

MORPHO-ECOLOGICAL STUDY OF WILD *Onobrychis altissima* GROSSH. AND *Onobrychis cyri* GROSSH. (*Onobrychis* MILL.) IN AZERBAIJAN

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Abstract

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Knowing the distribution and eco-geography of native *Onobrychis altissima* and *O. cyri* in Azerbaijan is critical for the implementation of strategies for their conservation and sustainable use. This study includes 27 specimens from nine wild populations of *O. altissima* and *O. cyri* collected from their natural habitats in Azerbaijan. The aims of this study are to provide a detailed morphological and eco-geographic characterization of *O. altissima* and *O. cyri*, to determine the distribution and environmental variables that define the eco-geographic zones native to Azerbaijan, and also to determine the priority areas for *in situ* conservation of native *Onobrychis* germplasm in Azerbaijan. Besides, research work was carried out for the purpose of clarifying the distribution areas, discovering more resistant species for selection, as well as enriching the genetic resources and Herbarium Fund. The specimens were biometrically assessed using 34 quantitative and 10 qualitative morphological characters. At each site, we recorded eco-geographic data regarding longitude, latitude, altitude, slope inclination, slope orientation, maximum and minimum temperatures, annual precipitation, relief, as well as soil traits including texture, humus, organic carbon, total nitrogen, pH, phosphorus pentoxide, potassium oxide, and magnesium. Cluster analysis of morphological characters showed that the nine populations could be divided into three main groups. Furthermore, principal component analysis (PCA) of eco-geographic data is discussed. The article presents tables and phenograms reflecting some eco-geographic information and a distribution map of the species.

Key words: *Onobrychis*, PCA, ecology, morphology, UPGMA.

Introduction

The genus *Onobrychis* Mill. (Miller, 1754) belongs to the legume family (*Fabaceae*) tribe *Hedysareae* and about 170 species of *Onobrychis* are distributed around the world. They occupy a wide area ranging from the territories near the Baltic Sea to the Mediterranean region to the south, as well as far western Asia and Siberia to the east. The diversity and concentration of these species are high in the Anatolia–Iran–Caucasia region (Shomurodov et al., 2021). Azerbaijan is one of the main centers of diversity for the genus *Onobrychis* Mill. In these regions, *Onobrychis* species are cultivated to produce protein-rich animal food. Also, these species have a crucial influence on the environment of soil by developing its nutritive value (Talebi et al., 2020; Hadadi et al., 2022; Yucel et al., 2022). Sainfoin has the potential to enhance the diversity and sustainability of agroecosystems by providing multiple revenue streams, numerous ecosystem services, and the flexibility to adapt management strategies to climate change (Craine et al., 2023).

In Azerbaijan, 24 wild species of *Onobrychis* are noted, representing five sections (*Dendobrychis* DC., *Laphobrychis* Hand.-Mazz., *Onobrychis* Mill., *Heliobrychis* Bunge, and *Hymenobrychis* DC.) (Asgarov, 2016). Taxonomically, *Onobrychis* is a critical section. This section seems to be the most problematic, and the boundary of species is not completely clear due to polymorphism in the morphological characters (Avcı et al., 2013). There is a whole range of morphological characters relative to constants that may be of taxonomic value in distinguishing species of the section *Onobrychis* (Khinthibidze, 1960). The species in this section are perennial herbs, rarely shrubs (unarmed) with many-flowered racemes, usually short wings, ovary rarely more than 1-ovulate, the fruit not stipitate and with a straight suture, and the crest and disc often with spines or teeth, rarely unarmed, the length of the standard varies relative to the keel (Toluei et al., 2013a). *Onobrychis altissima*, which belongs to this section, occurs in the northern part of the Lesser Caucasus, the southeast part of the Greater Caucasus, Nakhchivan and Talish regions in Azerbaijan (Azizkhanli, Asgarov, 2022). People also

use *O. altissima* in the treatment of several diseases (Mukemre et al., 2015). This species is of significant use in agriculture as a perennial forage and fodder legume, for increasing the nutritive value of drought-resistant pastures due to nitrogen fixation, and for soil conservation. At the same time, *O. altissima*, despite its great external resemblance to the cultivated sainfoin (*O. viciifolia*), is a completely wild, autochthonous plant that has no connection with the cultivated sainfoin races in some places in the same area. Ecologically, *O. altissima* is a more mesophilic grassland race of wild sainfoin, growing in wetter habitats with deeper soil. From a feeding point of view, the described sainfoin is of great importance, perhaps the greatest among all the wild sainfoins of the Caucasus (Grossheim, 1926).

