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A new distribution area of the *Ophrys isaura* Renz & Taubenheim in Türkiye and some ecologic characteristics

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Abstract: *Ophrys isaura* Renz & Taubenheim., is one of the rarest endemic species in Türkiye and belongs to the Orchidaceae family. According to a literature survey, the species at risk of extinction in nature is distributed in the C3 and C4 squares of the Turkish flora. In this study, it was observed that the plant has also distribution within a narrow range of between 1100 and 1200 meters in the *Pinus nigra* subsp. *pallasiana* forest clearing near Akdoğan village in the Eğirdir district in Isparta from southern Türkiye. *O. isaura* was identified for the first time at this locality and represents the southwestern most record of this species in Türkiye. Some morphological, phenological and ecological characteristics of the species were revealed by examinations of the newly detected population. The morphological characteristics of the new population were compared with the descriptions of *O. isaura* in Flora of Türkiye and Illustrated Flora of Türkiye. Some morphological characteristics exhibit minor differences compared to the information available in the early studies. In the study, some ecological characteristics of the new distribution area of the species were identified. The dominant rock formation in the area is fractured and cracked limestone. The radiation index was calculated to be 0.46, while the temperature index was determined to be 0.89. These measurements indicate that the area generally receives sunlight in the early morning and has moderate illumination throughout the day. Soils up to 10 cm depth layer within areas characterized by humid and wet soil conditions are of clay type and exhibit very low skeletal content. The high levels of lime (30.65%), organic matter (6.12%), and weakly alkaline conditions (pH: 7.11) in the habitat of the species provide a favorable environment for water, air, and nutrient economy within this soil depth layer. Overall, the plant species diversity in the open forest area where *O. isaura* was sampled was found to be higher compared to that in the understorey of the surrounding black pine forest areas.
Keywords: *Ophrys isaura*, Endemic, IUCN, New distribution area, Morphological and ecological characteristics, Türkiye

Ophrys isaura Renz & Taubenheim'in Türkiye'de yeni bir yayılış alanı ve bazı ekolojik özellikleri

Öz: *Ophrys isaura* Renz & Taubenheim, Orchidaceae familyasına ait Türkiye'nin en nadir endemik türlerinden biridir. Literatür araştırmasına göre doğada nesli tükenme riski altında olan tür, Türkiye florasının C3 ve C4 karelerinde yayılış göstermektedir. Bu çalışmada, *O. isaura*'nın Türkiye'nin güneyinde yer alan Isparta'nın Eğirdir ilçesine bağlı Akdoğan köyü yakınlarındaki *Pinus nigra* subsp. *pallasiana* orman açıklığında 1100-1200 metreler arasında dar bir alanda yayılış gösterdiği gözlenmiştir. *O. isaura* bu lokalitede ilk kez tanımlanmıştır ve bu türün Türkiye'deki en güney batı kaydını temsil etmektedir. Yeni tespit edilen popülasyon üzerinde yapılan incelemeler ile türün bazı morfolojik, fenolojik ve ekolojik özellikleri ortaya konmuştur. Yeni popülasyonun morfolojik özellikleri, *O. isaura*'nın Flora of Turkey'deki tanımları ve ayrıca Resimli Flora of Turkey ile karşılaştırılmıştır. Bazı morfolojik özellikler, ilk çalışmalarda mevcut olan bilgilere kıyasla küçük farklılıklar göstermektedir. Çalışmada, türün yeni yayılış alanının bazı ekolojik özellikleri belirlenmiştir. Alandaki baskın kaya formasyonu kırıklı ve çatlaklı kireçtaşıdır. Radyasyon indeksi 0,46 olarak hesaplanırken, sıcaklık indeksi 0,89 olarak belirlenmiştir. Bu ölçümler, alanın genellikle sabahın erken saatlerinde güneş ışığı aldığını ve gün boyunca orta derecede aydınlığa sahip olduğunu göstermektedir. Nemli ve ıslak toprak koşullarıyla karakterize edilen alanlardaki 10 cm derinlik katmanına kadar olan topraklar kil türündendir ve iskelet içeriği çok düşüktür. Türün yaşam alanındaki yüksek kireç (%30,65), organik madde (%6,12) ve zayıf alkali koşullar (pH: 7.11) bu toprak derinliği katmanında su, hava ve besin ekonomisi için elverişli bir ortam sağlamaktadır. Genel olarak, *O. isaura'nın* örneklendiği açık orman alanındaki bitki türü çeşitliliği, çevredeki karaçam orman alanlarının alt tabakasındakine kıyasla daha yüksek bulunmuştur.
Anahtar kelimeler: *Ophrys isaura*, Endemik, IUCN, Yeni yayılış alanı, Morfolojik ve ekolojik özellikler, Türkiye

