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Original Research

Comparative Petiole Anatomy in Members of Polygonaceae as Medicinal Plants from District Muzaffarabad, Kashmir Himalayas

Syed Waseem Gillani[1*], Mushtaq Ahmad[1], Muhammad Zafar[1], Muhammad Manzoor[1***], Javeed Hussain[2], Abd El-Zaher M.A. Mustafa[3], Mohamed S Elshikh[3], Mohamed Ragab AbdelGawwad[4], Shazia Sultana[1], Abdul Basit Mehmood[5], Bushra Ali[1]**

[1]Department of Plant Sciences, Quaid-i-Azam University, Islamabad 45320, Pakistan
[2]Department of Botany, University of Kotli Azad Jammu and Kashmir
[3]Department of Botany and Microbiology, College of Science, King Saud University, P.O. 2455, Riyadh 11451, Saudi Arabia
[4]Genetics and Bioengineering, Faculty of Engineering and Natural Sciences, International University of Sarajevo, 71210 Sarajevo, Bosnia and Herzegovina
[5]Department of Botany University of Azad Jammu & Kashmir

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Abstract

The petiole anatomy of some medicinally important species was investigated. The comprehensive explanation of Polygonaceae species is connected to their systematic significance. One of the reliable characteristics that is used for precise species identification is microanatomical attributes. The main objective of the current study was to study the taxonomic significance of the petiole anatomy and ethnobotanical uses of eight Polygonaceae taxa from the Kashmir Himalayan Region. A microtome was used to assess the microanatomical quantitative and qualitative features of the petiole. The qualitative characteristics of the petiole were observed, such as collenchyma cell shape, parenchyma cell shape, epidermal cell shape, and vascular bundles (VBs), with a lot of variations. The number of VBs and length of the petiole were also examined as quantitative characteristics. The largest petiole length was observed in *Antigonon leptopus* (430 µm), and the shortest was in *Rumex hastatus* (130 µm). The highest number of VBs were observed in *Antigonon leptopus* (10), and the lowest in *Rumex chalepensis* (2). The largest parenchyma length was observed in *Rumex chalepensis* (23 µm), and the shortest was in *Rumex hastatus* (4.3 µm). The largest collenchyma width (10.6 µm) and length (14.6 µm) were observed in *Rumex crispus*, and the minimum was observed in *Persicaria berbata*. Studied taxa also have high local ethnobotanical uses. Both questionnaires and group discussions were used to

*e-mail: sgillani@bs.qau.edu.pk
**e-mail: mushtaq@qau.edu.pk
***e-mail: mmanzoor@bs.qau.edu.pk

collect the ethnobotanical data. The collected data was analyzed using a variety of quantitative metrics, such as use value (UV) and relative frequency citation (RFC). The majority of medicinal plants utilized by local communities were herbs (87.5%), with leaves (53.85%) being the most commonly used plant parts. Among the investigated taxa, *Oxyria digyna* had the highest use value (1.08), and it also had the highest relative frequency citation (0.88). This study describes the anatomical and ethnobotanical exploration of the Polygonaceae and can be further elaborated for the correct identification using phylogenetic-based micromorphological taxonomic approaches.

Keywords: epidermal cells, microanatomy, systematics relevance, Medicinal Quantitative metrics

Introduction

The family Polygonaceae is one of the most eurypalynous among the dicotyledons, consisting of a varied range of mostly herbaceous plants [1]. Eriogonoideae, Polygonoideae, and Symmerioideae are the three subfamilies that currently make up Polygonaceae [2, 3]. According to phylogenetic analyses conducted in the past [4, 5], Polygonoideae is the largest subfamily, with 21 genera and nearly 900 species. The Polygonoideae family, which shares structural similarities with ocrea and quincuncial aestivation, is an important part of the flora of various countries [6]. Though most species are located in the northern temperate zone, members of this group are also found all over the world [6-8]. Overall, Polygonaceae species are found in nearly every type of ecosystem, including mountains, wetlands, deserts, and dunes. Some species of *Persicaria* (L.) Mill. have adapted to aquatic environments, and other plants in the family, like *Rheum* L., have been found to grow in these environments [1, 9-11]. Notably, a number of species (such as *Atraphaxis* L., *Calligonum* L., and *Bistorta* L.) with strong ecological adaptations flourish in areas with limited plant diversity.

Reproductive traits that were not always readily accessible have been used since ancient times to identify species and their relationships. Alternatively, plant anatomy can be employed as a huge information-generating tool for intra- or interspecific taxonomic differentiation, circumventing the prior issue. Taxonomists examine anatomy to help identify, locate, and classify plant taxa [12-20]. It is one of the most crucial techniques that modern taxonomists employ to distinguish and identify closely related taxa [21]. Therefore, identification of vegetative organs at the species level is important, as it will help determine adulteration. Thus, anatomical research on petioles could potentially be useful from that perspective. As a result, anatomical studies of the petiole are one of the investigative tools used to identify taxa at the species level [18, 20]. The petiole structure has proven to be quite important in plant classification and categorization [20]. Taxonomists used the anatomical features of the petiole to classify plants and assign taxa to their correct taxonomic ranks [22]. They can help identify how the environment influences the structural properties of petioles in specific species by comparing them to those found in other regions. Plant scientists use several traits related to taxonomy to help in the methodical identification, grouping, and classification of plants [23, 24]. Epidermal cells, parenchyma, sclerenchyma, vascular bundles, and sizes were among the many features they investigated. Connecting the lamina to the stem, the petiole is a portion of the leaf that has anatomical characteristics that are important for survival [25]. It has been demonstrated that the microstructure of the vascular bundles in petiole histology is helpful for identifying species [26]. In a similar manner, they were examined to identify distinguishing characteristics of the species [17, 27]. Getting insight into the differences in anatomical features between related species can help us understand the evolutionary aspects of plants [28]. In order to examine problematic taxa in the genus, the anatomical data may be utilized as diagnostic features for specific species [18, 29, 30].

