Rendiconti Lincei. Scienze Fisiche e Naturali
https://doi.org/10.1007/s12210-023-01208-7

RESEARCH PAPER

A comparative anatomical study on leaf and scape of *Androsace* taxa (Primulaceae): contribution to *Androsace* taxonomy

Sibel Ulcay[1] · Abdurahman Sefali[2] · Yakup Yapar[3] · İbrahim Demir[4] · Betül Gıdık[5]

Received: 10 August 2023 / Accepted: 24 October 2023

Abstract

In the present study, the leaf and scape anatomical features of ten *Androsace* taxa distributed in Türkiye were defined to contribute to plant identification. Hand sections from both the scape and leaf examined 43 anatomical characters. Principal component analysis (PCA) and hierarchical cluster analysis (HCA) were performed for the first time for *Androsace*. The results show that there are differences as well as similarities between *Androsace* species. It is noteworthy that *Androsace artvinensis* forms a separate main group with HCA. In addition, the fact that two of the five basic components are represented by a single variable according to the PCA results suggests that the anatomical characters examined are determinative for *Androsace* taxa.). In PCA, eigenvalues of PC1 (2.435), PC2 (1.749), PC3 (1.369), PC4 (1.223), and PC5 (1.006) were greater than 1.0. Cortex width, aerenchyma, arrangement of vascular bundles, and four-branched non-glandular hairs are among the important anatomical characteristics of the scape. Leaf cross-sectional shape, epidermis dimensions, glandular hair types, presence of five branched non-glandular hairs, stoma sizes, presence of crystals in the mesophyll, and types are among the qualitative and quantitative anatomical characteristics that attract attention in the leaf. This study examined almost all taxa of the genus *Androsace* distributed in Türkiye. The results will be important in finding new species among the examined taxa and revealing the anatomical features of the taxa for the first time. They will contribute to other studies on the genus.

Keywords Anatomy *Androsace* · Leaf · Scape · Cluster analysis · Primulaceae

1 Introduction

The family Primulaceae consists of about 22 genera with approximately 1000 species and is mainly distributed in the high mountains of the northern temperate zone (Boucher et al. 2016; Bai 2020). The family is among the world's three most critical horticultural plants (Richards 2014; Tütüncü 2020). The family has two large genera, *Primula* (1753: 142) and *Androsace* Linnaeus (1753: 141) (Roquet et al. 2013; Baasanmunkh 2020). *Androsace* is distinguished from the genus *Primula* by having a corolla future (narrowed-annular throat and a short corolla tube) (Xu et al. 2016).

The genus *Androsace* is encountered with many well-known alpine plant members through studies, such as climate change and plant ecology (Schönswetter et al. 2003a, 2003b; Grabherr et al. 2010; Dentant et al. 2018; Bonelli et al. 2021; Eustacchio et al. 2023). The genus consists of about 158 taxa distributed in the northern hemisphere's extratropical mountain ranges (Mabberley 2008; Jacquemoud and Jordan 2020; Sefalı 2021; Sefalı and Yapar 2022). The ancestor of the genus is thought to have come from Asia,

✉ Sibel Ulcay
sibelulcay@gmail.com

Abdurahman Sefali
asef4petal@gmail.com

Yakup Yapar
yyapar25@gmail.com

İbrahim Demir
hosap65@gmail.com

Betül Gıdık
betulgidik@bayburt.edu.tr

1 Department of Field Corps, Faculty of Agriculture, Kırsehir Ahi Evran University, Bağbaşı, 40100 Kırsehir, Turkey

2 Department of Primary Education, Faculty of Education, Bayburt University, Bayburt, Turkey

3 Department of Biology, Faculty of Science and Art, Bingöl University, 12000 Bingöl, Turkey

4 Department of Biology, Faculty of Arts and Sciences, Bitlis Eren University, Bitlis, Turkey

5 Department of Organic Farming Management, Bayburt University, 69000 Bayburt, Turkey

Published online: 11 November 2023

in terms of biogeography, 35 million years ago (Boucher et al. 2016). This ancestral species from Asia spread through the mountain ranges of the Northern Hemisphere, first reaching Europe and then North America (Schönswetter et al. 2003b; Roquet et al. 2013). Adaptation of the *Androsace* to the harsh environmental conditions, especially the cold climate, in the mountains increased the diversity (Boucher et al. 2012; Roquet et al. 2013). The recent discoveries of new *Androsace* taxa in the Eurasian mountains show that we know little about plant diversity on mountain summits (Dentant 2018).

The genus *Androsace* has been consisting of six distinct sections (*Chamaejasme* Koch, *Douglasia* (Gray) Wendelbo, *Androsace* (syn. sect. *Andraspis* (Duby) Koch), *Pseudoprimula* Pax., *Aretia* (L.) Duby, and *Aizoidium* Hand.-Mazz.) (Smith and Lowe 1997). Xu et al. (2016), whose research shows that these sections are changeable, added *Samuelia* Schlechtd., *Mirabiles* (Hand.-Mazz.) Yang & Huang, and *Orthocaulon* Hand.-Mazz. sections. Throughout the history of plant taxonomy, the classification of plants is constantly changing and is a matter of discussion (Talebi et al. 2015). Türkiye has nine species, and two subspecies of *Androsace* belong to *Androsace* (syn. sect. *Andraspis* (Duby) Koch) and *Chamaejasme* Koch sect. (Davis 1980; Davis et al. 1988; Smith and Lowe 1997; Sefalı 2021; Sefalı and Yapar 2022). The sect. *Chamaejasme* includes *Androsace villosa* L. and *A. caduca* Ovcz, while the sect. *Androsace* includes *Androsace albana* Steven, *A. maxima* L., *A. armeniaca* Duby, *A. intermedia* Ledeb, *A. azizsancarii* Sefalı, *A. artvinensis* Sefalı & Yapar, and *A. multiscapa* Duby (Sefalı 2021; Sefalı and Yapar 2022).

The comparative study of plant structure, morphology, and anatomy has always been the backbone of plant systematics, which seeks to elucidate plant diversity, phylogeny, and evolution. The second half of the twentieth century has been a fascinating period in which systematic and structural studies benefited greatly from new techniques and methods (Boonprajan et al. 2023; Villalva et al. 2023). Although morphological features are one of the main factors in identifying plants, morphological features may be insufficient occasionally. In cases where morphological characters are insufficient, anatomical features are used (Birjees et al. 2022; Marzinek et al. 2022; Raza et al. 2022). Plant anatomy reveals phylogenetically important features (Glos et al. 2022; Boonprajan et al. 2023).

Anatomical studies on *Androsace* taxa are very limited and scarce. This research aimed to determine the anatomical features of 10 *Androsace* taxa distributed in Türkiye. The anatomical features of the species belonging to the genus will be brought to the literature for the first time. In addition, this study aims to use anatomical characters in the diagnosis of taxa belonging to the *Androsace*, which is a taxonomically problematic genus, and to reveal the similarities and differences of taxa through statistical analysis of anatomical features.

2 Materials and methods

Plant samples of 10 taxa naturally distributed in Türkiye were collected, their photographs were taken in nature (Canon EOS 60D), and locality and habitat information were given (Fig. 1). During the diagnosis, "Flora of Turkey and the East Aegean Islands 6, 10 and 11" (Davis 1978; Davis et al. 1988) was used as the main source, and in addition to this work, species discovered in Türkiye in recent years A literature review was conducted for (Sefalı 2021; Sefalı and Yapar 2022). The Plant List (2010) and International Plant Name Index (2008) websites were used to determine the current names of taxa. The locality information and herbarium voucher numbers of the plant samples that are the subject of the research are presented in Table 1.