Onobrychis cyri is one of the most common species in the central Transcaucasus and occurs in all regions in Azerbaijan, except Absheron. It grows mainly in the region of the middle reaches of the Kura River. Despite such a wide distribution, plants vary relatively little: the main features and sizes of parts of the flower and fruit always remain constant. The general habitat, as well as the width and consistency of the leaves vary somewhat depending on the habitat conditions (they are sometimes more and sometimes less xerophilous).

Many studies were done on the genus *Onobrychis* in neighboring countries by morphological, ecological, molecular and karyological traits (Navasardyan, 2013; Avcı et al., 2013; Ranjbar et al., 2018; Zarrabian et al., 2022). However, our literature is not rich in studies of the genus *Onobrychis*, including its Caucasian species.

This paper summarizes some of the results of the study on establishing the exact coordinates of habitats, characterizing the ecological conditions of growth, and determining the biometric indicators of two wild-growing species of sainfoin, common in regions of Azerbaijan.

Material and methods

Collection of materials

Twenty-seven specimens of *O. altissima* and *O. cyri* were collected from nine populations across Azerbaijan during 2013, 2018, 2021, and 2022. The specimens were prepared according to established herbarium techniques, and voucher specimens were deposited in the herbarium of the Institute of Genetic Resources of MSERA. Information on collection sites is listed in Table 1. Identification of the specimens was performed using the regional (Tamamshyan, 1954) and other related floras (Rechinger, 1984; Hedge, 1970; Grossheim, 1948; Takhtajan, 1962). Soil samples were collected from a depth of 0–20 cm and analyzed in the laboratory of the Institute of Soil Science and AgroChemistry of MSERA.

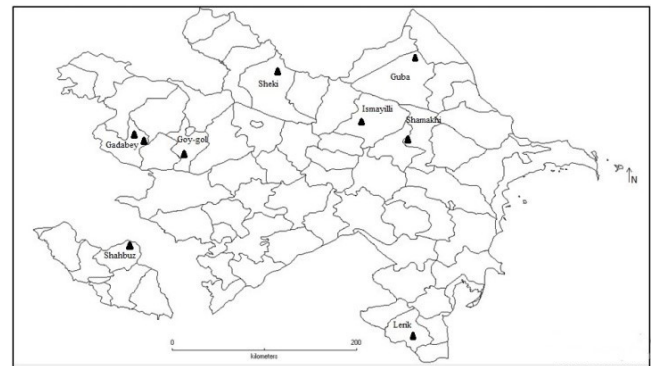


Fig. 1. Maps of the areas from where the samples had been collected (*Onobrychis altissima* and *Onobrychis cyri*).

Table 1. Population list of *Onobrychis altissima* and *Onobrychis cyri*.

Pop.№	Locality	Coordinates	Voucher number	Extension (m ²)
<i>Onobrychis altissima</i>				
1.	Goy-gol district, Togana village	N 40°46'373" E 46°33'348"	GH-3 2022	1
2.	Gadabey district, Slavyanka village	N 40°66'912" E 45°79'866"	GH-1 2022	10–100
3.	Nakhchivan AR, Shahbuz district, Bichenak village	N 39°53'524" E 45°79'866"	NVSh-4 2021	10–100
4.	Lerik district, Hiledara village	N 40°33'576" E 49°48'564"	Z ₁ 2018	>1000
<i>Onobrychis cyri</i>				
5.	Gadabey district, Soyudlu village	N 40°37'705" E 45°48'923"	G ₁₋₁ 2013	>1000
6.	Guba district, Vladimirovka village	N 41°38'545" E 48°53'717"	QV-1 2022	100–1000
7.	Ismayilli district, Ivanovka village	N 40°46'265" E 48°05'647"	A ₁₋₈ 2013	100–1000
8.	Sheki district, Kish village	N 41°15'428" E 47°12'923"	K ₁₋₃ 2013	>1000
9.	Shamakhi district, Agsu-Shamakhi road	N 40°38'04.0" E 48°28'28.6"	ShA-1 2022	10–100

Table 2. List of characters and related numerical codes used in morphological studies.