1. Introductions

There are 11466 plant taxa of 3649 naturally growing in Türkiye and are endemic (Güner et al., 2012). Türkiye is a rich country for endemic species, and the Orchidaceae (Salepgiller) family has an important place in this richness. Endemic orchid species risk extinction due to the rapidly increasing population, the uprooting of their tubers for salep production, intensive agricultural practices and overgrazing (Kreutz, 2000; Kreutz and Krüger 2014; Sezik, 1984). The *Ophyrs* genus, generally known as Salep in Türkiye, is one of the essential genera containing the most common species of this family. The *Ophrys* genus is represented by approximately 200 species distributed in the European

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continent, North Africa, Anatolia, the Middle East and the Caucasus (Deniz, 2012; Sezik, 2002). In Türkiye, the *Ophyrs* genus is characterized by 65 taxa 14, which are endemic (Güner et al., 2022). *Ophyrs* sp. tubers, as are other orchid species, are used in medicine, beverages, and ice cream production. Therefore, they are threatened by collection from nature (Sezik, 2002).

Ophrys isaura Renz & Taubenheim species belonging to the *Ophyrs* genus is an endemic plant for Türkiye. The species was described by Renz and Taubenheim (1980) from the region between Gülnar and Bozağaç (Mersin). The species' distribution areas are C3 Antalya: Akseki, C4 Mersin: Gülnar in Türkiye. The species is found in pine forests, maquis areas, seasonal waterfronts, oak groves and clearings, and calcareous places between 495 and 1200 m. (Güner et al., 2022). *O. isaura* is one of the rarest orchid species in Europe and neighboring regions due to its low number of individuals and small distribution area. It is, therefore, classified as highly endangered (Kreutz and Krüger 2014).

A new distribution area of *O. isaura* was determined during the field studies. In our research, it was determined that the plant is distributed within a narrow range of between 1100 and 1200 meters in the C3 square, specifically in a forest clearing of *Pinus nigra* subsp. *pallasiana* (Lamb.) Holmboe near Akdoğan village in the Eğirdir (Isparta) district of Türkiye. Compared with the literature, some similarities and differences were found in the morphological characteristics of *O. isaura*. In addition, some ecological characteristics (climate, soil, biodiversity, etc.) of the area where *O. isaura* species is found were determined. This study aimed to add a new distribution area of the species and examine some ecological characteristics of the species.

2. Material and Methods

The study's material was samples of the species *O. isaura*. The samples were collected in May 2024 from the *Pinus nigra* subsp. *pallasiana* forest clearing (1100-1200 m) near C3 Isparta: Eğirdir, Akdoğan Village, the natural distribution area of *O. isaura.* Flora of Turkey (Davis, 1984) and Illustrated Flora of Turkey (Güner et al., 2022) have been used to identify plant samples. Plant samples of *O. isaura* were examined, and various morphological features (such as stem, tuber, leaf, flower, fruit, and seed) were identified. A stereo microscope, digital caliper and digital camera were used to determine the features. The morphological and phenological characteristics obtained in the study were compared with the literature.

In May of 2024, sampling was conducted to determine the ecological characteristics of the area where the *O. isaura* species was found. The geographical coordinates of the area where the plant is distributed were recorded according to the WGS 84 / UTM zone 36N system. The area is within the C3 square according to Davis's grid system (1984) (Figure 1).

In the sampling area, altitude (m) with GPS, slope (°) with a clinometer, and aspect (°) with a compass were measured. Based on the slope (°) and aspect (°) values, radiation index and temperature index values were calculated in the sampling area (Moisen and Frescino, 2002). The area's slope position and landform characteristics were determined through field observations. In the sampling area, stoniness (%) and soil depth (m) were determined at 10 random points using the iron rod penetration method (Eriksson and Holmgren, 1996).