2.1 Anatomical methods

The collected fresh samples were fixed in 70% ethanol. Cross sections were taken by hand from the stem and leaves of the fixed specimens. Glycerin was used as the examination medium in the sections. The preparations were made semi-permanent with the glycerin–gelatin method (Vardar 1987). In addition, superficial sections were taken from the lower and upper surfaces of the leaves. Analyses were made on 25 measurements. Photographs were taken with the SOIF BK500-L microscope and the AmScope FMA050 camera system from the transverse and superficial sections (Figs. 2, 3).

2.2 Statistical analysis

The statistical analysis of this study was performed using the SPSS 26.0 software program. This study applied principal component analysis (PCA) to determine the main components and hierarchical cluster analysis (HCA) to determine the relationship between *Androsace* spp. Hierarchical cluster analysis (HCA) is a clustering method that explores the organization of samples. Furthermore, it allows for determining similarities and differences within and between groups by depicting a hierarchy (Lee and Yang 2009).

3 Results

3.1 Stem anatomy

Twenty qualitative (Table 2) and quantitative anatomical features (Table 3) of the handle are investigated. According to

Fig. 1 *Androsace* species of Türkiye: **A** *A. villosa,* **B** *A. caduca,* **C** *A. maxima,* **D** *A. armeniaca* var. *macrantha*, **E** *A. armeniaca* var. *armeniaca,* **F** *A. artvinensis,* **G** *A. albana,* **H** *A. multiscapa,* **I** *A. azizsancarii* and **J** *A. intermedia*

the cross-sectional width, some species' scapes were polygonal (undulate-edged), and some were circular (Table 2). The epidermis cells in the outermost part of the stem are usually polygonal in shape and have a single row. In all species, there is a cuticle layer on the epidermis cells. The epicuticular part, which forms the outermost part of the cuticle,

Table 1 Locality and herbarium voucher numbers of the studied taxa

Taxa	Locality	Voucher number
Section *Chamaejasme* Koch		
Androsace villosa L	Bayburt, Kop Mountains, Bahtlı Mountain, stony places, 2800 m, 20 Jun 2018, A. Sefalı 394	BIN
A. caduca Ovcz	Van, Başkale, İspiriz Mountain, stony places, 3600 m, 13 Jun 2021, A. Sefalı 727	BIN
Section *Androsace* (syn. sect. *Andraspis* (Duby) Koch)		
Androsace albana Steven,	Bayburt; south side of Anzer Mountain, 2860 m, 5 Jul 2020, A. Sefali 494 Ardahan: Artvin to Ardahan, Sahara Pass, on the cap of the hills, 2300 m, 10 June 2021, A. Sefalı 691 Giresun: Karagöl Mountain, 2700 m, 18 June 2021, A. Sefalı 702 Kastamonu: Ilgaz Mountain, 2300 m, 30 June 2021, A. Sefalı 721	ISTE 117268 BIN BIN BIN
A. maxima L.,	Bayburt; Aslandede Village, about 1700 m, 18 May 2021, A. Sefali 639	BIN
A. armeniaca Duby var. *armeniaca*	Ardahan; Artvin to Ardahan, Sahara Pass, on the cap of the hills, 2300 m, 10 June 2021, A. Sefalı 688	VANF 165226
A. armeniaca Duby var. *macrantha* (Boiss. & A.Huet) Martelli	Bayburt; Kop pass, sandy places, 2200 m, 02 June 2021, A. Sefalı 661	VANF 165227
A. intermedia Ledeb	Artvin; Yusufeli, Sarıgöl, Taşkıran Village, rout of the Rindiker Highland, 1650 m, 09 June 2021, A. Sefalı 686	BIN
A. azizsancarii Sefalı	Bayburt; Soganlı Mountains, south of Anzer Mountain, on moraines, 2831 m, 1 July 2020, A. Sefalı 507	BIN 9405
A. artvinensis Sefalı & Yapar	Artvin: southern foothills of the Kackar Mountains, Yaylalar Village, forest side, 2129 m, 40°51′N, 41°16′E, 09 June 2021, A.Sefalı 686	BIN 10099
A. multiscapa Duby	Niğde, Ulukışla, Horoz Village, volcanic gravel, 2750–2900 m a.s.l., 14 June 2021, A. Sefalı 694	VANF 165224

differed in species. The epicuticle layer has flat, highly wavy, slightly wavy margin shapes (Table 2). The stem surface was glabrous in *A. intermedia*. Indumentum density is the highest taxon *A. armeniaca* var. *armeniaca*.

Papillae have been identified in species, such as *A. artvinensis*, *A. multiscape*, *and A. azizsancari*. *A. armeniaca* var. *armeniaca*, and *A. villosa*. Non-glandular cover hair types also differ among taxa. Taxa such as *A. armeniaca* var. *macrantha*, *A. intermedia*, *A. caduca*, *A. albana,* and *A. armeniaca* var. *armeniaca* do not have simple, non-glandular hairs. Separately from the simple non-glandular hair, branched non-glandular hair is also noteworthy. In non-glandular hairs, the number of branches is 2–3. Only in *A. artvinensis* were *4* branched non-glandular hairs found. The richest species in terms of glandular hairs is *A. armeniaca* var. *armeniaca*. No glandular hairs are found in A. *intermedia*. Goblet-shaped glandular hairs are found only in taxa, such as *A. artvinensis* and *A. multiscapa*. In scape cross sections, the cortex parenchyma cells are circular or polygonal in shape. In particular, the presence of idioblasts in the cortex of A. *intermedia* is remarkable. Secretory spaces are formed between these parenchymatous cells. The cortex of taxa is usually narrowed, while only the cortex of *A. caduca* covers a large area. Aerenchyma is observed in A. *caduca.* The sclerenchyma consists of multi-row cells and is in the form of a continuous ring (except for *A. maxima* and *A. villosa*). There is no sclerenchyma in the *A. caduca;* a row of collenchyma cells is formed just below the epidermis. The pith region is usually filled with parenchymatous cells. In taxa, vascular bundles are usually spaced apart (except *A. caduca*). The phloem is 5–10 layered and prominent. The most phloem layer was formed in *A. artvinensis* (10 layered). The cambium is prominent in some species and is in 1–4 rows (*A. multiscapa*, *A. albana, A. armeniaca* var. *armeniaca, and A. armeniaca* var. *macrantha*).

3.2 Leaf anatomy

When investigating leaf anatomy, 22 qualitative and quantitative anatomical features of the studied taxa are determined (Tables 2, 3). The leaves reveal different shapes according to the midrib cross sections taken from the examined taxa: shuttle elliptical, linear, oblong, two-knuckle shuttle, or pointed shuttle. The midrib region is not prominent in species, such as *A. azizsancari*, *A. caduca, A. armeniaca* var. *macrantha,* and *A. maxima*. The epidermis cells in the outermost part of the leaf are generally circular and polygonal in shape. The anticlinal walls of the epidermis cells differ between taxa and are flat, slightly curved, curved, and highly curved. Very thick anticline cell walls are formed in *A. armeniaca* var. *macrantha* and *A. villosa*. Simple, 2–5-branched cover hairs are formed on the epidermis. No non-glandular cover hairs are found on A. *intermedia* and *A. villosa*. Again, glandular hairs were determined in all taxa.