No.	Characters	Code	Unit of measurement
1.	Plant height	46-0/49-1/63-2/65-3/86-4/90-5/115-6	cm
2.	Stem length	33-0/36-1/38-2/48-3/50-4/71-5/74-6/90-7	cm
3.	Leaf length of upper leaf	4-0/4.5-1/5-2/7-3/7.5-4	cm
4.	Leaf length of lower leaf	5-0/6-1/7-2/5-3/9-4/10-5/11-6/14-7	cm
5.	Petiole length of upper leaf	1-0/2-1/2.5-2/5-3/7-4	mm
6.	Petiole length of lower leaf	1.5-0/3-1/3.5-2/4-3	cm
7.	Number of leaflets	11-0/13-1/15-2/17-3/21-4	Number
8.	Leaflet length of upper leaf	10-0/12-1/13-2/15-3	mm
9.	Leaflet length of lower leaf	10-0/15-1/20-2	mm
10.	Leaflet width of upper leaf	1.5-0/2-1/2.5-2/3-3/4-4	mm
11.	Leaflet width of lower leaf	2-0/2.5-1/3-2/4-3/5-4/6-5/7-6	mm
12.	Stipule length	4-0/5-1/6-2/7-3/10-4	mm
13.	Stipule width	2-0/2.5-1/5-2/6-3	mm
14.	Inflorescence length	4-0/5-1/7-2/9-3/10-4/12-5/17-6	cm
15.	Peduncle length	14-0/15-1/16-2/17-3/18-4/20-5/30-6	cm
16.	Flowers per raceme	10-0/12-1/15-2/22-3/26-4/30-5/35-6	Number
17.	Bract length	2-0/2.5-1/3-2/4-3	mm
18.	Pedicel length	1-0/1.5-1	mm
19.	Calyx length	5-0/6-1/6.5-2/7-3	mm
20.	Length of calyx tube	1.5-0/2-1	mm
21.	Length of calyx teeth	3-0/3.5-1/4-2/5-3/5.5-4	mm
22.	Standard length	6-0/7-1/9-2/10-3	mm
23.	Standard width	2.5-0/3-1/4-2/4.5-3/5-4	mm
24.	Keel length	6-0/8-1/9-2/10-3	mm
25.	Keel width	2-0/3-1/4-2	mm
26.	Wing length	2-0/2.5-1/3-2/3.5-3/4-4	mm
27.	Wing width	1-0/1.5-1/2-2	mm
28.	Wing claw length	0.5-0/1-1/1.5-2	mm
29.	Pod length	5-0/6-1/7-2	mm
30.	Pod width	3.5-0/4-1/4.5-2/5-3	mm
31.	Crest spines	1-0/3-1/4-2/5-3/6-4	Number
32.	Areoles per pod	4-0/5-1/7-2/8-3/9-4	Number
33.	Seed length	2.5-0/3-1/4-2	mm
34.	Seed width	1.5-0/2-1/2.5-2	mm
No.	Characters	Code	Numerical code
35.	Leaflet shape	1-0/2-1/3-2	1-oblong-elliptic 2-oblong-elliptic 3-linear-oblong
36.	Hair state of abaxial surface of leaflet	1-0/2-1	1-sparse 2-dense
37.	State of stem hair (density)	1-0/2-1	1-sparse 2-rare
38.	State of stem hair	1-0/2-1	1-appressed 2-protuberant
39.	State of peduncle hair	1-0/2-1	1-sparse 2-dense
40.	Stipule shape	1-0/2-1	1-lanceolate 2-triangular
41.	State of stipule hair	1-0/2-1/3-2	1-glabrous 2-ciliate 3-sparse
42.	State of bract hair	1-0/2-1/3-2	1-glabrous 2-ciliate 3-sparse
43.	State of pedicel hair	1-0/2-1	1-dense 2-sparse
44.	State of pod hairs	1-0/2-1	1-dense 2-sparse

Morphological data

The studied specimens (three in each population) were assessed by biometric data: 34 quantitative and 10 qualitative morphological characters were chosen and evaluated (Table 2). The phe-

netic analysis was carried out with three individuals (a, b, c) per population using the unweighted pair-group method with arithmetic averages (UPGMA) (Sneath, Sokal, 1973) and percent similarity coefficients in MVSP Vers. 3.22 (Kovach, 1985–2013). The results are illustrated in a phenogram.