The Polygonaceae family was chosen for the Muzaffarabad district due to its extensive range of applications, both traditional and medical. The members of the family Polygonaceae are medicinally very important, mostly used as a wild vegetable and used to treat different diseases among the indigenous communities of the Kashmir Himalayas [31-34]. The rhizome of *Rumex* species contains essential oils, tannins, naphthalene derivatives, flavonoids, steroidal chemicals, leucoanthocyanidol, fixed oils, polysaccharides, and essential oils [35-41]. It has been discovered that certain species work well against bacteria and fungi. Furthermore, it was discovered that a few species had anti-inflammatory properties. Based on the substance groups they contain, antitumoral, antihistaminic, cardiovascular, hematological, and laxative properties were reported [41-43]. Additionally, it was mentioned that a few species within the family have antibacterial and antioxidant properties [44, 45]. Family members were employed in traditional medicine. *Rumex crispus* L. was effective in treating rheumatism, piles, diarrhea, oedema, lung bleeding, and excretion, as well as cutaneous, respiratory, and digestive disorders [46]. Important species of *Rumex* were also ethnobotanically collected. Typically, family species leaves were eaten raw in salads, but their stems, branches, and leaves were also cooked as vegetables [31, 34]. Traditional medical practitioners usually classify herbs into two groups: those in their prepared form (after post-treatment activities like steaming and fermenting) and fresh or dried herbs that can be used directly. Since ancient times, medicinal plants have been recognized and utilized in traditional medicine. Humans have relied on traditional medicines, more than 90% of which are made from plants, to treat diseases throughout the history of human civilization [47-49]. Though certain plants are also employed directly as medications, most modern herbal remedies are based on their extracts [50, 51].

Although Polygonaceae has been well-known for its lengthy history of usage in folk medicine to treat a variety of illnesses, there is a conspicuous lack of anatomical study on the petiole within the Polygonaceae family. The petiole anatomy is important in medicinal plants because it facilitates nutrition delivery and provides mechanical support. Petioles in the Polygonaceae family, which have some prominent medicinal plants, frequently have specialized features like vascular bundles and sclerenchyma cells. Understanding petiole anatomy in these species helps to detect medicinal ingredient distribution and contributes to taxonomic classification. Furthermore, changes in petiole structure can shed light on evolutionary adaptations and potential therapeutic characteristics within the Polygonaceae family. The objective of this research is to investigate the potential medical uses of species while simultaneously documenting their anatomical characteristics using light microscopy in a number of Polygonaceae species. The diversity in microanatomical characteristics was found to be important for classification within the genus, and the investigation was to correlate these features, especially those of the tissues of the petioles, in order to distinguish between various Polygonaceae taxa.

Materials and Methods

Study Area

The current study has been conducted in Muzaffarabad district, Azad Jammu and Kashmir (Fig. 1). Azad Jammu and Kashmir (AJK) is described by the geographic coordinates 33°54′-34°44 North latitude and 73°31-74°50 East longitude [31, 52, 53]. The area is dominated by steep mountains, deep ravines, and rocky landscapes with undulating topography. It is bounded to the south by the Punjab Province's Gujrat district, to the east by the occupied Jammu and Kashmir Region, and to the west by the districts of Kahota, Murree Region, and KPK [31]. The climate in the area ranges from subtropical to moderately humid. During the winter months, high temperatures typically range between 25 and 34 degrees Celsius, while low temperatures typically range between 4 and 10 degrees Celsius [54].

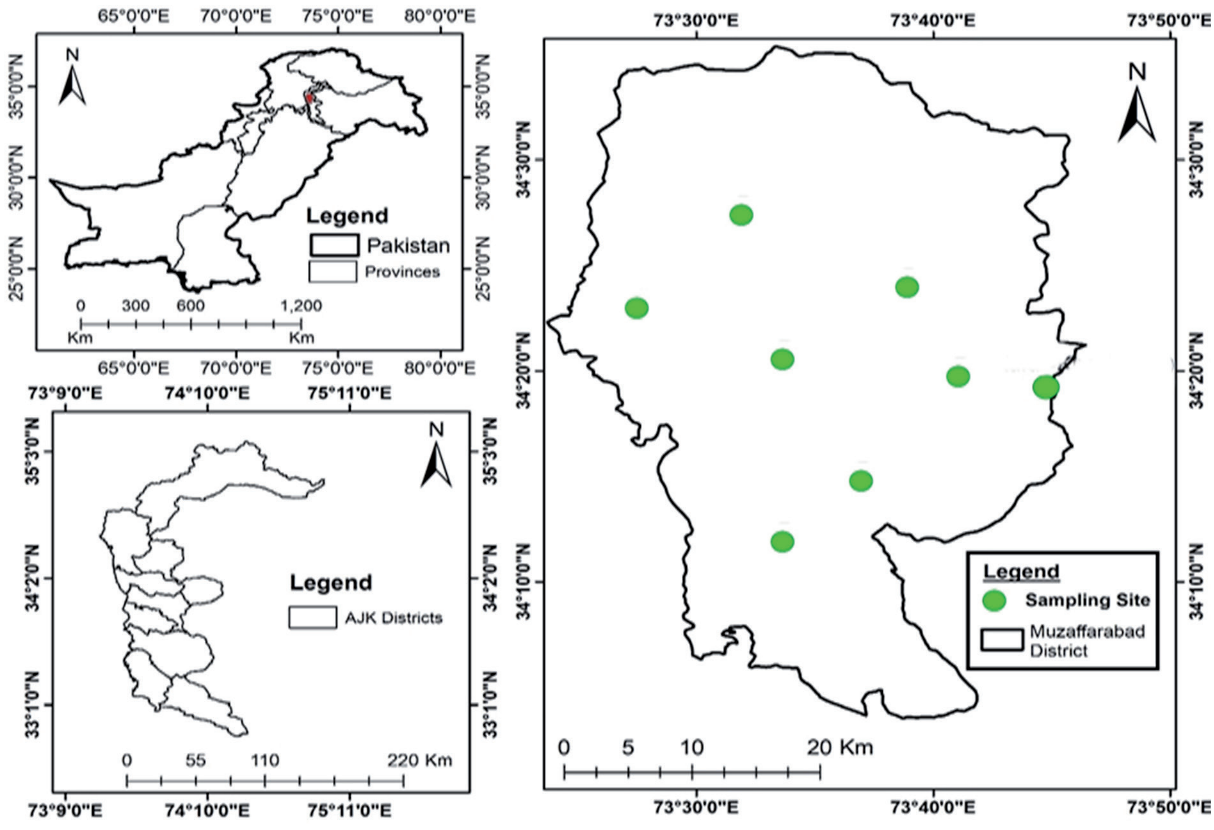

Fig. 1. Map of the study area.

