**Figure 5.** *Eotetranychus thujae* – Male: (a) Lateral view; (b) Aedeagus.

Host plant. This species was recorded only on *T. occidentalis* (Table 2). The population density was extremely low with only rare active stages (Figure 6a) and eggs (Figure 6b) recorded. No visible symptoms of scale damage were detected. During this research, *E. thujae* was not found cohabiting with other species of spider mites.



**Figure 6.** *Eotetranychus thujae* on *Thuja occidentalis*: (a) Female on scale; (b) Egg on scale.

#### 4. Discussion

Together with previously published data [31,40,41], this record of *E. libocedri* and *E. thujae* increased the number of known spider mite species in Serbia to 47. *E. libocedri* and *E. thujae* have so far been reported in Europe from only a few countries. Both species are recorded in Hungary and the European parts of Russia; in the European part of Turkey only *E. libocedri* has been reported, while in Georgia only *E. thujae*, has been documented. Thus, their discovery in Serbia expands the known distribution of both species in Europe.

According to available literature, most records of *E. libocedri* in Eurasia are associated with ornamental plants in urban environments, suggesting that the actual distribution may be considerably larger than currently known [15]. Therefore, further research is expected to expand the range of known host plants, including several relevant conifer families in Serbia.

During our research, *E. libocedri* and *E. thujae* were found on infested plants only in pure populations, i.e., without the presence of the commonly associated species *O. ununguis* [3–5], although they can cohabit with it on conifers [6]. In mixed populations with other spider mites, *O. ununguis* usually reaches higher population density, spins a stronger web, and causes more severe damage to host plants, making it difficult to objectively assess the harmfulness of other species on the same host plant [6]. In the present study, this issue did not arise, as *E. libocedri* and *E. thujae* were observed in isolation.

The relatively low population densities recorded through quantitative assessment of the samples were consistent with the weak manifestation of damage symptoms on the host plants. However, such results do not necessarily indicate that the two species are harmless, given the prevailing environmental conditions during the study period. Dry summer months are generally not favourable for spider mite populations on conifers to reach their maximum densities [6]. Research conducted during ecologically more favourable periods for population growth may yield different results.

Both species are of Nearctic origin, and are relatively rarely recorded within their native range, which therefore remains insufficiently studied. Although their presence in the Palearctic and Europe has been confirmed, the study of their distribution in these regions is even more limited, stressing the need to collect further data on these allochthonous species. Future studies in Serbia should focus on monitoring *E. libocedri* and *E. thujae* expand their host range or cause damage to ornamental conifers, and studying their biology, distribution, symptomatology, and control measures.

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**Conflicts of Interest:** The authors declare no conflicts of interest.

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