Eco-geographic data

To record the coordinates, global positioning system (GPS) device was used. Maps of the areas from where the samples had been collected were developed using DIVA-GIS software (Version 7.3.0.1) (Fig. 1). In particular, the distribution of *Onobrychis* species, localization, and their collection site were identified (Table 1).

Eco-geographic data were encoded and analyzed with Past 4.03 (Hammer et al., 2001) by the principal component analysis (PCA) method.

Data obtained from each site included altitude, longitude, latitude, slope inclination, slope orientation, min. and max. annual temperature, mean annual precipitation, relief and soil traits including texture, humus %, organic carbon (OC %), total nitrogen (Total N %), pH, phosphorus pentoxide, potassium oxide, and magnesium. Climatic information for each collecting site was obtained from the Bureau of Hydrometeorological Forecasts of the Department of National Hydrometeorology of the Ministry of Ecology and Natural Resources of Azerbaijan (Table 3).

Results

The results of the morphometric analysis are represented in an UPGMA phenogram with percent similarity coefficients (Fig. 2). All individuals derived from the same population cluster together. There are three groups with similarity level 94.6%: populations 3, 4 formed group (A), population 6 formed a separate group (B), and populations 1, 2, 5, 7, 8, and 9 clustered as group (C).

Eco-geographic analysis, as shown by PCA, illustrates the relative importance of each environmental variable in the distribution of the populations. The PCA analysis of the populations based on the overall similarity of the eco-geographic data showed three groups (Fig. 3).

Discussion

Onobrychis altissima is an Iranian element. The geographic distribution range of *O. altissima* in Azerbaijan is not limited, and the species occurs in almost the whole of Azerbaijan. *O. cyri* is a Caucasian element. By geographic range, *O. cyri* is distributed in all regions of Azerbaijan. As a detailed description of *O. altissima* and *O. cyri* is not found, we present here a relevant morphological circumscription. This description differs in some morphological characters from the populations in Turkey (Hedge, 1970), Armenia (Navasardyan et al., 2009; Takhtajan, 1962), Georgia (Khinthibidze, 1981), and Iran (Toluei et al., 2013b) (Table 4).

O. altissima Grossh. – Zap. Nauchno-Prikl. Otd. Tiflissk. Bot. Sada 6: 141 (1929) (Tamamshyan, 1954) – Perennial plant is high, up to 90 cm. Stems are erect, almost glabrous. Stipules are free, wide triangle, acuminate with loose, ciliated edge. Lower leaves are long petiolate, of six to eight pairs. Leaflets are 12–14 mm long, 5–7 mm wide, oblong-oval or elliptic, obtusely acuminate, and almost glabrous. Upper leaves are six to eight pairs, with leaflets that are 12–20 mm long and 5–6 mm wide. Peduncle is two or three times as long as a leaf. Racemes are cristate, during fruit elongating. Bract is lanceolate-subulate, long pilose, and two or three times as long as tube. Corolla is up to 12 mm long and pink with dark veins. Standard is equal to the wing. Legume is semi-