ACCEPTED MANUSCRIPT

Plant Collection

A number of field trips were conducted from 2022-2023 in order to get plant specimens and traditional ethnomedical knowledge. Throughout the collection procedure, every plant was given a proper label and recognized using its local name. Plant sample pressing, drying, and mounting on herbarium sheets were done with precision. Using the "Flora of Pakistan" as a guide and following the instructions provided by Stewart [55], in addition to using a number of internet sites, made it easier to identify these specimens. Every plant name was checked by looking up the information in the World Online Flora.

Petiole Section Cutting

Sections from the center of the stalk to the base of the petiole were selected for the cross-sectional study. Two four-hour treatments with a 10% saline formal solution were applied to the petiole samples in order to prepare them for fixation. Dehydration was carried out for an hour at various concentrations of methanol (70%, 80%, 90%, and two rounds of 100%). After that, alcoholization with xylol was performed for one hour with two replacements, followed by wax impregnation with two replacements at temperatures ranging from 58-62ºC. Petiole samples were embedded or blocked by slicing them into 3-5 µm thick sections (microtomies). The slides were then melted at 62ºC according to the proven method [56].

Staining

Dewaxing was done with xylene for five minutes, with two changes. Following dewaxing, methanol was used for rehydration, with progressive dilutions of 100%, 90%, and 70% for one minute. After that, tap water was used for one minute. The slide was then stained with basic hematoxylin for 5 minutes before being washed with tap water for another minute. The slide was then soaked in 1% acid alcohol and washed with tap water for a minute. A 1% eosin solution was applied for 30-60 seconds. The part was then rinsed again with tap water for a minute. Following that, dehydration was performed with methanol concentrations of 70%, 80%, 90%, and 100% for 30 seconds each. The prepared slides were cleared with xylol for 1 minute for two changes before being mounted with dibutylphthalate polystyrene xylene (DPX). The slides were then labeled and observed under a light microscope by following the procedure [18].

Ethnobotanical Quantitative Data Analysis

The first step was compiling a list of Poygonaceae taxa for further evaluation. This included information on the different plants used in phytoremediation as well as any potential therapeutic benefits. Then, specific indices were calculated:

Use Value (UV)

Use value (UV) is the local identification and citation of plants to assess their value within a certain area. The use value is calculated using a specific methodology [33, 57]. It was calculated by the following method:

$$UV = \sum Ui/N$$

Relative Frequency Citation (RFC)

It highlights the significance of each species and is determined using the method developed by Tardio and Pardo-de-Santayana [58], which is based on the frequency of citation (FC) – the number of informants who mention the use of a certain species. The FC value is calculated by dividing the total number of survey participants (N) by the disease categories. Its value varies from "0" to "1".

$$RFC = FC/N$$

Results

The quantitative and qualitative characteristics of petiole anatomy, along with the local ethnobotanical uses of the studied taxa, were investigated. More details on each taxa are given below in detail.

Antigonon Leptopus

The length of the adaxial surface was noted as (4-6) 5±1.0 (µm), and the width was (4-5) 4.3±0.5 (µm). The length of the abaxial surface was noted as (5-6) 5.3±0.5 (µm), and the width was (4-6) 4.6±1.1 (µm). The length and width of the parenchyma ranged from 13-27 µm and 13-25 µm, respectively. The length of the petiole was noted as 450 µm, and the number of vascular bundles was 10 (Table 2). The shape of the petiole was observed as circular. The abaxial and adaxial surfaces were thickened and irregular. The collenchyma has a polygonal shape, whereas the parenchyma is spherical in shape. The shape of the collenchyma was noted as polygonal, and the parenchyma was spherical. Vascular bundles were observed to be elliptical (Fig. 2). *Antigonon leptopus* is a climber and does not have significant medicinal uses. This species is mostly used as an ornamental plant. This species is used to treat skin ailments; a fresh leaf paste is applied externally (Table 1).

Oxyria Digyna

The adaxial surface length (10-11) was 10.3±0.5 (µm) and the breadth (5-8) was 6.6±1.5 (µm), respectively. The length of the abaxial surface was (10-15) 11.6±2.8 (µm) and the width was (6-10) 8.3±2.0 (µm).

Table 1. Checklist of the Polygonaceae taxa with local medicinal uses.

Taxa	Habit	Life Form	Leaf Form	Part Used	Use Category	Traditional Medicinal Uses	Marketing	Use Value	RFC
Antigonon leptopus Hook. & Arn.	Cl	Pha	Mic	Leaves	Medicinal, Ornamental	For skin problems, a fresh leaf paste is administered externally	-	0.14	0.33
Oxyria digyna (L.) Hill	H	Hem	Mic	Leaves, Aerial parts	Medicinal, Vegetable, Raw	Leaves are used as a vegetable for stomach problems	-	1.08	0.88
Persicaria barbata (L.) H. Hara	H	Ther	Nan	Leaves	Medicinal	Used to cure ulcers	-	0.19	0.38
Persicaria nepalensis (Meisn.) Miyabe	H	Ther	Mic	-	-	-	-	0.00	0
Rumex chalepensis Mill	H	Hem	Mes	Leaves	Medicinal, Vegetable	Skin diseases, coughs, and root decoctions are used to relieve pulmonary disorders. Used as a vegetable in constipation	-	0.33	0.68
Rumex crispus L	H	Hem	Mic	Root, Leaves, stem	Medicinal	Gastrointestinal disorders, burns, skin conditions, rashes, sores, and jaundice	-	0.56	0.86
Rumex dentatus L	H	Hem	Mes	Root, Leaves	Medicinal, Vegetable	Diarrhoea, constipation, bacterial and fungal infection, stomach problems, and leaves are consumed as vegetables in mountainous areas of Kashmir	-	0.73	0.83
Rumex hastatus D. Don	S	Np	Mic	Aerial parts, Roots	Medicinal, Vegetable, Raw	Leaves are used as a vegetable; the juice of the plant is astringent and used in the treatment of amoebic dysentery, cough, headache, and skin problems. It is also used as a vegetable	-	0.90	0.81

Abbreviations: Cl = Climber, H = Herb, S = Shrub, Ther = Therophytes, Hem = Hemicryptophytes, Geo = Geophytes, Mac = Macrophylls, Mic = Microphylls and Nan = Nanophylls.