Bedrock samples from the field were identified at the Rock and Mineral Analysis Laboratory of the Department of Geological Engineering, Faculty of Engineering and Natural Sciences, Süleyman Demirel University. Thus, local-scale determinations were made regarding the rock formation of the sampling area and the general geology of the region. The skeleton content (%) and moisture status in the soils around the plant's roots were determined through field observations. Additionally, soil samples were taken from the 0-10 cm depth layer. Soil samples from three different plant root environments were transported to the Isparta Soil and Plant Analysis Laboratory under appropriate conditions. The samples, free from inorganic materials (e.g., stones, gravel larger than 2 mm) and various organic materials (e.g., leaves, branches, roots), were left to air dry in the laboratory. Once air-dried, the samples were sieved (2 mm or 0.5 mm), mixed, and prepared for various soil analyses. In the laboratory, texture analysis was performed using the Bouyoucos hydrometer method (Bouyoucos, 1962), total lime content (%) was determined using the Shiebler calcimeter method (Allison and Moodie, 1965), and total organic matter content (%) was determined using the Walkley-Black wet oxidation method (Allison, 1965). Finally, soil acidity (pH) and Electrical Conductivity (dS/m) measurements were carried out in the filtrate of a soil-water mixture prepared at a 1:2.5 ratio (Soil Survey Staff, 1992).

Figure 1. The location of the sampling area of the *O. isaura* species in the grid system of Davis (1984)

To determine the climatic characteristics of the sampling area, data from the CHELSA version 2.1 database were used. According to the coordinates of the sampling area, 19 bioclimatic variables were obtained from the CHELSA database (Karger et al., 2017).

Finally, a plant inventory was conducted in two separate 400 m² sample areas, one in a forest opening and the other under a black pine forest, where the species was sampled. Species richness (S) (Formula 1) calculations were initially performed in the sample areas (Peet, 1974).

$$S=\sum_i^S S_i \quad 1$$

In the formula, S_i represents the number of different species in the sampling area.

Additionally, the plant species identified in the sample areas were recorded according to the Braun-Blanquet (1932) abundance scale and then converted into numerical values suitable for biological diversity calculations, as suggested by Fontaine et al. (2007). Using the numerical values of each plant species, within-area (alpha) diversity values were calculated according to the Shannon-Wiener (H') index (Formula 2) (Shannon, 1948).

$$H' = -\sum_{i=1}^{S} p_i \ln p_i \quad 2$$

In the formula, S represents species richness, and p_i represents the proportional values of the species.

For determining the dissimilarity (beta) values between the forest opening and the black pine forest areas, the Bray-Curtis distance (D) formula (Formula 3) was used (Bray and Curtis, 1957).

$$D=1-2\frac{\sum_{i=1}^{S}\min(a_i,b_i)}{\sum_{i=1}^{S}(a_i+b_i)} \quad 3$$

In the formula, S represents species richness in the sampling areas, and a_i and b_i represent the abundance values of the i^{th} species in the sampling areas.

The Biological Diversity Components (BİÇEB) Calculation Software was used for diversity calculations (Özkan et al., 2020).

3. Results and discussion

The field studies determined a new distribution area of *O. isaura*. In this study, it was revealed that the species also spread in a narrow area in Isparta. It was determined that there were approximately 77 natural individuals in this newly identified locality (Figure 2). New field studies have been ongoing to determine whether this area's population distribution is limited. When the samples of the newly identified *O. isaura* population were compared with the literature regarding morphological characteristics, some similarities and differences were determined. The results of the examination of the morphological characteristics of the species and comparison with the literature data (Davis, 1984; Güner et al., 2022) are given in Table 1.

According to the literature survey, *O. isaura* is reportedly distributed in the Mediterranean phytogeographic region and is an Eastern Mediterranean element (Davis, 1984; Güner et al., 2022). The presence of *O. isaura* has been determined to be between Gülnar and Aydıncık (Mersin) in Southern Anatolia and also in a wider area in Cevizli (Akseki) and Alanya (Antalya) (Kreutz 2000, Kreutz and Krüger 2014; Güner et al. 2022). The locality of the newly identified population is quite far from the localities given in the literature, and the bird flight distances between the localities given in the literature are 108-271 km. Interestingly, the literature has long distances between the new locality and the localities.