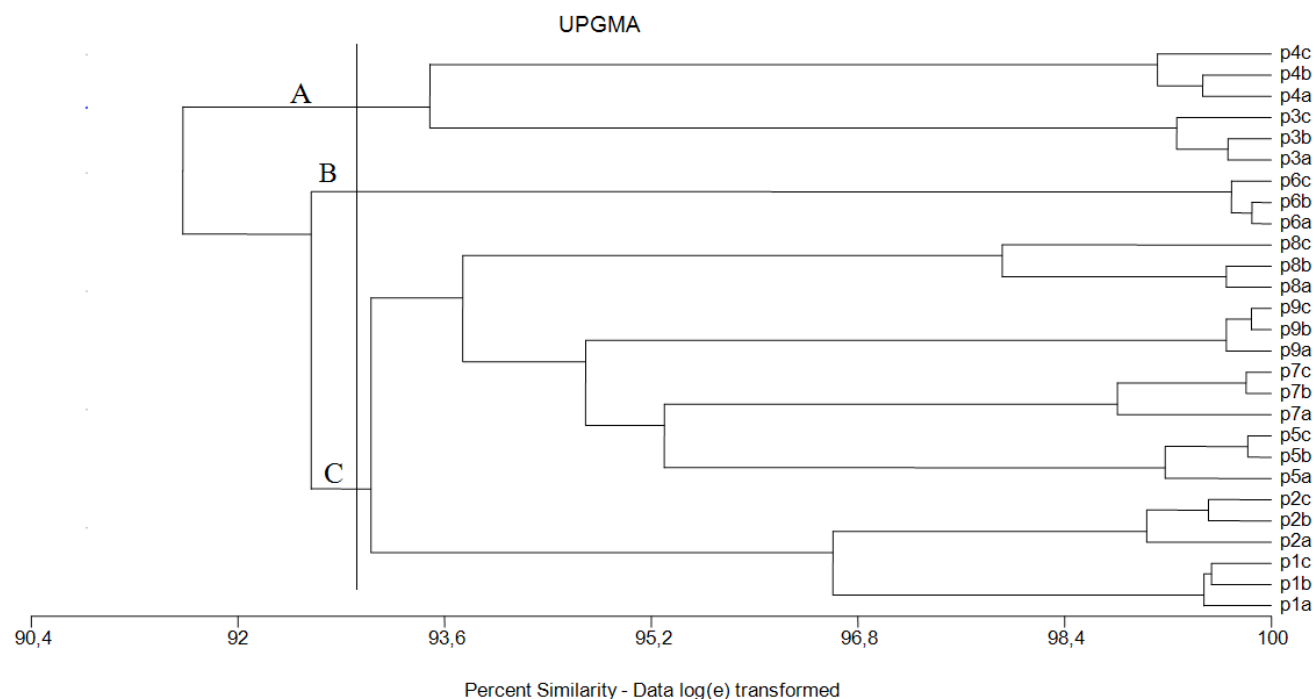


Fig. 2. UPGMA phenogram based on the analysis of morphological data in three individuals (a, b, c) per population (1–9) of *Onobrychis altissima* and *Onobrychis cyri*.