The length and width of the parenchyma ranged from 7-10 µm and 5-9 µm, respectively. Collenchyma length ranged from 9-15 µm and width ranged from 3-7 µm. The number of vascular bundles was four, and the length of the petiole was 220 µm. The shape of the petiole was examined as sulcate. The abaxial and adaxial surfaces were polygonal. The shape of the parenchyma was seen to range from pentagonal to polygonal. The shape of the collenchyma was lamellar, whereas the shape of the vascular bundle was observed to be rounded (Table 3). *Oxyria digyna* is an important herb, and locally, it is very important for different purposes. This species is used as a vegetable, may be individually or in a mix of vegetables, and is also used as an alternative to water in alpine mountainous areas to quench thirst where water is not available, mostly during fields and hiking. According to the elderly inhabitants of the study area, it is a very effective vegetable to cure different stomach problems.

Persicaria Barbata

The abaxial epidermal cells have (6-7) 6.3±0.5 (µm) length and (4-5) 4.6±0.5 (µm). Similarly, adaxial surface length was noted (5-6) 5.3±0.5 (µm) and width was (4-5) 4.3±0.5 (µm). The length of the collenchyma was observed in the range (4-6 µm) and width (2-5) µm, respectively. The length and width of parenchyma were recorded at 9-11 µm and 8-10 µm, respectively. The length of the petiole was noted at 220 µm, and the number of vascular bundles was two to three. Qualitatively, the shape of the petiole was sulcate. The shape of the adaxial surface was smooth, and the abaxial surface was convex. The shape of the collenchyma was annular, whereas the parenchyma was noted as polygonal. The shape of the vascular bundle was observed to be elliptical (Fig. 2). *Persicaria barbata is not frequently u*sed among the locals of the study area, but sometimes it is used to cure ulcers (Table 1).

Persicaria Nepalensis

The length and width of the abaxial surface were noted as (5-8) 6.6±1.52 (µm) and (4-7) 5.6±1.5 (µm), respectively. The length and width of the adaxial surface were recorded at (7-9) 8±1.0 (µm) and (6-7) 6.3±0.5 (µm), respectively. The length of the collenchyma ranged from 6-9 (µm) and width (5-6) (µm). Parenchyma length ranged from 8-10 (µm) and width ranged from 6-10 (µm). The length of the petiole was 240 µm, and six vascular bundles were counted (Table 2). The shape of the petiole was triangular with grooves. The shape of the vascular bundle was oval. Both the abaxial and adaxial surfaces were isodiametric. The shape of the parenchyma cell was polygonal, and the collenchyma shape was annular. *Persicaria nepalensis* was not reported to be used medicinally during encounters with locals.

Rumex Chalepensis

Abaxial epidermal cells measured (9-11) 10±1.0 (µm) in length and (5-6) 5.3±0.5 (µm) in width. The adaxial surface length was noted (11–20) 15±4.5 (µm) and width (9-10) 9.5±0.5 (µm). Collenchyma ranges 12-15 µm in length and 10-11 µm in width, while parenchyma ranges 15-35 µm in length and 9-12 µm in width,

Table 2. Quantitative characteristics of petiole anatomy of family Polygonaceae.

Species	Petiole Length (µm)	Number of VB	Abaxial Epidermis		Adaxial Epidermis		Parenchyma		Collenchyma	
			L (µm)	W (µm)	L (µm)	W (µm)	L (µm)	W (µm)	L (µm)	W (µm)
			(Min-Max) Mean±SD							
Antigonon leptopus	450	10	(5-6) 5.3±0.5	(4-6) 4.6±1.1	(4-6) 5±1.0	(4-5) 4.3±0.5	(13-27) 20±7.0	(13-25) 17.6±6.4	(7-12) 8.6±2.8	(6-11) 8±2.6
Oxyria digyna	220	4	(10-15) 11.6±2.8	(6-10) 8.3±2.0	(10-11) 10.3±0.5	(5-8) 6.6±1.5	(7-10) 8±1.7	(5-9) 7±2.0	(9-15) 11.3±3.2	(3-7) 5.3±2.0
Persicaria barbata	220	3	(6-7) 6.3±0.5	(4-5) 4.6±0.5	(5-6) 5.3±0.5	(4-5) 4.3±0.5	(9-11) 10±1.0	(8-10) 9±1.4	(4-6) 5±1	(2-5) 3.6±1.5
Persicaria nepalensis	240	6	(5-8) 6.6±1.52	(4-7) 5.6±1.5	(7-9) 8±1.0	(6-7) 6.3±0.5	(8-10) 9.3±1.1	(6-10) 8.0±2.8	(6-9) 7.6±1.5	(5-6) 5.3±0.5
Rumex chalepensis	290	2	(9-11) 10±1.0	(5-6) 5.3±0.5	(11-20) 15±4.5	(9-10) 9.5± 0.5	(15-35) 23±10.5	(9-12) 10.5±2.1	(12-15) 13.6±1.5	(11-10) 10.3±0.5
Rumex crispus	420	6	(15-17) 16±1.0	(6-9) 7.3±1.5	(12-18) 15.3±3.05	(5-10) 7.6±2.5	(6-8) 6.6±11.1	(4-12) 7±4.3	(10-18) 14.6±4.1	(8-12) 10.6±2.3
Rumex dentatus	280	4	(6-12) 9.3±3.0	(5-11) 8.3±3.0	(5-8) 6.3±1.5	(4-6) 5±1.0	(9-13) 11.3±2.0	(8-10) 9±1.0	(7-12) 9.6±2.5	(5-7) 6.3±1.1
Rumex hastatus	130	3	(12-23) 17.3±5.5	(5-9) 7.3±2.0	(14-24) 18±5.2	(3-10) 7±3.6	(3-6) 4.3±1.5	(2-4) 2.6±1.1	(6-14) 10±4.04	(3-9) 6.3±3.0

Abbreviations: L = length, W = Width, VB = Vascular bundles.