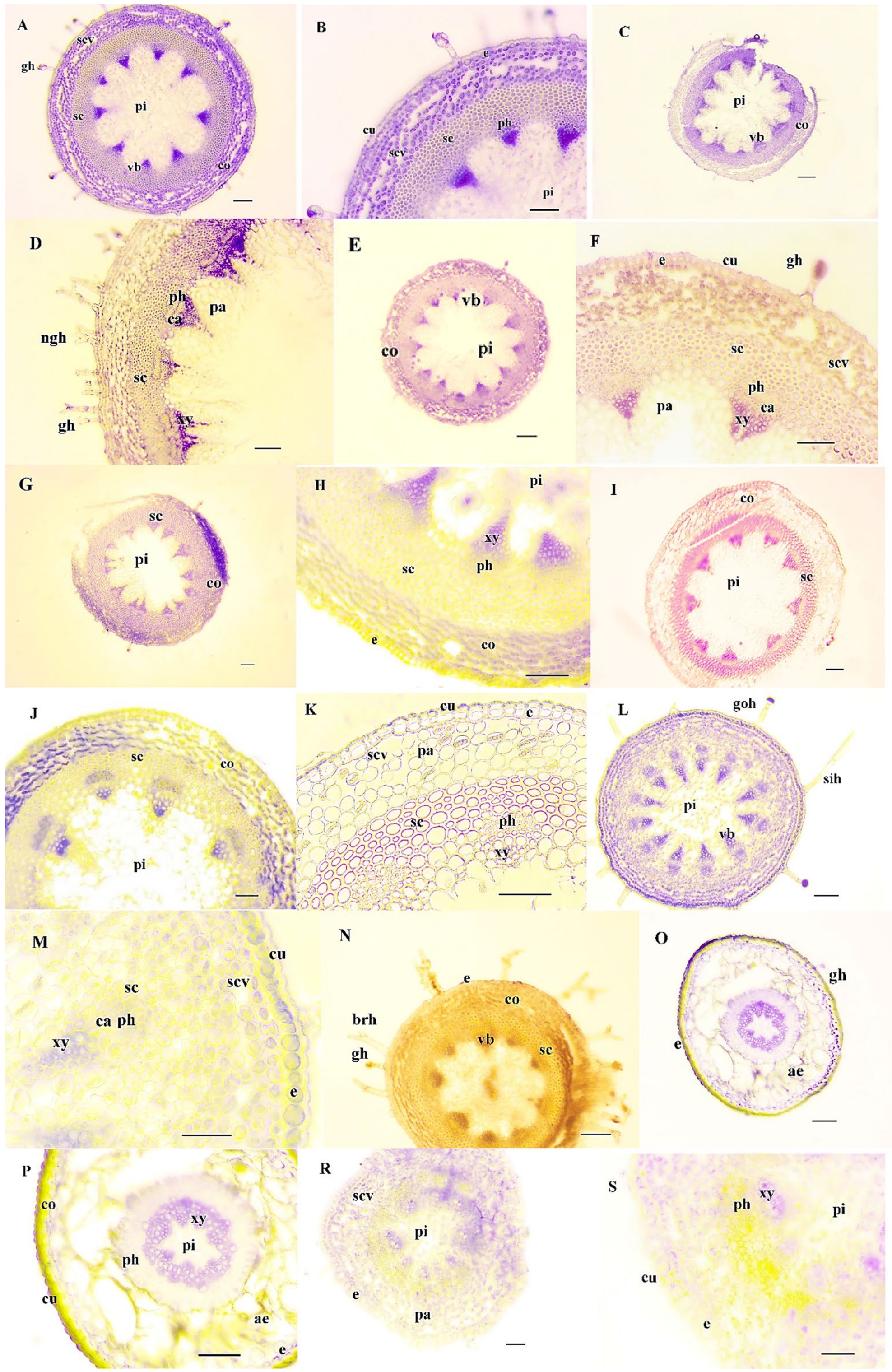

Fig. 2 Scape cross sections of taxa. **A**, **B** *A. albana*, **C**, **D** *A. armeniaca* var. *armeniaca***,** **E**, **F** *A. armeniaca* var. *macrantha*, **G**, **H** *A. artvinensis*, **I** *A. azizsancarii*, **J**, **K** *A. intermedia*, **L**, **M** *A. maxima*, **N**, **O** *A. multiscapa*, **O**, **P** *A. caduca*, **R**, **S** *A. villosa*. ae—brh—branched non-glandular hair, *ca* cambium, *co* cortex, *cu* cuticle, *e* epidermis, *gh* glandular hair, *goh* goblet-shaped glandular hair, *ngh* non-glandular hair, *sc* sclerenchyma, *scv* secretory cavity, *sih* simple non-glandular hair, *pa* parenchyma, *ph* phloem, *pi* pith, *xy* xylem, *vb* vascular bundles