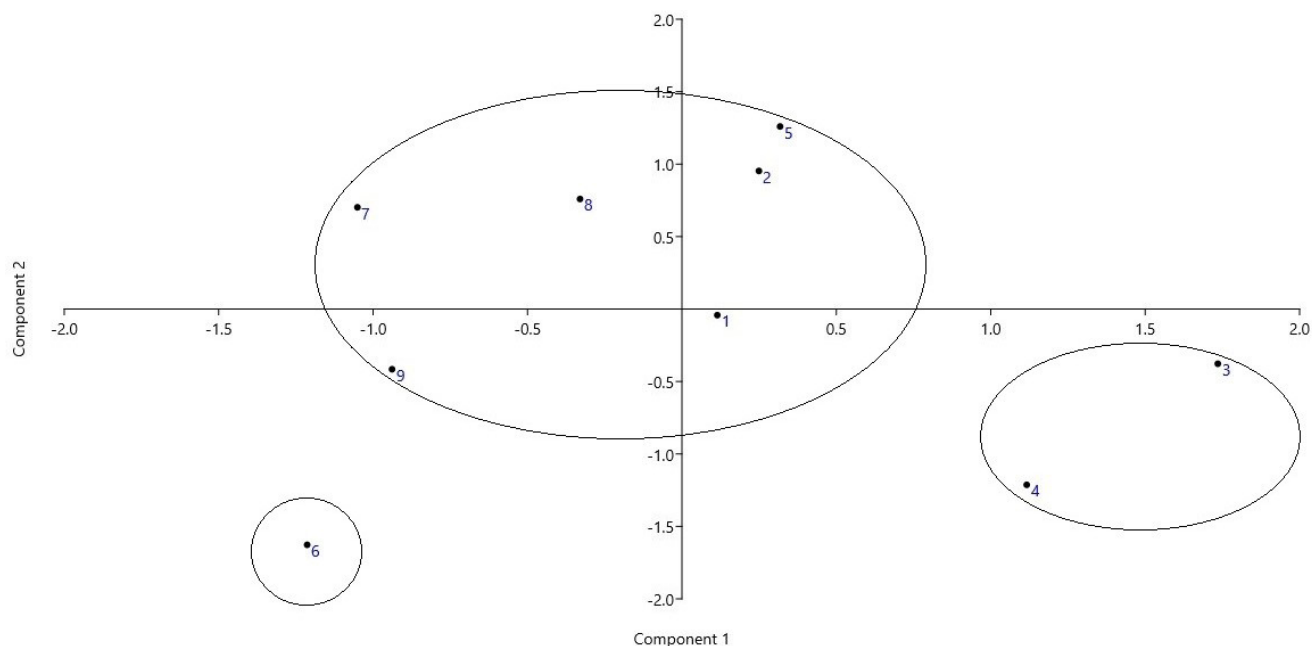


Fig. 3. PCA of nine populations of *Onobrychis altissima* and *Onobrychis cyri* in Azerbaijan based on the 15 eco-geographic data.

Table 3. Eco-geographic variables of *Onobrychis altissima* and *Onobrychis cyri* populations.

Eco-geographic factors	Population numbers								
	1	2	3	4	5	6	7	8	9
Altitude (m)	1340	1420	2209	1870	1459	614	719	1107	771
Slope inclination%	16–30	8–16	8–16	8–16	30+	0–3	16–30	30+	8–16
Slope orientation	SW	SW	SE	S	N	NW	S	SE	E
Max. temp (°C)	38	38	40	36	35	38	32	35	37
Min. temp (°C)	–5	–5	–12	–10	–11	–9	–2	–5	–7
Ann. prec. (mm)	499	599	402	338	627	385	622	599	501
Relief	O5	R4	R4	R4	E6	L2	O5	E6	R4
Texture	HO	HO	SC	SA	HO	CY	GR	ST	ST
HU %	3.58	3.98	3.48	2.49	4.13	3.40	5.02	6.01	5.60
OC%	5.4	4.13	4.11	5.19	5.10	2.20	6.4	4.79	5.02
pH	6.1	7.2	7.9	6.5	7.3	7.3	6.7	7.2	6.9
Total N%	0.68	0.38	0.28	0.19	0.35	0.29	0.34	0.52	0.18
P ₂ O ₅ (mg/kg)	0.22	0.14	0.12	0.1	0.19	0.17	0.23	0.27	0.21
K ₂ O (mg/kg)	1.46	2.2	2.20	1.45	2.24	2.16	2.79	3.5	2.0
Mg (mg/kg)	2.3	2.8	2.7	6.2	2.6	2.5	7.3	9.4	9.3

Notes: Slope orientation - S- south; SW - southwest; SE - southeast; N - north; NW - northwest; E - east; relief - O5- foothill; R4 - hill; E6 - steep slope; L2 - plain; texture - HO- high humus; SC - sandy clayey; SA - sandy; CY - clay soil; GR - grave; ST - stony.

orbicular, 5–8 mm long, short pilose, with a crest of width 1.5–2 mm, unarmed or with shorter spines.

O. cyri Grossh. – Zap. Nauchno-Prikl. Otd. Tiflissk. Bot. Sada 6: 123 (1929) (Tamamshyan, 1954) – Perennial plant of height 30–60 cm, rarely of 50–80 cm height. Stems are branched and appressed hairy. Stipules are ovoid and acuminate. Lower leaves are long petiolate, of 8–10 pairs. Leaflets are 10–12 mm long, (3)

4–5 mm wide, oval or elliptic, and rarely elongated (30–40 mm long). Upper leaves are short petiolate, with 7–12 elliptic or lanceolate-linear leaflets. Peduncle is two or three times as long as the leaf. Racemes are cristate before flowering, during elongation of fruit. Bract is 2 mm long. Calyx teeth are two or three times as long as the tube. Corolla is pink and 6–12 mm long. Legume is semi-orbicular, appressed pilose, with spines and cavities on the

Table 4. Comparison of *Onobrychis altissima* and *Onobrychis cyri* with morphologically similar species (1- *O. altissima*, 2- *O. cyri*).