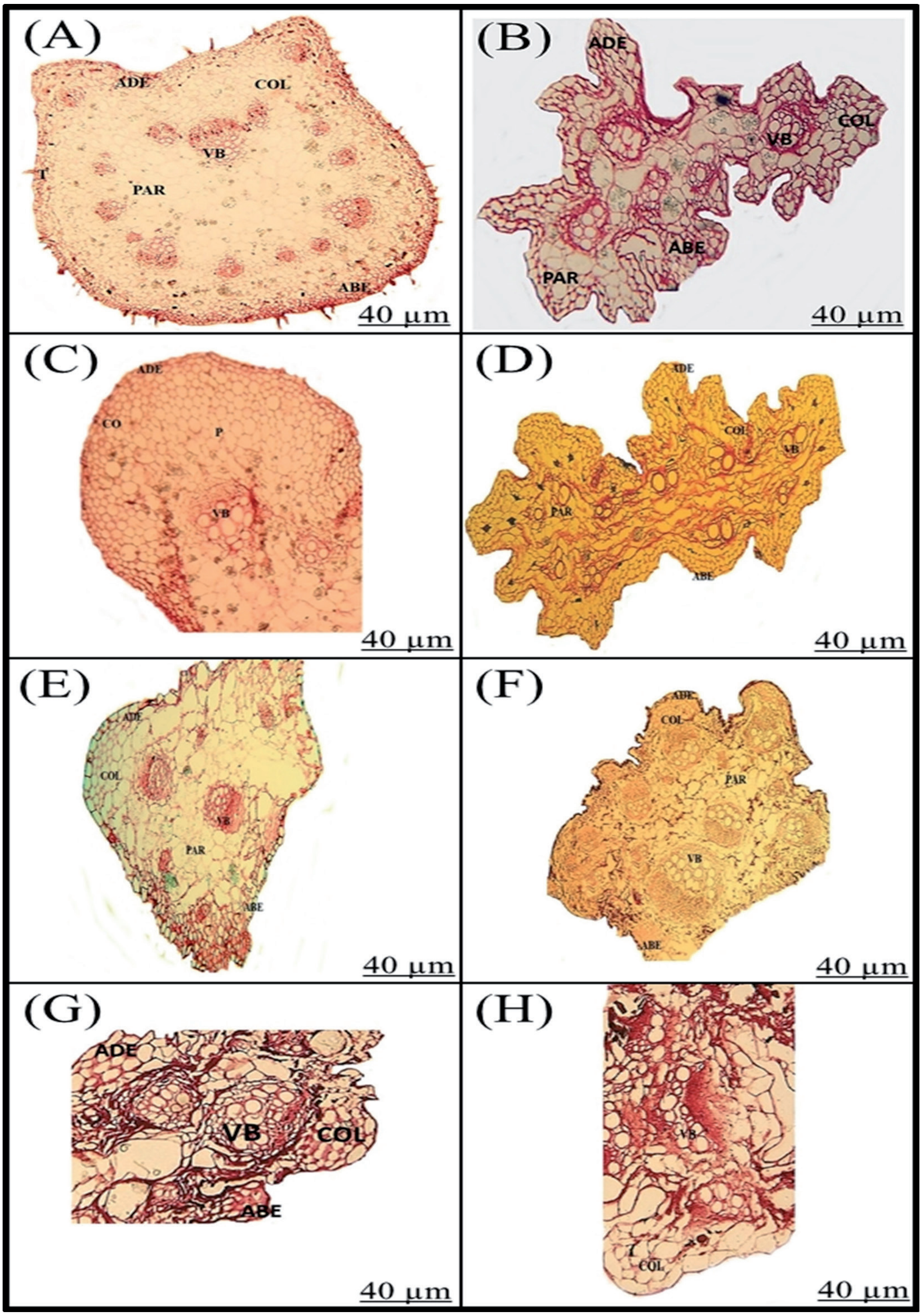

Fig. 2. Petiole anatomy of the family Polygonaceae, where A = *Antigonon leptopus*, B = *Oxyria digyna*, C = *Persicaria barbata*, D = *Persicaria nepalensis*, E = *Rumex chalepensis*, F = *Rumex crispus*, G = *Rumex dentatus*, and H = *Rumex hastatus*.

respectively. The petiole measured 290 µm in length and had six vascular bundles. The petiole had a sulcate shape. The abaxial and adaxial epidermal surfaces were isodiametric. The collenchyma cell was annular, whereas the parenchyma cell was polygonal and irregular in shape (Table 3). The vascular bundle was seen to have a sulcate shape. *Rumex chalepensis* is a valuable medicinal vegetable used to treat a variety of ailments, including skin illnesses, coughs, and pulmonary disorders. It is used as a vegetable to treat constipation.

Rumex Crispus

The abaxial surface measured (15-17) 16±1.0 (µm) in length and (6-9) 7.3±1.5 (µm) in width. The adaxial surface's length (12–18) was 15.3±3.05 (µm) and breadth (5-10) was 7.6±2.5 (µm). The collenchyma was 10-18 µm in length and 8-12 µm in breadth. The parenchyma length ranged from 6 to 8 µm. The width ranged from 4 to 12 µm. The length of the petiole was 420 µm, and the number of vascular bundles was six. The shape of the petiole was sulcate. The shape of the vascular bundle was elongated. Both the abaxial and adaxial surfaces were thickened and irregular. The shape of the parenchyma cell was irregular, and the collenchyma shape was annular. *Rumex crispus* is used to treat various disorders among the locals; these include gastrointestinal disorders, burns, skin conditions, rashes, sores, and jaundice (Table 1).

Rumex Dentatus

The length and width of the adaxial epidermal cell were (5-8) 6.3±1.5 (µm) and (4-6) 5±1.0 (µm), respectively. Abaxial surface length and width were noted at (6-12) 9.3±3.0 (µm) and (5-11) 8.3±3.0 (µm), respectively. The parenchyma cell has a length of 9-13 µm and a width of 8-10 µm. Collenchyma cells had a recorded length of (7-12 µm) and width of (5-7 µm). The length of the petiole was noted at 280 (µm). The shape of the petiole was flat. The number of vascular bundles was recorded as four, and the shape was elongated. Both abaxial and adaxial epidermal cells were polygonal in shape. The shape of the parenchyma cell was spherical, and the collenchyma was angular in shape. *Rumex dentatus* is a medicinally important herb and cures many diseases, including diarrhea, constipation, bacterial and fungal infections, and stomach problems. Leaves are consumed as vegetables in the mountainous areas of Kashmir.