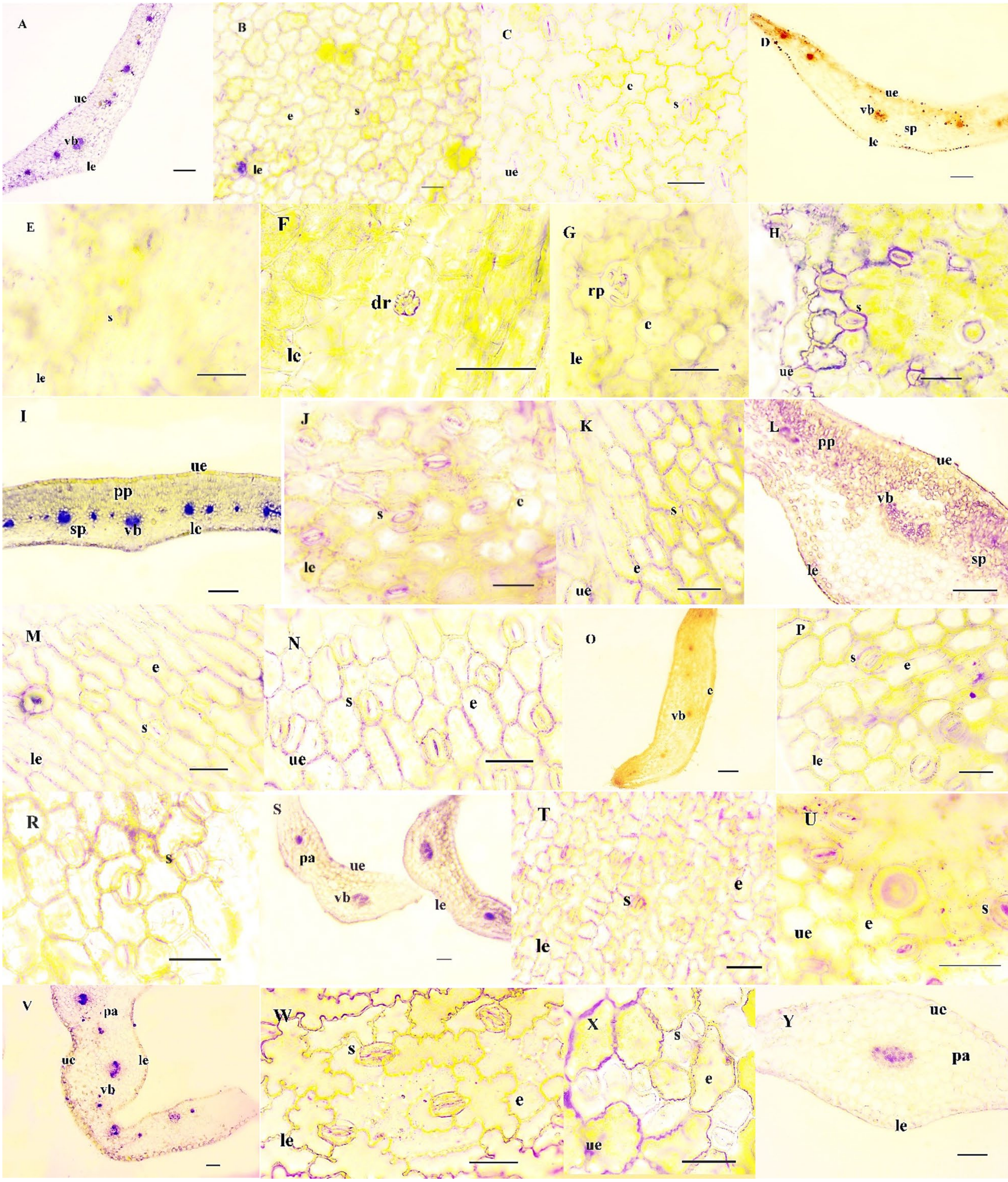

Fig. 3 Leaf cross sections and superficial sections of taxa. **A, B**, **C** *A. albana*, **D**, **E**, **F**, **G**, **H** *A. armeniaca var. armeniaca*, **I**, **J**, **K** *A. armeniaca* var. *macrantha*, **L**, **M**, **N** *A. artvinensis*, **O**, **P**, **R** *A. aziz-sancarii*, **S**, **T**, **U** *A. intermedia*, **V**, **W**, **X** *A. maxima*, **Y**, **Z**, **AB** *A. multiscapa*, **AC**, **AD**, **AE** *A. caduca*, **AF**, **AG**, **AH** *A. villosa*. **AI** glandular hair, **AJ**–**AL** goblet-shaped glandular hair, **AM** mushroom-shaped hairs, *e* epidermis, *le* lower epidermis, *dr* druse crystal, *pa* parenchyma, *ph* phloem, *pp* palisade parenchyma, *rp* raphide crystal, *s* stomata, *sp* spongy parenchyma, *xy* xylem, *ue* upper epidermis, *vb* vascular bundles

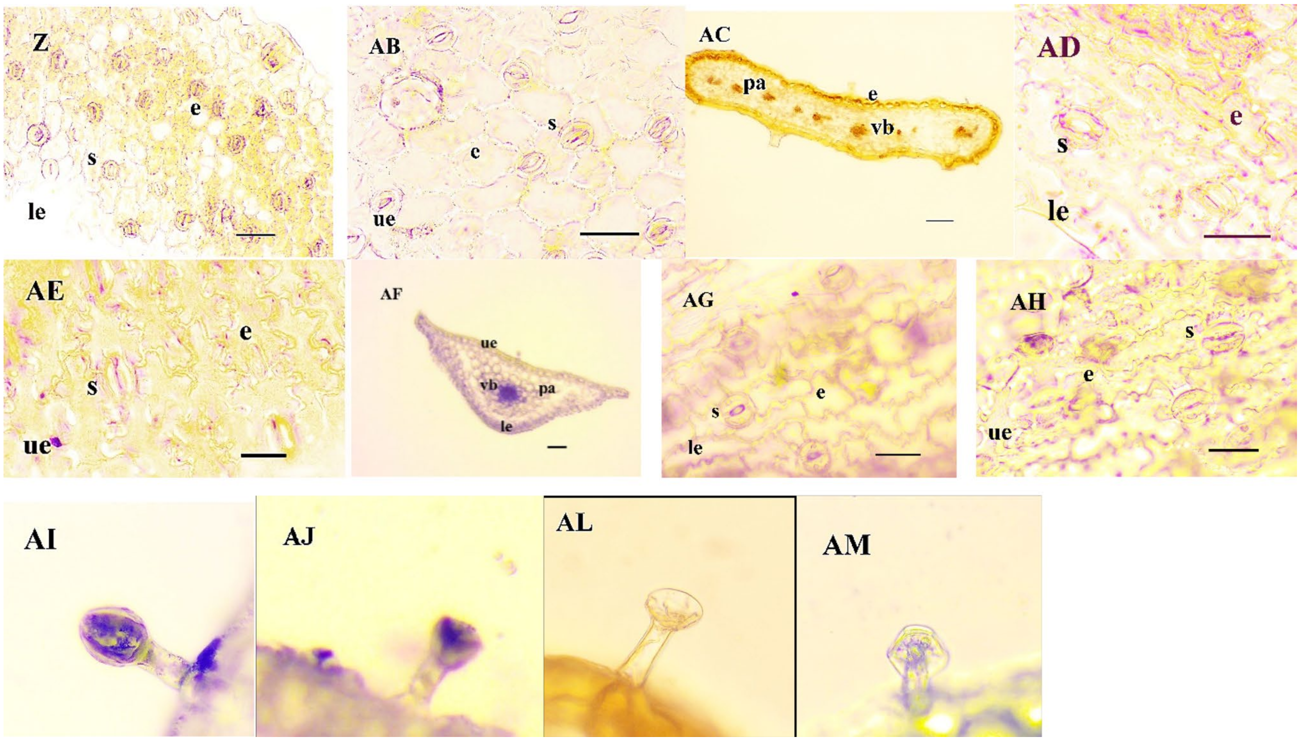

Fig. 3 (continued)

In particular, goblet and mushroom-shaped hairs are determined only in taxa, such as *A. artvinensis* and *A. caduca*. In *A. villosa*, the cuticle is noticeably thicker than in other taxa. The mesophyll of taxa has two different structures. While the mesophyll of taxa such as *A. artvinensis, A. albana,* and *A. armeniaca* var. *macrantha* is of bifacial type, unifacial mesophyll formation was determined in other taxa (Fig. 3).

The palisade cells are in 1–4 layers in the bifacial mesophyll. Spongy parenchyma cells in all taxa are circular. In species, such as *A. multiscapa* and *A. villosa*, one vascular bundle is formed in the midrib region, while the number of vascular bundles varies between 6 and 14 in other taxa. Raphides and druse crystals (*A. armeniaca* var. *armeniaca*) were formed in mesophyll parenchyma cells. There are stomata on both the lower and upper surfaces of the leaves, and they are of the amphistomatic leaf type. The anomocytic stoma is determined in all taxa. In addition, anisocytic stomata are found in taxa, such as *A. azizsancari*, *A. intermedia,* and *A. maxima*.

3.3 Statistical results

Principal component analysis (PCA) is known as a method that reveals the variance structure of the original p variable with fewer new variables that are the linear components of the variables. Principal components were determined for morphological features of *Androsace* spp. Eigenvalues and variance percentages of PCA analysis are provided in Table 4. The graph is provided in Fig. 4.

The HCA results show that the two main groups are further subdivided within themselves. While it is seen that the first main group is divided into more subgroups, it is noteworthy that only *A. azizsancarii* is in the second main group. This situation shows that *A. azizsancarii* is differentiated from other species regarding anatomical features. The number for clusters graphic of *Androsace* spp. is shown in Fig. 5.

The results of HCA are generally presented in a dendrogram, a plot that shows the organization of *Androsace* spp. and their relationships in tree form. There are two common approaches to resolving the grouping problem in HCA. They are divisive and agglomerative (Fig. 6).