In this study, plant individuals of the *O. isaura* population in the newly identified distribution area were compared with the literature regarding morphological characteristics. Some morphological characteristics show minor differences from the information in the literature. These minor morphological differences can be thought to be due to ecological conditions.

Although some morphological features of the new population may differ slightly from the literature, the taxonomic status of the population can be re-evaluated based on the results obtained by conducting anatomical, palynological, molecular, etc. studies on the species.

Table 1. Comparison of the morphological properties

O. isaura			
	Flora of Turkey (Davis, 1984)	Illustrated Flora of Türkiye (Güner et al., 2022)	Newly detected population
Stem	To 40 cm	17-45 cm	15-36 cm
Leaves	Leaves ± basal, broadly lanceolate	4-7 leaves oblong to broadly lanceolate, pointed 5,5-11 x 0,9-3 cm	4-7 leaves broadly lanceolate 5,1-10,3 x 0,8-2,6 cm
Flowering Time	-	April-May	May-June
Inflorescence	To 12 flowered	4-12 flowered, sparse	4-10 flowered
Bracts	Bracts exceeding flowers	Lanceolate, 11-32 (50) x3-10 mm, equal to or longer than the ovary	10,6-28,7 mm Equal to or longer than the ovary
Sepals	strongly reflexed, green, sometimes suffused greyish-violet, 12 x 5 mm.	Ovate to oblong, 9-13 x 5-8 mm, yellowish green, sometimes tinged with violet, usually strongly recurved.	7-13 x 3-8 mm yellowish green, sometimes tinged with violet, sometimes recurved
Petals	ligulate, 4 x 2,5 mm, whitish-green, ± suffused with rose, minutely hairy.	triangular-ligulate, 3-5 x 2-2,7 mm, yellowish green, sometimes whitish to violet, backward, sometimes slightly auricated at the base, whitish gray short hairs.	Triangular-ligulate, 2,7-4 x 1,8-2,4 mm, yellowish green
Labellum	Labellum horizontally spreading, middle lobe narrow-based, margins recurved, maroon or dark brown, velvety, with yellowish subglabrous margins towards the apex, and with yellowish-green, glabrous appendix upturned; lateral lobes small, with short, outward-directed horns, densely villous outside, whitish and glabrous inside; speculum on the inner half of labellum, of white ribbons encircling brownish spots.	Labellum pattern basically H-shaped, sometimes with scattered lines and spots, usually round false eyes bright, greenish to black in the center. The labellum is fragmented, three-lobed, narrowly ovoid to rectangular, dark reddish brown to blackish purplish near the base, usually yellowish towards the tip, velvety hairy, curved back. The middle lobe is rectangular to ovoid, narrow at the base; the lateral lobes are small, straight to slightly triangular protrusions near the base of the lip, yellowish.	The labellum features detected in the Flora of Turkey and the Illustrated Flora of Turkey are similar. In the newly identified population, it was determined that the labellum pattern with the spaces inside the letter H was more similar to *O. phrygia* in some individuals.
Tuber	-	2 tubers 1-3 x 0,8-3 cm	2 tubers 1-2,5 x 0,7-2,6 cm
Fruit	-	Cylindrical, 18-30 x 4,5-6 mm.	Cylindrical,18-24 x 3,5-5 mm