Rumex Hastatus

The adaxial surface has length (14–24) 18±5.2 (µm) and width (3-10) 7±3.6 (µm). The abaxial surface has length and width (12-23) 17.3±5.5 (µm) and (5-9) 7.3±2.0 (µm), respectively. The collenchyma measured 6-14 µm in length and 3-9 µm in width, while the parenchyma measured 3-6 µm in length and 2-4 µm in width (Fig. 2). The petiole measured 130 µm in length and had three vascular bundles. The shape of the petiole was flat. The shape of the abaxial and adaxial surfaces was polygonal. The shape of the collenchyma was lamellar, and the parenchyma was irregular in shape. Vascular bundles were spherical. Leaves are used as a vegetable; the juice of the plant is astringent and used in the treatment of amoebic dysentery, cough, headache, and skin problems. It is also used as a vegetable.

Table 3. Qualitative Characteristics of petiole anatomy of Polygonaceae.

Species	Shape of Petiole	VB Shape	Shape of adaxial epidermal cell	Shape of abaxial epidermal cells	Shape of parenchyma cells	Shape of collenchyma cells
Antigonon leptopus	Circular	Elliptical	Irregular, thickened	Irregular, thickened	Spherical	Annular
Oxyria digyna	Sulcate	Rounded	Polygonal	Polygonal	Polygonal	Lamellar
Persicaria barbata	Sulcate	Elliptical	Smooth	Convex	Polygonal	Annular
Persicaria nepalensis	Triangle with grooves	Oval	Isodiametric	Isodiametric	Irregular	Annular
Rumex chalepensis	Sulcate	Rounded	Isodiametric	Isodiametric	Polygonal	Annular
Rumex crispus	Sulcate	Elongated	Irregular, thickened	Irregular, thickened	Irregular	Annular
Rumex dentatus	Flat	Elongated	Polygonal	Polygonal	Spherical	Angular
Rumex hastatus	Flat	Spherical	Polygonal	Polygonal	Irregular	Lamellar

Abbreviations: VB = Vascular bundles.

Plant Parts Used

Several plant parts were recorded for their medicinal properties in the present investigation (Fig. 4). Leaves were the most commonly used plant parts with 53.85%, followed by the roots with 23.08% (Table 1). Following that, aerial parts were documented at 15.38%, and then the stem was documented at 7.69% (Fig. 3). The aerial parts of plants, particularly the leaves, are thought to have a higher concentration of extractable phytochemicals, crude medicines, and other active molecules with potential applications in phytotherapy.

Use Categories

The ethnoflora was divided into four use categories: medicinal, vegetable, raw, and ornamental. The most widely used category was medicinal, which included 7 plant species. Following that, there were 4 species of vegetables, 2 species as raw, and 1 species as ornamental. The range of uses was particularly prominent in two plant species, *Oxyria digyna* and *Rumex hastatus*, which were associated with three distinct usage categories. In addition, three plant species displayed utilization across two use categories, while two plant species were identified as having applicability in one use category and *Persicaria nepalensis* had no specific medicinal use (Table 1).

Use Value

Use value is very crucial for the identification of the most valuable medicinal plants used in the investigated area. The use value of medicinal plants ranges from 0 to 1.08 (Table 2). The highest UV was calculated for *Oxyria digyna* (1.08), followed by *Rumex hastatus* (0.90), *Rumex dentatus* (0.73), and the lowest use value was recorded for *Persicaria nepalensis* (0.00), *Antigonon leptopus* (0.14), and *Persicaria barbata* (0.19) (Table 1).

Relative Frequency Citation

The RFC value ranges from 0 to 0.88 (Table 2). The highest RFC value was calculated for *Oxyria digyna* (0.88), followed by *Rumex crispus* (0.86) and *Rumex dentatus* (0.83), and the lowest use value was recorded for *Persicaria nepalensis* (0), *Antigonon leptopus* (0.33), and *Persicaria barbata* (0.38). Locals are familiar with a range of high-RFC plant species that are common in the area, mostly because they have been used for a long time in traditional medicine (Table 1).

Statistical Analysis

The Pearson correlation statistical technique was employed on the quantitative data of petiole anatomy, such as abaxial epidermis, adaxial epidermis, parenchyma, and collenchyma. The statistical analysis result revealed that all the variables showed a positive relationship except for the negative correlation observed between parenchyma length and width (Fig. 4).

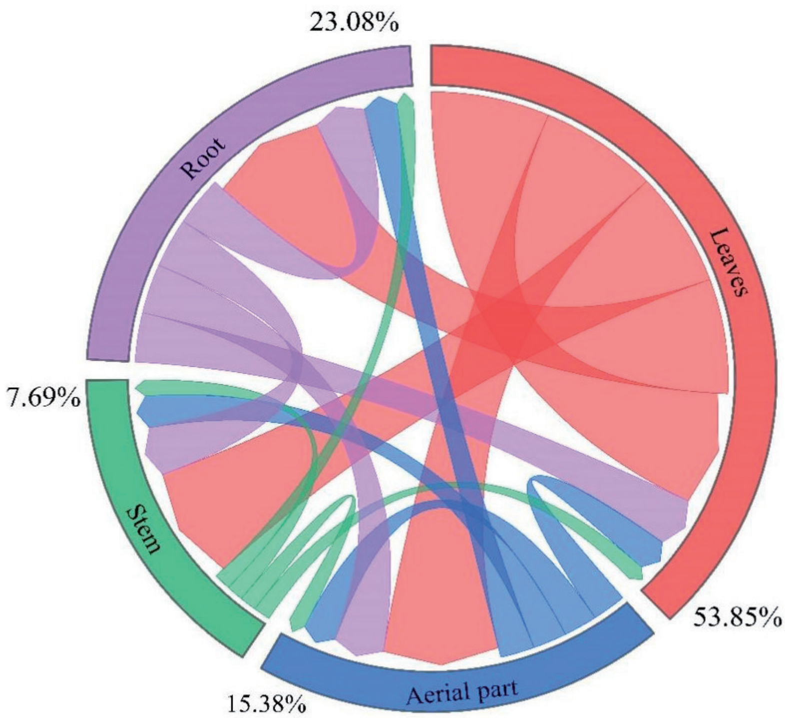

Fig. 3. Plant part used percentage among the local inhabitants.