The Kaiser–Meyer–Olkin (KMO) test was utilized for Principal Component Analysis to assess sample adequacy. Kaiser (1960) asserts that the KMO coefficient is unacceptable between 0 and 0.5, that 0.5 is the least, that 0.5 and 0.7 are the middle, that 0.7 and 0.8 are fine, that 0.8 and 0.9 are very good, and that 0.9 and above are great. Accordingly, principal components (PC) consisted of PC1—lower epidermis height and lower epidermis width; PC2—upper epidermis size and upper epidermis width; PC3—upper stomata width, upper stomata size, and lower stomata size; PC4—scape cortex general and PC5—width of lower stomata, respectively. This shows that there is a very important grouping. The width of the lower stomata and scape cortex

Table 2 Qualitative anatomical features of *Androsace* taxa

Characters	*A. albana*	*A. armeniaca var. armeniaca*	*A. armeniaca* var. *macrantha*	*A. artvinensis*	*A. azizsancarii*	*A. intermedia*	*A. maxima*	*A. multiscapa*	*A. caduca*	*A. villosa*
Secretory cavity	+	+	+	+	+	+	+	+	–	+
Layout of the vascular bundles	Intermittent	Intermittent	Intermittent	Intermittent	Intermittent	Intermittent	Intermittent	Intermittent	Continuous ring	Intermittent
Scape general shape	Full circle	Full circle	Undulate-edged	Full circle	Full circle	Undulate-edged	Full circle	Full circle	Undulate-edged	Undulate-edged
Cambium	2 layered	4 layered	2 layered	Indis	Indis	Indis	Indis	2 layered	Indis	Indis
Epicuticular layer	Flat	Higwavy	Higwavy	Higwavy	Sliwavy	Flat	Sliwavy	Sliwavy	Sliwavy	Sliwavy
Papilla	–	+	–	+	–	–	–	+	–	+
Simple cover hair	+	+	–	+	+	–	+	+	–	+
Branched non-glandular hair	2–5 branched	2–3 branched	2–3 branched	4 branched	–	–	–	2–3 branched	–	–
Aerenkima	–	–	–	–	–	–	–	–	+	–
Total number of hairs	13	64	12	19	7	–	15	27	–	22
Number of glandular hairs	15	18	1	3	3	–	7	6	–	11
Number of sclerenchyma layers	9	8	8	9	8	5	6	6	2	5
Pith	Full	Half full	Half full	Full	Full	Half full	Half full	Full	Full	Half full
Number of phloem layers	6	6	5	10	6	5	5	6	7	6
Goblet glandular hair	–	–	–	+	–	–	–	+	–	–
Sclerenchyma status	Conring	Conring	Conring	Conring	Conring	Conring	Tvasbund	Conring	–	Tvasbund
Leaf cross-sectional shape	Shuttle	Shuttle	Oblong	Shuttle	linear	Two-knuckle shuttle	Two-knuckle shuttle	elliptical	Oblong	Pointed shuttle
Mesophyll type	Bifacial	Unifacial	Bifacial	Bifacial	Unifacial	Unifacial	Unifasial	Unifacial	Unifacial	Unifacial
Leaf midrib	Distin	Distin	Indis	Distin	Indis	Distin	Indis	Distin	Indis	Distin
Number of vascular bundles	14	10	14	8	6	5	9	1	8	1
Number of sclerenchyma cells	–	–	–	4	–	–	2	–	–	–

Table 2 (continued)

Characters	*A. albana*	*A. armeniaca var. armeniaca*	*A. armeniaca* var. *macrantha*	*A. artvinensis*	*A. azizsancari*	*A. intermedia*	*A. maxima*	*A. multiscapa*	*A. caduca*	*A. villosa*
Non-glandular hair shape	2 branched	Simple, 2–3 branched	2–3 branched	2 branched	Simple	–	Simple	5 branched	Simple	–
Epidermis anti-cline walls	Curved	Slightly curved	Curved	Flat	Flat	Sightly curved	Highly curved	Curved,	Highly curved	Curved
Type of stomata	Anomocytic	Anomocytic	Anomocytic	Anomocytic	Anisocytic, Anomocytic	Anomocytic	Anisocytic, Anomocytic	Anomocytic	Anomocytic	Anomocytic
Presence of gladular hair	+	+	+	+	+	+	+	+	+	+
Goblet-shaped glandular hair	–	–	–	+	-	-	-	-	+	–
Mushroom-shaped hairs	–	–	–	+	–	–	–	–	+	–
Crystals	–	Raphide, druse	–	–	–	–	–	–	–	–

Indis indistinguishable, *Distin* distinguishable, *Conring* continuous ring, *Tvasbund* top of vascular bundles, *Sliwavy* slightly wavy, *Higwavy* Highly wavy

generally constituted single basic components, which have shown that they are important for separating *Androsace* taxa. According to Kaiser rules, eigenvalues greater than 1.0 are accepted as the principal component and descriptor of the variance (Kaiser 1960). In PCA, eigenvalues of PC1 (2.435), PC2 (1.749), PC3 (1.369), PC4 (1.223), and PC5 (1.006) were greater than 1.0.

4 Discussion

Environmental conditions highly influence most morphological features (Caferri and Bassi 2022). Although morphological features are one of the main factors in diagnosing plants, anatomical features are used in cases, where they are insufficient (Hameed et al. 2020; Gissi et al. 2022). In addition, although some anatomical characters are affected by environmental factors, these characters can make important contributions to plant taxonomy and biosystemic studies (Ozcan and Akinci 2019; Ulcay 2022a, 2023). Some morphological characters may be insufficient in distinguishing the genera belonging to the Primulaceae family (Xu et al. 2020). There are also problems in naming species in the genus *Androsace*, which belongs to the same family (Schneeweiss et al. 2004; Xu et al. 2020). In cases where morphological characters are missing or insufficient, anatomical features gain importance to facilitate diagnosis. This study determined some qualitative and quantitative anatomical features of 10 *Androsace* taxa distributed in Türkiye. The determined features will be useful in distinguishing taxa from a systematic point of view.

Scape anatomies of taxa are similar to each other. In general, a narrow cortex area is seen just below the single-row epidermis cells. Aerenchyma was observed only in the cortex of the *A. caduca*, which is wider than in other taxa. Scape structure can be considered an important distinguishing feature in distinguishing the *A. caduca*. The pith region of the taxa is usually filled with parenchymatic cells. According to the numerical and statistical analysis results, the scape widths are different between taxa.

Scape anatomies of taxa are similar to each other. In general, a narrow cortex area is seen just below the single-row epidermis cells. Notably, *A. artviensis*, a new species, has 4 branched non-glandular and goblet-shaped glandular hairs in its epidermis. Aerenchyma was observed only in the cortex of *A. caduca*, which is wider than in other taxa. Scape structure can be considered an important distinguishing feature in distinguishing *A. caduca*. The pith region of the taxa is usually filled with parenchymatic cells. According to the numerical and statistical analysis results, the scape widths are different between taxa.