No.	Characters	Azerbaijan populations		Iran populations	Flora of Turkey	Flora of Georgia		Flora of Armenia		Flora of Azerbaijan	
		1	2	1	1	1	2	1	2	1	2
1.	Plant height (cm)	63–115	46–86	38–125	50–90	70–100	30–60	72–105	48–75	90	30–80
2.	Stipule length (mm)	5–10	4–7	3–16	5	-	-	-	-	-	-
3.	Number of leaflets (in pairs)	15–17	11–21	4–11	6–8	6–8	7–12	6–8	7–12	6–8	7–12
4.	Leaflet length (mm)	10–20	10–20	6–43	12–20	12–30	10–15 (17)	-	-	-	10–40
5.	Leaflet width (mm)	2–7	1.5–4	(1) 4–10.5	4–6	5–36	34	-	-	-	(3) 4–5
6.	Bract length (mm)	3–4	2–3	3	-	-	-	-	-	-	2
7.	Calyx length (mm)	5–6	6–7	5–9	7	8–10	-	-	-	-	-
8.	Standard length (mm)	6–10	7–10	8–15	9.5–11	11–13	-	-	-	12	6–12
9.	Keel length (mm)	6–10	8–10	7–15	9.5–11	-	-	-	-	-	-
10.	Wing length (mm)	2–4	2–4	2.7–5	2.5–4	-	-	-	-	-	-
11.	Pod length (mm)	5–6	6–7	5–7(8)	5–7 (8)	5–8	-	5–7	3.5–5	5–8	-
12.	Pod width (mm)	4–4.5	3.5–5	4–5.5	-	-	-	3–5	3–3.5	-	-
13.	Number of crest spines	1–4	4–6	-	-	-	-	-	-	-	5–7
14.	Seed length (mm)	3–4	2.5–3	3–3.2	-	-	-	-	-	-	-
15.	Seed width (mm)	2–2.5	1.5–2.5	2.3–2.5	-	-	-	-	-	-	-

disc; the edge cavities are big, and the crest has five to seven wide triangle teeth.

Based on morphological variations, three groups were identified; here, we have presented the description of each group based on important and distinguishing features.

O. altissima Group A (populations 3, 4): plant height 63–65 cm, stem length 45–50 cm, inflorescence 5–12 cm long, peduncle 15–17 cm long, length of leaflet 10–20 mm, width of leaflet 2–6 mm, pods 6×4–4.5 mm.

O. cyri Group B (population 6): plant height 75–86 cm, stem length 60–71 cm, inflorescence 5 cm long, peduncle 20 cm long, length of leaflet 10–20 mm, width of leaflet 2.5–4 mm, pods 6×5 mm.

Group C (populations 5, 7, 8, 9) – *O. cyri*: plant height 46–65 cm, stem length 30–45 cm, inflorescence 4–10 cm long, peduncle 14–20 cm long, length of leaflet 10–15 mm, width of leaflet 2–6 mm, pods 6×4–4.5 mm; (populations 1, 2) – *O. altissima*: plant height 90–115 cm, stem length 74–90 cm, inflorescence 12–17 cm long, peduncle 25–30 cm long, length of leaflet 10–20 mm, width of leaflet 5–6 mm, pods 4.5 mm

Our results showed that three main groups based on overall similarity of eco-geographic data are in concordance with three morphological groups. The morphological character of each species is in close correlation with a combination of ecological factors, representing the best criteria for the diagnosis of its ecological factors. Based on the PCA analysis result, populations 1, 2, 5, 7, 8, and 9 formed one group in terms of their closer environmental index. Among the 2 and 5 populations are the same region (Gadabey district), are located close to each other. Although *O. altissima* was collected from population 2, *O. cyri* was collected from population 5. According to the results of the analysis, population 6 is separated from the other populations with the lowest altitude, slope inclination, and orientation. Besides, the Guba district is one of the warmer and arid regions, from where the specimens of *O. cyri* were collected. Populations 3 and 4 were located close to each other because of the higher altitude, the minimum percentage of humus and phosphorus

pentoxide, and soil texture. Our results showed that the three groups based on overall similarity of eco-geographic data are in concordance with the morphological groups.

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