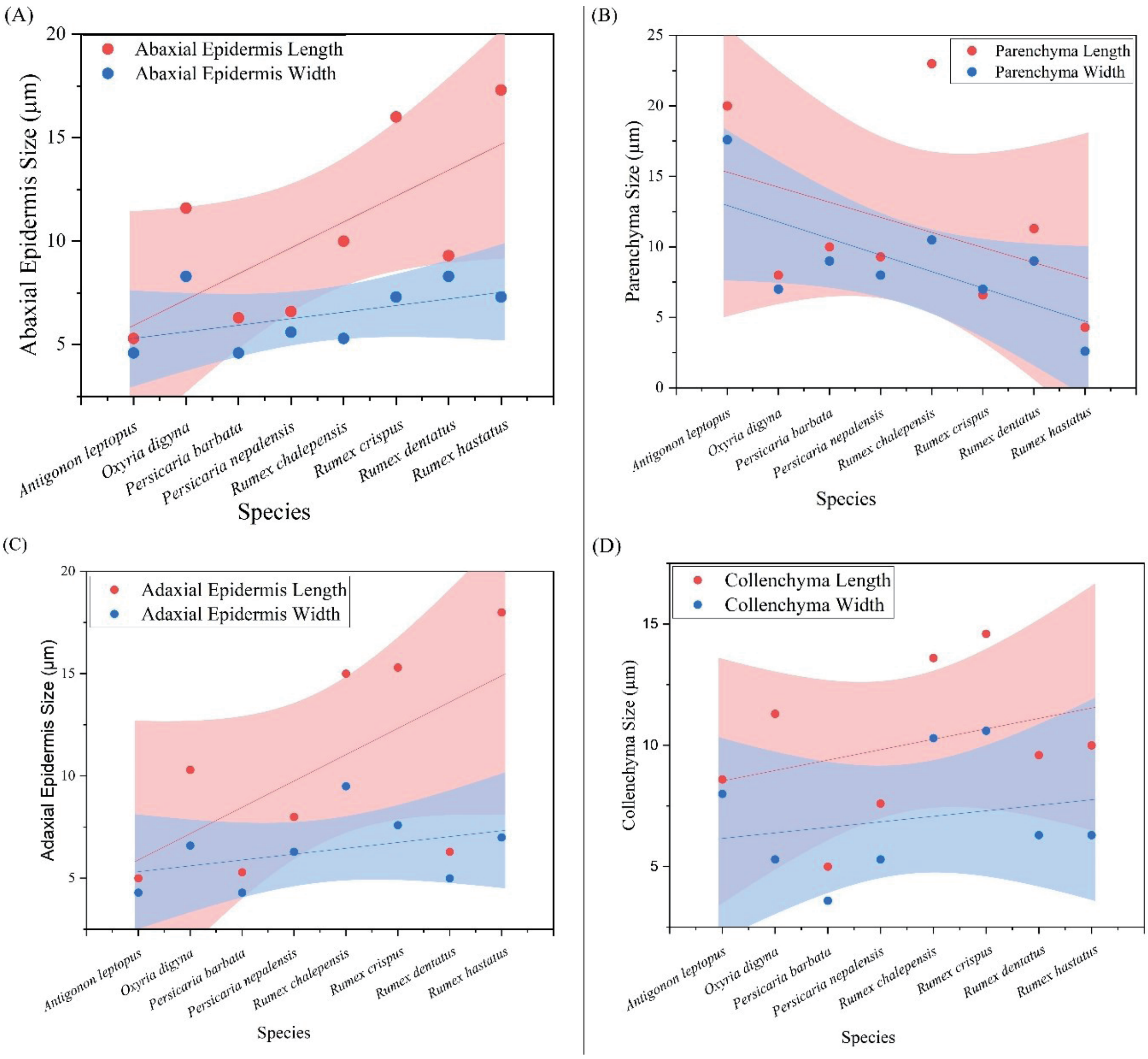

Fig. 4. Pearson correlation among the different quantitative characteristics of the Polygonaceae species.

Discussion

The current research was evaluated to determine the anatomical features of petioles and the local ethnobotanical uses of Polygonaceae taxa from the Kasmir Himalayan Region. Many researchers investigated variations in length and width of the abaxial and adaxial features, the number of vascular bundles in petiole anatomy, and many other parameters [16, 18]. These micromorphological characters were used for taxonomic and systematic studies of Polygonaceae. The petiole anatomy was investigated many years ago and is similar to our results [59-63]. The quantitative and qualitative features of the leaf and petiole anatomy of each species vary with each other, showing that polygonaceae were very distinct in Muzaffarabad. Each taxonomic group exhibited distinct anatomical traits that serve as indications for distinguishing between them, making the data documented important in differentiating the taxa under study. Anatomical features that were observed included the size and shape of petioles, the size and shape of vascular bundles, the size and shape of epidermal cells, the type and size of collenchyma, the size and shape of parenchyma, and the position of vascular bundles in relation to one another, as in earlier studies [14, 17-19, 24, 56, 62, 64, 65].