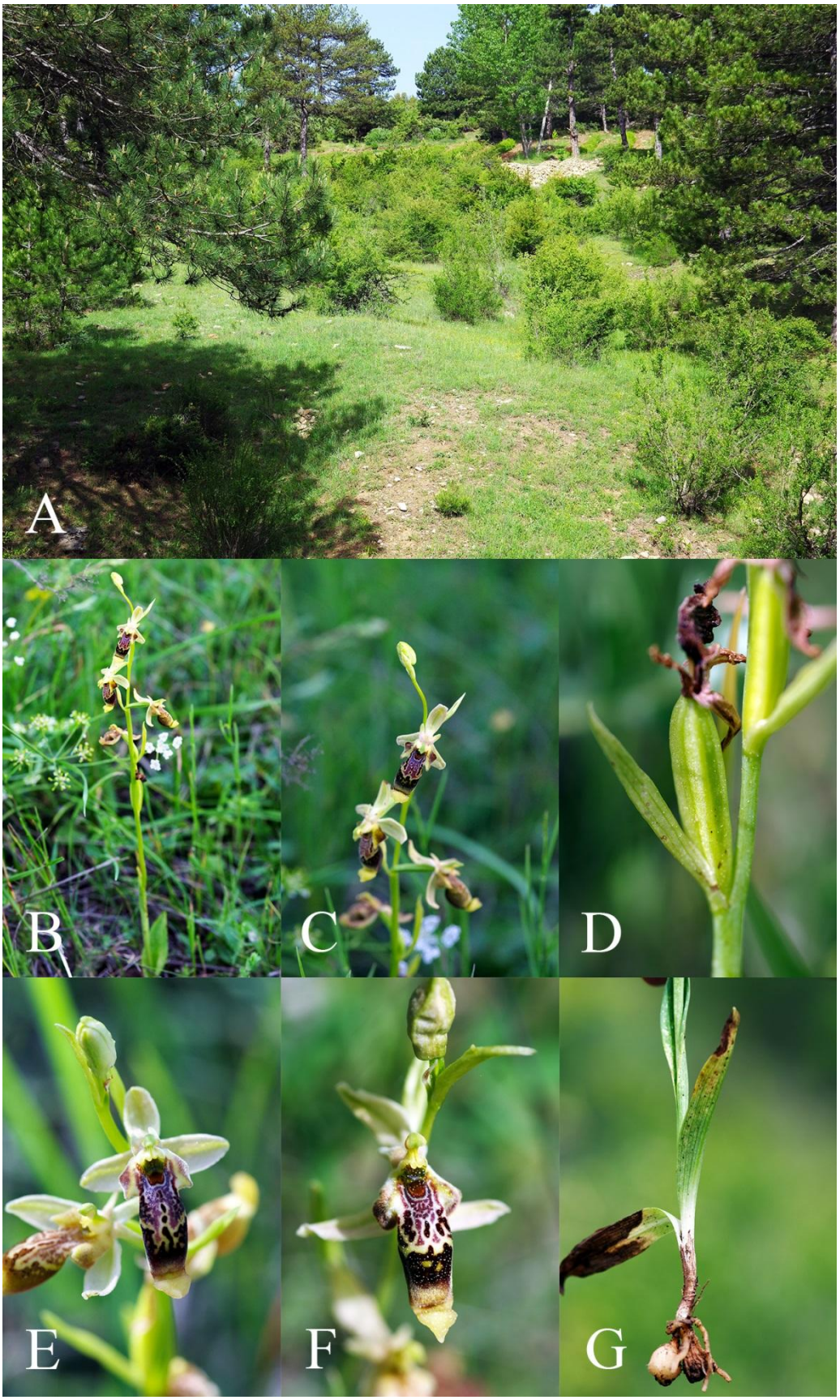

Figure 2. A) New distribution area, B and C) General appearance D) Fruit E) Front and side view of the flower G) Tuber

The elevation of the forest opening area where the species was sampled ranges approximately between 1100 and 1200 meters. The location is surrounded by black pine stands of varying ages and canopies. The average slope of the area is 17°, and the aspect is 115°, facing between east and southeast. According to the aspect degree, the radiation index value was calculated as 0.46, and the temperature index value was calculated as 0.89 based on the aspect and slope degree values. These measurements indicate that the sampling area receives early morning sunlight and has moderate illumination during the day. The high-temperature index is a vital forest site affecting the length of the vegetation period, especially in spring and summer.

The dominant rock formation in the area is fractured and cracked limestone. The fractures and cracks are generally filled with secondary calcite, with variable thicknesses (up to 1 cm). The rock surface appears smeared with silt and clay-sized detrital material, likely due to the area encountering a limestone band within the detrital rocks. Besides the locally described limestone rock formation, considering the general geology, it is possible to say that sandstone, siltstone, shale, and chert rock formations form the dominant lithology in the region.