The leaf shapes of some taxa of *Androsace* are grouped into five groups (shuttle, elliptical, linear, oblong, dioecious, ridged shuttle) (Table 2). *A. albana*, *A. azizsancari*, *A.*

Table 3 Quantitative anatomical features of the s *Androsace* taxa

Characters	*A. albana*	*A. armeniaca* var. *armeniaca*	*A. armeniaca* var. *macrantha*	*A. artvinensis*	*A. azizsancarii*	*A. intermedia*	*A. maxima*	*A. multiscapa*	*A. caduca*	*A. villosa*
The overall diameter of the scape cortex	530.43 ± 18.47	456.64 ± 14.35	476.62 ± 54.09	453.83 ± 14.05	440.61 ± 18.15	347.32 ± 11.99	271.93 ± 11.40	309.37 ± 8.51	506.38 ± 19.52	315.43 ± 18.89
Diameter of scape cortex parenchyma	23.60 ± 1.44	27.30 ± 1.14	14.78 ± 3.52	26.96 ± 1.14	18.92 ± 1.11	15.94 ± 1.03	20.46 ± 0.64	15.74 ± 1.05	24.02 ± 1.34	82.84 ± 66.42
The overall diameter of the scape center cylinder	1212.37 ± 23.84	1511.25 ± 45.87	1609.69 ± 140.92	1494.02 ± 47.25	1105.95 ± 36.06	751.20 ± 22.15	1089.44 ± 64.38	1793.96 ± 75.73	715.80 ± 26.27	431.09 ± 17.12
Diameter of scape pith parenchyma	32.43 ± 1.57	34.56 ± 2.29	28.73 ± 8,44	35.03 ± 2.25	18.86 ± 1.12	24.84 ± 1.22	37.16 ± 0.95	30.83 ± 1.19	20.98 ± 0.96	27.81 ± 1.17
Width of leaf upper epidermis	22.61 ± 0.84	22.05 ± 0.73	20.73 ± 5.18	22.12 ± 0.70	22.06 ± 0.85	26.00 ± 0.98	24.87 ± 0.62	25.25 ± 1.18	24.41 ± 0.6047	17.81 ± 0.81
The length of the upper epidermis of the leaf	29.57 ± 1.28	38.37 ± 1.01	33.47 ± 5.60	38.37 ± 1.01	28.49 ± 1.0	49.94 ± 1.55	50.54 ± 1.08	54.94 ± 0.77	36.13 ± 0.68	29.49 ± 0.90
Width of leaf lower epidermis	25.70 ± 0.75	14.48 ± 0.58	18.26 ± 3.08	14.58 ± 0.56	16.90 ± 0.85	25.52 ± 0.77	27.44 ± 0.88	25.25 ± 1.187	25.08 ± 0.55	20.44 ± 0.85
The length of the lower epidermis of the leaf	34.12 ± 0.62	16.91 ± 0.84	27.19 ± 2.98	16.81 ± 0.81	27.50 ± 0.99	47.99 ± 1.26	54.86 ± 0.99	55.46 ± 0.94	36.53 ± 0.68	27.36 ± 1.11
Diameter of leaf spongy parenchyma	23.50 ± 0.90	34.91 ± 1.59	21.05 ± 2.40	34.72 ± 1.54	28.74 ± 2.03	31.21 ± 1.71	29.048 ± 1.57	33.98 ± 2.18	31.39 ± 1.07	34.05 ± 2.66

Table 3 (continued)

Characters	*A. albana*	*A. armeniaca* var. *armeniaca*	*A. armeniaca* var. *macrantha*	*A. arvinensis*	*A. azizsancarii*	*A. intermedia*	*A. maxima*	*A. multiscapa*	*A. caduca*	*A. villosa*
Width of leaf lower stomata	29.34 ± 1.05	30.77 ± 0.74	28.40 ± 4.63	30.50 ± 0.76	1244.64 ± 1207.68	27.61 ± 1.03	30.04 ± 0.70	35.61 ± 1.47	33.05 ± 1.16	32.31 ± 2.29
Length of leaf lower stomata	53.04 ± 1.63	46.71 ± 1.28	46.20 ± 7.83	46.89 ± 1.24	50.68 ± 1.13	51.76 ± 1.53	54.43 ± 1.26	52.40 ± 1.66	44.07 ± 1.30	44.44 ± 1.58
Width of leaf upper stomata	39.34 ± 0.88	33.45 ± 1.86	28.91 ± 5.38	33.30 ± 1.79	38.17 ± 0.94	25.85 ± 0.58	32.23 ± 0.94	26.75 ± 1.38	27.46 ± 1.50	34.43 ± 1.37
Length of leaf upper stomata	50.24 ± 1.16	52.39 ± 2.31	50.48 ± 3.70	52.19 ± 2.22	51.08 ± 1.37	45.44 ± 1.36	57.42 ± 1.21	47.82 ± 1.18	40.20 ± 1.79	45.48 ± 1.13

Values in the table are mean and standard error

Table 4 Eigenvalues and percentage of variance for investigated parameters of PCA analysis

	Eigenvalue	Variability (%)	Cumulative (%)
PC1	2.435	18.733	18.733
PC2	1.749	13.451	32.184
PC3	1.369	10.530	42.714
PC4	1.223	9.408	52.123
PC5	1.006	7.739	59.862

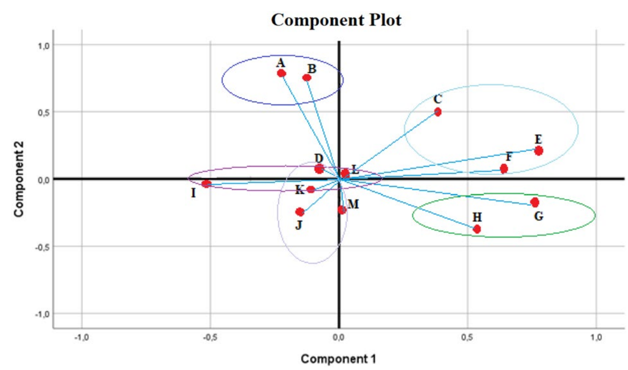

Fig. 4 PCA score plot bases on the anatomical features of *Androsace* species. **A** upper stoma width, **B** upper stoma size, **C** lower stoma size, **D** width of lower stoma, **E** upper epidermis size, **F** upper epidermis width, **G** lower epidermis height, **H** lower epidermis width, **I** scape cortex general, **J** scape cortex parenchyma diameter, **K** scape self-parenchyma, **L** caliper cylinder overall diameter, **M** spongy parenchyma

armenia var. *macrantha,* and *A. villosa* taxa show a distribution under similar ecological conditions (Table 1). Among these taxa, goblet and mushroom-shaped glandular hairs were found only in *A. villosa*. There are stomata on the taxa's lower and upper surfaces, and the general stomata type is anomocytic. The stomata sizes of the taxa are different from each other. The features of stomata are generally affected by environmental factors. However, some researchers have reported that stomatal sizes can be a distinguishing feature, especially in taxa with the same ecological and climatic characteristics (Ulcay 2022a, 2023). In addition, stomata sizes were used to diagnose some taxa, although they could not be in the same ecological conditions. For example, stomatal size was used as a distinguishing feature in the differentiation of *Anagallis* taxa (Davis 1978; Ulcay 2022b). In taxa leaf anatomies, the presence of goblet and mushroom-shaped glandular hairs, five-branched non-glandular hairs, and crystals in the mesophyll can be used as distinguishing features. Simpson (2019) states that trichomes may be valuable in plant systematics and may have important anatomical characters.

As a result of the statistical analysis, the stomatal width on the upper surface of the leaves and the stoma size on

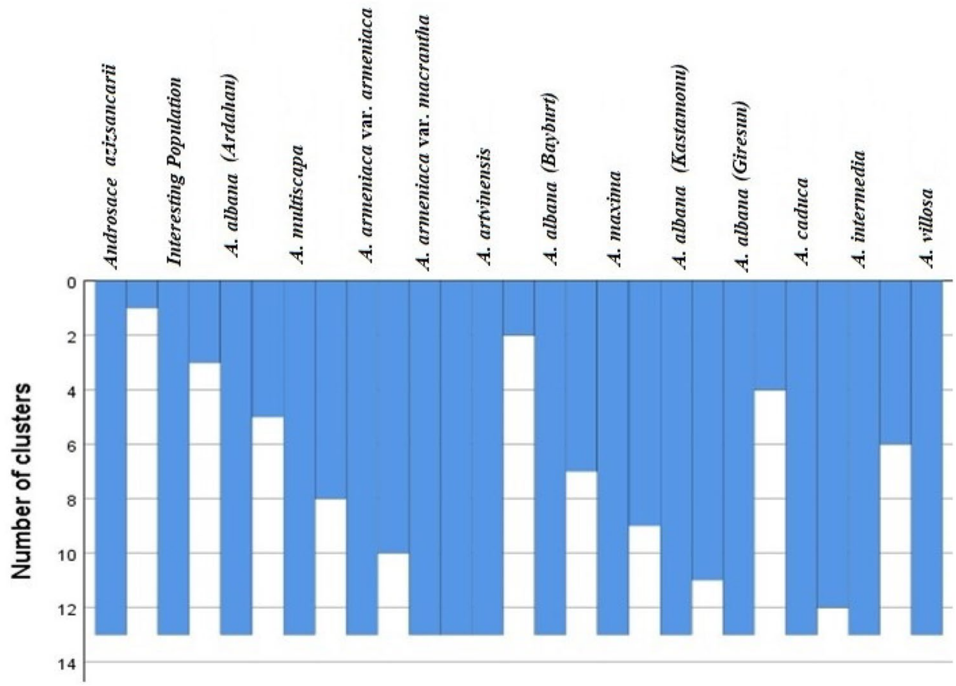

Fig. 5 Number of clusters graphic of *Androsace* spp.

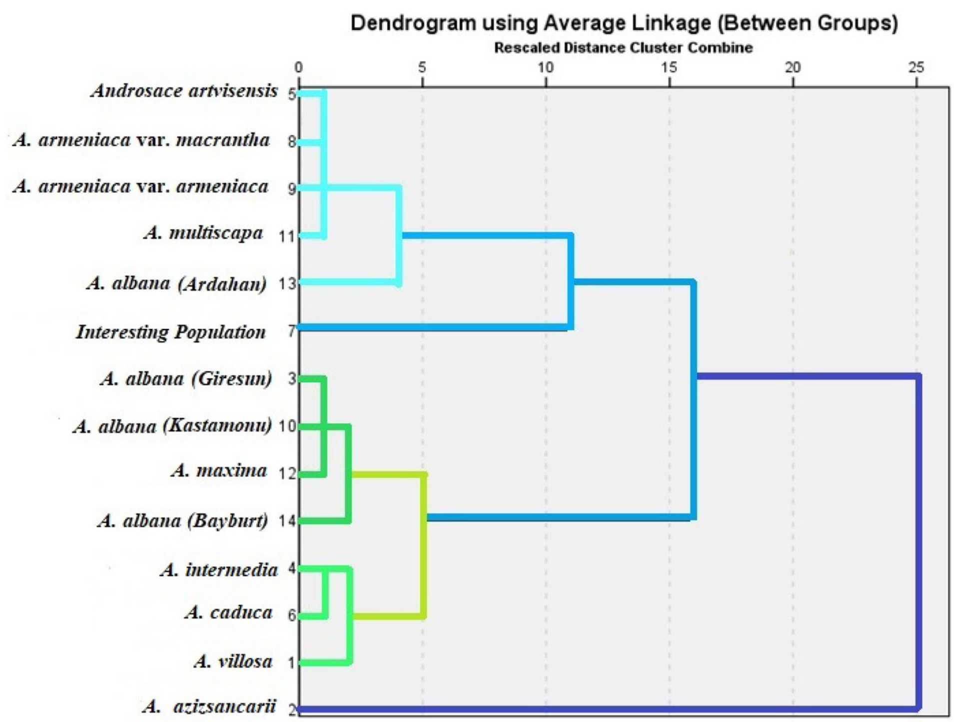

Fig. 6 HCA dendrogram according to the anatomical characters of *Androsace* spp.

the lower surface differ from each other in *Andrasace*. It was determined to be a new species by *A. azizsancarii*. Again, HCA and PCA analysis results show this species can be considered new.

5 Conclusions

In this study, *Androsace* species were evaluated regarding their anatomical characters and analyzed with PCA and HCA analyses. The results show that there are differences as well as similarities between *Androsace* species. It is noteworthy that *A. azizsancarii* forms a separate main group with HCA. The HCA results show that the two main groups are further subdivided within themselves. While it is seen that the first main group is divided into more subgroups, it is noteworthy that only *A. azizsancarii* is in the second main group. This situation shows that *A. azizsancarii* is differentiated from other species regarding anatomical features. In addition, the fact that two of the five basic components are represented by a single variable according to the PCA results suggests that the anatomical characters examined are determinative for *Androsace* taxa. In PCA, eigenvalues of PC1 (2.435), PC2 (1.749), PC3 (1.369), PC4 (1.223), and PC5 (1.006) were greater than 1.0. Again, the analysis results suggest that anatomical characteristics such as lower epidermis width, upper epidermis width, upper surface stomatal width, dimensions of stomata on the lower surface, and scape cortex width can be used as distinguishing features among taxa. The cross-sectional shape of the leaf, the presence of goblet and mushroom-shaped glandular hairs on the leaves, the presence of five branched non-glandular hairs, the presence and types of crystals in the mesophyll, the presence of aerenchyma in the scape, the scape indumentum density, the presence of four branched non-glandular hairs, and the arrangement of the bundles in the scape are among the important qualitative features.

Acknowledgements No financial support was received for this study.

Declarations

Conflict of interest There is no conflict of interest between the authors.

Research involving human and animal participants Humans or animals were not used as material in this study.

References

Baasanmunkh S, Kovtonyuk NK, Oyuntsetseg B, Tsegmed Z, Han IV, Choi HJ (2020) Diversity and distribution of the genus *Primula* L. (Primulaceae) in Mongolia. J Asia-Pac Biodivers 13(4):687–700

Bai YH, Zhang SY, Guo Y, Tang Z (2020) Conservation status of Primulaceae, a plant family with high endemism, in China. Biol Conserv 248:108675

Bonelli M, Eustacchio E, Minici A, Dinatale E, Melotto A, Mangili F, Caccianiga M (2021) Unravelling biology and ecology of the endangered endemic alpine plant *Androsace brevis* (Hegetschw) Cesati (Primulaceae.) by a multidisciplinary approach. Boll Soc Ticin Scinat 109:230–230

Boonprajan P, Leeratiwong C, Sirichamorn Y (2023) Morphological, anatomical, phytochemical, and phylogenetic evidences reveal into a new Derris species (Fabaceae) with rare flowers and reddish midribs, from Peninsular Thailand. ARPHA Preprints 4:e105209