The forest opening area, with a valley bottom and mid-slope position, contains brown forest soils of moderate or deep class. In the area with concave landform characteristics, the skeleton content of the soils was found to be very low (less than 1%). The area, with moist soil conditions, provides a rich water environment, with sand, silt, and clay proportions in the soils of the 0-10 cm depth layer being 23.9%, 34.0%, and 42.1%, respectively. According to these proportions, the forest site, dominated by "clay" texture soils, has an oxygen content just sufficient for the plant species distributed in the area. Additionally, in this depth layer, the lime content of the soil was determined to be 30.65%, and the organic matter content was 6.12%. According to these proportions, the soils in the 0-10 cm depth layer are classified as "very high lime" and "very rich" in organic matter content. The high lime and organic matter ratios in the upper clay soil layer help the soil reach a granular structure, providing a favorable water and air economy environment for the distribution of *O. isaura* and associated species in this soil depth layer. Indeed, in the samples taken from the specified depth layer, the density of fine roots is very high. This situation relates to the plant finding an optimal forest site in the 0-10 cm upper soil layer for utilizing nutrients and water. The Electrical Conductivity (EC) value in the specified soil depth layer was also 0.182 dS/m, indicating low salinity suitable for plant growth. The measured pH value (7.11) shows the soils are weakly alkaline. These values have provided optimal conditions in the soil for the presence of the plant. A blocky and platy structure prevails in soil conditions deeper than 10 cm due to the high clay content.

Analyzing the climatic data obtained from the 19 bioclimatic maps downloaded from the CHELSA database with a 30 arc-sec resolution in ESRI Grid format, the mean annual air temperature (Bio1) was determined as 11.4 °C, and the annual precipitation amount (Bio12) was 792.3 kg/m² (Figure 3).

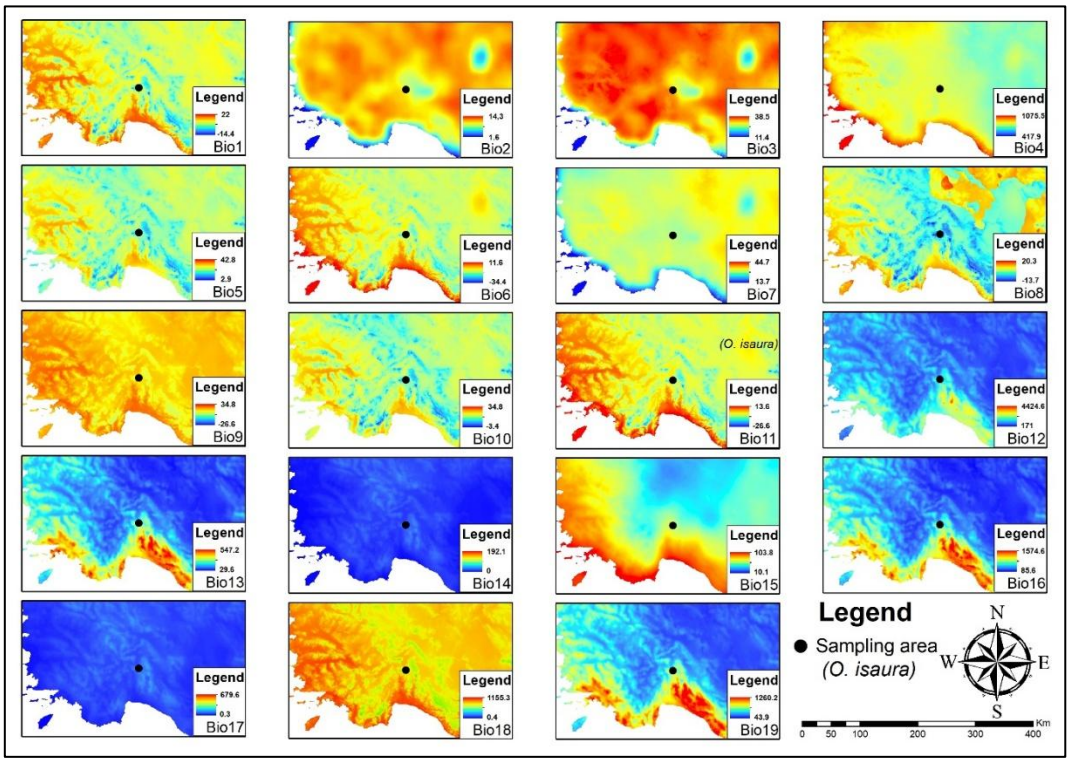

Figure 3. Bioclimatic maps

Bio1: mean annual air temperature (11.4 °C), Bio2: mean diurnal air temperature range (10.3 °C), Bio3: isothermality (32.6 °C), Bio4: temperature seasonality (770.0 °C/100), Bio5: mean daily maximum air temperature of the warmest month (27.7 °C), Bio6: mean daily minimum air temperature of the coldest month (-3.8 °C), Bio7: annual range of air temperature (31.5 °C), Bio8: mean daily mean air temperatures of the wettest quarter (1.7 °C), Bio9: mean daily mean air temperatures of the driest quarter (21.0 °C), Bio10: mean daily mean air temperatures of the warmest quarter (21.3 °C), Bio11: mean daily mean air temperatures of the coldest quarter (1.7 °C), Bio12: annual precipitation amount (792.3 kg m^2), Bio13: precipitation amount of the wettest month (136.5 kg m^2), Bio14: precipitation amount of the driest month (12.4 kg m^2), Bio15: precipitation seasonality (58.3 kg m^2), Bio16: mean monthly precipitation amount of the wettest quarter (342.2 kg m^2), Bio17: mean monthly precipitation amount of the driest quarter (52.1 kg m^2), Bio18: mean monthly precipitation amount of the warmest quarter (59.4 kg m^2), Bio19: mean monthly precipitation amount of the coldest quarter (342.2 kg m^2) (Karger et.al., 2017).

In the inventory conducted in the forest opening area where the species was sampled, species richness (*S*) was recorded as 48 taxa, while under the black pine forest, it was recorded as 33 taxa (Table 2). The Shannon-Wiener (*H'*) diversity value was determined as 3.575 in the forest opening area and 2.667 under the black pine forest. According to these values, plant species diversity in the forest opening area where the plant was sampled is higher than under the black pine forest. The beta dissimilarity value calculated using the Bray-Curtis distance (*D*) formula for these two habitats was 0.495. Species like *O. isaura*, which do not enter the forest and only distribute in the forest opening environment, increase the community dissimilarity value (beta diversity) due to their preference for similar local forest site conditions.

Table 2. Plant species found in the sample area taken from the new distribution area of *O. isaura*

No	Family	Taxa	Endemic status	IUCN category
1	Acanthaceae	*Acanthus hirsutus* Boiss.		
2	Asteraceae	*Anthemis cretica* subsp. *absinthifolia* (Boiss.) Grierson		
3		*Pilosella piloselloides* subsp. *piloselloides* (Vill.) Soják		
4	Berberidaceae	*Berberis crataegina* DC.		
5	Boraginaceae	*Myosotis lithospermifolia* Hornem.		
6		*Onosma aucheriana* DC.		
7	Brassicaceae	*Erysimum smyrnaeum* Boiss. & Balansa		
8	Campanulaceae	*Asyneuma virgatum* subsp. *cichoriiforme* (Boiss.) Damboldt		
9	Caryophyllaceae	*Silene dichotoma* subsp. *dichotom*a Ehrh.		
10	Cistaceae	*Helianthemum nummularium* (L.) Mill.		
11		*Cistus laurifolius* L.		
12	Cupressaceae	*Juniperus oxycedrus* subsp. *oxycedrus*		
13	Cyperaceae	*Carex flac*ca subsp. *erythrostachys* (Hoppe) Holub		
14	Fabaceae	*Lathyrus laxiflorus* subsp. *laxiflorus* (Desf.) O.Kuntze		
15		*Medicago sativa* subsp. *sativa* L.		
16		*Onobrychis oxyodonta* var. *oxyodonta* Boiss.		
17		*Trifolium hirtum* All.		
18		*Vicia lutea* var. *hirta* (Balbis) Loisel.		
19	Fagaceae	*Quercus cerris* L.		
20		*Quercus coccifera* L.		
21	Iridaceae	*Gladiolus italicus* Mill.		
22	Junaceae	*Juncus articulatus* L.		
23	Lamiaceae	*Ajuga chamaepitys* subsp. *chia* (Schreb.) Arcang.		
24		*Salvia tomentosa* Mill.		
25		*Stachys cretica* subsp. *anatolica* Rech.f.	Endemic	LR (nt)
26		*Scutellaria salviifolia* Benth.	Endemic	LR (lc)
27	Liliaceae	*Fritillaria acmopetala* Boiss.		
28		*Ornithogalum sphaerocarpum* A.Kern.		
29	Oleaceae	*Fontanesia phillyreoides* Labill.		
30	Onagraeacae	*Epilobium lanceolatum* Sebast. & Mauri		
31	Orchidaceae	*Anacamptis pyramidalis* (L.) Rich.		
32		*Ophrys phrygia* H.Fleischm. & Bornm.	Endemic	LR (lc)
33		*Ophrys reinholdii* Spruner ex Fleischm.subsp. *straussi* var. *leucotaenia* (Renz & Taubenheim) Deniz		
34	Pinaceae	*Pinus nigra* subsp. *pallasiana* (Lamb.) Holmboe		
35	Poaceae	*Avena barbata* subsp. *barbata* Pott ex Link		
36		*Bromus tectorum* L.		
37		*Dactylis glomerat*a subsp. *hispanica* (Roth) Nyman		
38	Polygalaceae	*Polygala anatolica* Boiss. & Heldr.		
39	Primulaceae	*Lysimachia atropurpurea* L.		
40	Ranunculaceae	*Ranunculus repe*ns L.		
41	Rosaceae	*Fragaria vesca* L.		
42		*Crataegus monogyna* var. *monogyna*		
43		*Crataegus orientalis* Pall. ex M.Bieb.		
44		*Pyrus elaeagnifolia* Pall.		
45		*Rosa canina* L.		
46	Salicaceae	*Salix alba* subsp. *alba* L.		
47	Styracaceae	*Styrax officinalis* L.		
48	Thymelaeaceae	*Daphne sericea* subsp. *sericea* Vahl.		