Birjees M, Ahmad M, Zafar M, Khan AS, Ullah I (2022) Palyno-anatomical characters and their systematic significance in the family Apiaceae from Chitral, eastern Hindu Kush. Pak Microsc Res Tech 85(3):980–995

Boucher FC, Thuiller W, Roquet C, Douzet R, Aubert S, Alvarez N, Lavergne S (2012) Reconstructing the origins of high-alpine niches and cushion life form in the genus *Androsace SL* (Primulaceae). Evol 66(4):1255–1268

Boucher FC, Zimmermann NE, Conti E (2016) Allopatric speciation with little niche divergence is common among Alpine Primulaceae. J Biogeogr 43(3):591–602

Caferri R, Bassi R (2022) Plants and water in a changing world: a physiological and ecological perspective. Rend Lincei Sci Fis Nat 33(3):479–487

Davis PH (1978) Flora of Turkey and the East Aegean Islands 6:139–141

Davis PH (1980) Flora of Turkey and the East Aegean Islands. Vols. 1–9. Edinburgh Univ. Press, Edinburgh

Davis PH, Mill RR, Tan K (1988) Flora of Turkey and the East Aegean Islands, vol 10. Edinburgh Univ. Press, Edinburgh

Dentant C, Lavergne S, Malécot V (2018) Taxonomic revision of West-Alpine cushion plant species belonging to *Androsace* subsect. Aretia Bot Lett 165(3–4):337–351

Eustacchio E, Bonelli M, Beretta M, Monti I, Gobbi M, Casartelli M, Caccianiga M (2023) Pollen and floral morphology of *Androsace brevis* (Hegetschw.) Ces. (Primulaceae), a vulnerable narrow endemic plant of the Southern European Alps. Flora 301:152256

Glos RA, Salzman S, Calonje M, Vovides AP, Coiro M, Gandolfo MA, Specht CD (2022) Leaflet anatomical diversity in Zamia (Cycadales: Zamiaceae) shows little correlation with phylogeny and climate. Bot Rev 88(4):437–452

Gissi DS, Seixas DP, Fortuna-Perez AP, Torke BM, Simon MF, Souza G, Rodrigues TM (2022) Leaf and stem anatomy of the *Stylosanthes guianensis* complex (Aubl.) Sw. (Leguminosae, Papilionoideae, Dalbergieae) and its systematic significance. Flora 287:151992

Grabherr G, Gottfried M, Pauli H (2010) Climate change impacts in alpine environments. Geogr Compass 4(8):1133–1153

Hameed A, Zafar M, Ullah R, Shahat AA, Ahmad M, Cheema SI, Majeed S (2020) Systematic significance of pollen morphology and foliar epidermal anatomy of medicinal plants using SEM and LM techniques. Microsc Res Tech 83(8):1007–1022

Jacquemoud F, Jordan D (2020) Androsace albimontana (Primulaceae): a new species from the Alps (France, Switzerland, Italy) to be distinguished from *A. pubescens*. Candollea 75(1):149–155. https://doi.org/10.15553/c2020v751a14

Kaiser HF (1960) The application of electronic computers to factor analysis. Educ Psychol Meas 20(1):141–151

Lee I, Yang J (2009) Common clustering algorithms. In: Brown SD, Tauler R, Walczak B (eds) Comprehensive chemometrics. Elsevier: Oxford, UK, pp 577–618

Mabberley DJ (2008) Mabberley's plant book: a portable diction-ary of plants, their classifications and uses. Cambridge Univ. Press, Cambridge

Marzinek J, Nakajima JN, Marques D, De-Paula OC (2022) Heterocarpy in dipterocypselinae (Asteraceae): morphology, anatomy and systematic significance. S Afr J Bot 147:263–274
Ozcan M, Akinci N (2019) Micromorpho-anatomical fruit characteristics and pappus features of representative Cardueae (Asteraceae) taxa: their systematic significance. Flora 256:16–35
Raza J, Ahmad M, Zafar M, Yaseen G, Sultana S, Majeed S (2022) Systematic significance of seed morphology and foliar anatomy among *Acanthaceous* taxa. Biologia 77(11):3125–3142
Richards J (2014) *Primula*. Batsford books. England, London
Roquet C, Boucher FC, Thuiller W, Lavergne S (2013) Replicated radiations of the alpine genus Androsace (Primulaceae) driven by range expansion and convergent key innovations. J Biogeogr 40(10):1874–1886
Schneeweiss GM, Schönswetter P, Kelso S, Niklfeld H (2004) Complex biogeographic patterns in *Androsace* (Primulaceae) and related genera: evidence from phylogenetic analyses of nuclear internal transcribed spacer and plastid trnL-F sequences. Syst Biol 53(6):856–876
Schönswetter P, Tribsch A, Niklfeld H (2003a) Phylogeography of the high alpine cushion plant *Androsace alpina* (Primulaceae) in the European Alps. Plant Biol 5(06):623–630
Schönswetter P, Tribsch A, Schneeweıss GM, Niklfeld H (2003b) Disjunctions in relict alpine plants: phylogeography of *Androsace brevis* and *A. wulfeniana* (Primulaceae). Bot J Linn Soc 141(4):437–446
Sefali A (2021) *Androsace azizsancarii* sp. nov. (Primulaceae): a new species from northeastern Anatolia, Turkey. Nord J Bot 39(7):1–8
Sefali A, Yapar Y (2022) *Androsace artvinensis* sp. nov. (Primulaceae): a new species from northeastern Anatolia, Turkey. Phytotaxa 552(3):171–181
Smith G, Lowe D (1997) The genus *Androsace*. Alpine Garden Society. Avon Bank, The UK
Talebi SM, Rashnou-Taei M, Sheidai M, Noormohammadi Z (2015) Use of anatomical characteristics for taxonomical study of some Iranian Linum taxa. Environ Exp Bot 13(3):123–131
Tütüncü M (2020) In vitro culture of primula: a review. Nt J Agric Nat Resour 13(2):118–125
Simpson MG (2019) Plant systematics. Academic press
Xu Y, Hu CM, Hao G (2016) Pollen morphology of *Androsace* (Primulaceae) and its systematic implications. J Syst Evol 54(1):48–64. https://doi.org/10.1111/jse.12149
Xu Y, Hu CM, Hao G (2020) Proposal to conserve the name *Androsace bulleyana* against *A. coccinea* (Primulaceae). Taxon 69(4):830–831
Ulcay S (2022a) Anatomy, palynology, seed and leaf micromorphology of Turkish endemic *Allium brevicaule* Boiss. & Balansa and *Allium scorodoprasum* ssp. *rotundum* (L.) stearn. Acta Biol Crac Ser Bot 64(1):27–38
Ulcay S (2022b) A research on the histo-anatomical characteristics of some species of *Anagallis* (Primulacaeae) distributed in Turkey. In: Demirçali A (ed) Current Debates in Science and Mathematics Duvar Publishing, pp 187–197
Ulcay S (2023) Comparative anatomy of some *Sedum* species (Crassulaceae) in Turkey and distinguishing characteristics of these species. J Anim Plant Sci 33(1):95–102
Vardar Y (1987) Preparation technique in botany. Ege University Faculty of Science Publications.
Villalva AS, Gnaedinger S, Zavattieri AM (2023) Systematic and organ relationships of Neocalamites (Halle) Vladimirovicz, and Nododendron (Artabe and Zamuner) emend from the Triassic of Patagonia. Palaeobiogeographic, palaeoenvironments and palaeoecology considerations. Rev Palaeobot Palynol 316:104939