LR (nt): Lower risk (near threatened); LR (lc): Lower risk (Least concern)

O. isaura is an orchid species endemic to Turkey and has two main distribution areas: Cevizli (Antalya-Alanya) and Gülnar (Mersin). The habitat of the Cevizli location is found in damp areas, along streamsides and meadows, and rarely in pine forests. The habitat of the Gülnar location, on the other hand, is located on a slope with a water source, on limestone cliffs, under oak, and rarely pine forests. The habitat of the Eğirdir (Isparta) location has been found on a slope with a water source, in meadows and rarely under pine forests, in an area with limestone. Despite the similarity of their habitats in these 3 locations, a hot and arid climate has been observed in Gülnar, a cold and humid climate in Cevizli, and a temperate and rainy climate in Eğirdir. Although their ecological environments are similar, their climate types are different. Furthermore, discovering a new distribution area for this species, previously known to occur in only two regions, is highly significant. While the westernmost distribution area was previously known as Cevizli, this study has identified Eğirdir as the westernmost distribution area and has contributed this finding to the literature.

In the Red Data Book of Turkish Plants, *O. isaura* is listed in the EN category, which means that the existence of this species in nature is endangered (Ekim et al., 2000). This threat level is even higher for species with narrow distribution areas and small populations. *O. isaura* is in danger of extinction due to grazing pressure and the uprooting of tubers for salep production. In particular, the presence of an endemic species in more natural regions is essential for the continuation of that species' generation and for biodiversity. In this context, finding a new distribution area for *O. isaura*, an endemic species, is significant.

4. Conclusions

The danger category of *O. isaura* was re-evaluated by considering the population size, distribution areas and anthropogenic threat situations. Accordingly, *O. isaura*, which is in the 4th EN category, was collected by us from Akdoğan village, Eğirdir (Isparta) region, and we concluded that it is under high threat in the future. The EN category of this taxon should continue. It should be protected, especially against animal grazing and other biotic pressures. In this context, the new distribution area of *O. isaura* is located within the borders of the Isparta Eğirdir Forest Management Directorate. It has been determined that the people in the region do not have sufficient knowledge and awareness about the importance of *O. isaura*. They overgraze it and collect it to be used in salep production. Eğirdir Forest Management Directorate officials have been informed about this issue to protect the species.

Considering the location of the habitat where the species is located, its accessibility and the usage status of its immediate surroundings, the species should be protected in situ. Protecting biodiversity, especially endemic and rare taxa, is a national and international obligation. Türkiye is making the necessary legal arrangements and studies on this issue. However, it is seen that some problems have emerged in practice. These problems arise from implementing the laws and regulations rather than their existence. In conservation efforts, convincing the local people to internalize this issue and turn it into behavior is very important. In addition, conservation efforts should be carried out using a habitat protection approach rather than species protection.

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