In petiole anatomy, eight species showed variations among them. In the context of phylogenetic lineages, the petiole vascular system offers an important feature to support plant tissue growth and adaptation to a broad range of ecological environments [66]. Accordingly, recent micro-systematic research indicates that Polygonaceae taxa have a vascularization system that is restricted to some specific tissue layers. The shapes of the petiole were sulcate, flat, circular, and triangular. Four species were found to be sulcate-shaped. Flat-shaped was found in two species. Circular and triangular shapes were found in one species each. Petiole was absent in *Polygonum plebieum*. *Rumex crispus* has recorded the largest length and width of collenchyma cells at 14.6 µm and 10.6 µm, respectively. *Persicaria barbata* has a minimum length and width of collenchyma cells at 5 µm and 3.6 µm sequentially, and the largest length and width of parenchyma were found in *Rumex chalepensis* at 23 µm and 17.6 µm, respectively. On the other hand, *Rumex dentatus was* recorded to have the smallest length and width, 4.3 µm and 2.6 µm, respectively. On the adaxial surface, the largest and smallest lengths were noted in *Rumex hastatus* and *Antigonon leptopus* (18 µm and 5 µm, respectively), while the maximum and minimum widths were found in *Rumex chalepensis* (9.5 µm) and *Persicaria barbata* (4.3 µm). The largest length and width of the adaxial surface were examined in *Rumex hastatus* (17.3 µm), *Oxyria digyna,* and *Rumex dentatus,* which were found to have a common maximum width of 8.3 µm. The smallest length was noted in *Antigonon leptopus (*5.4 µm), and the smallest width was found in *Antigonon leptopus* and *Persicaria barbata (*5.3 µm). Crystal bodies were found in *Oxyria digyna.* Our current quantitative and qualitative results are inconsistent with previous findings across the globe in petiole anatomy; eight species showed variations among them. The shapes of the petiole were sulcate, flat, circular, and triangular. Four species were found to be sulcate-shaped. Flat-shaped was found in two species. Circular and triangular shapes were found in one species each. *Rumex crispus* has recorded the largest length and width of collenchyma cells at 14.6 µm and 10.6 µm, respectively. While *Persicaria barbata* has a minimum length and width of the collenchyma cell at 5 µm and 3.6 µm sequentially, the largest length and width of parenchyma were found in *Rumex chalepensis,* 23 µm and 17.6 µm, respectively. On the other hand, *Rumex dentatus was* recorded to have the smallest length and width, 4.3 µm and 2.6 µm, respectively. On the adaxial surface, the largest and smallest lengths were noted in *Rumex hastatus* and *Antigonon leptopus* (18 µm and 5 µm, respectively), while the maximum and minimum widths were found in *Rumex chalepensis* (9.5 µm) and *Persicaria barbata* (4.3 µm). The largest length and width of the adaxial surface were examined in *Rumex hastatus (*17.3 µm), *Oxyria digyna,* and *Rumex dentatus, which were* found to have a common maximum width of 8.3 µm. The smallest length was noted in *Antigonon leptopus* (5.4 µm), and the smallest width was found in *Antigonon leptopus* and *Persicaria barbata* (5.3 µm). Crystal bodies were found in *Oxyria digyna.* Our current results are inconsistent with or comparable with previous findings across the globe [67-76]. Both the number and arrangement of collenchymatous cell layers are relevant in a systematic manner [14].

Metcalfe and Chalk [77] distinguished five distinct petiole types within the Polygonaceae based on their vascular structure. They said that more research into the petiole's vascular architecture might produce patterns useful for diagnosis in particular species and families. Petiolar anatomy was employed by Haraldson [78] as a tool for taxonomy at the generic and tribal levels. Osterova [79] investigated the petiole anatomy of a few *Polygonum* species belonging to the *Persicaria* section. He emphasized that it has a mix of hydromorphic and xeromorphic characteristics, meaning that it has both well-developed aerenchyma and well-developed mechanical and conducting tissue.

The Himalayas have long been a source of essential medicinal plants for local herbalists and plant explorers [80], but over the last two decades, human activities have resulted in the extinction of a number of plants that are beneficial in the wild. The growing demand for raw plant material from the pharmaceutical sector for both domestic and export purposes has led to increased overharvesting of wild medicinal plant species [31, 33, 81]. The Himalayan region's medicinal flora and biodiversity are facing significant human-caused pressures, such as overgrazing, landslides, soil erosion, construction activities, unplanned development, overexploitation, mining, stone quarrying, overharvesting, conversion of grasslands and forests to agriculture, and invasion by alien species [82-84].

Generally, all parts of the plant were employed to heal various ailments. In line with this, Ahmad [85] showed that leaves were the most widely used plant portion in herbal treatments, accounting for approximately 33%. However, our findings are comparable with those of [86], who explored the ethnobotanical use of medicinal plants in Pashtun tribal areas and discovered that the locals primarily employed whole plants and leaves, respectively, to cure a variety of ailments. In numerous cases, multiple components of the same species, such as leaves and aerial parts, were employed in herbal remedies and treatments. Previous research has indicated that leaves are the most common component of traditional herbal medications [31, 53, 87-89].

The RFC results of the current study are comparable to those of previous investigations [31, 33, 34, 53, 87]. Residents are familiar with a variety of plant species that have high RFC values and are extensively dispersed in the region due to traditional therapeutic usage. These plants have the potential to be useful in medication discovery and commercialization [32, 90, 91]. High RFCs indicate a deep understanding and widespread use of traditional uses of plant species throughout communities, emphasizing their importance from both indigenous and cultural perspectives. Moreover, high RFC values indicate that a specific species is commonly used to treat diverse diseases, providing a risk of overexploitation and posing an important threat to conservation efforts. The higher usage value suggests that medicinal plants are used for a variety of purposes and that local communities depend on them to treat a wide range of illnesses [90, 92, 93]. The fact that medicinal plants are widely used in this area highlights how important it is to prioritize their preservation [94]. Because medicinal plants with greater use values are more vulnerable in the area, community-based efforts must be involved in their sustainable management and conservation [31, 34, 53].

Conclusions

The micromorphology of petioles in the Polygonaceae family can help resolve taxonomic problems at the subfamily, tribe, genus, and species levels. The histological structure of the petiole and ethnobotanical uses of 8 species were described and discussed, with a focus on their diagnostic relevance. *Antigonon leptopus* showed the largest petiole length and the shortest lengths were observed in *Rumex dentatus*. *Rumex hastatus* has the maximum number of vascular bundles. *Rumex chalepensis* has the largest parenchyma length, whereas the minimum width was noted in *Oxyria digyna*. *Rumex crispus* has the largest length and width of the collenchyma cell, whereas *Persicaria barbata* has the minimum length and width. The species in this family also have medicinal values. The collected data was analyzed using a variety of quantitative metrics, such as use value (UV) and relative frequency citation (RFC). The majority of medicinal plants utilized by local communities were herbs (87.5%), with leaves (53.85%) being the most commonly used plant parts. Among the investigated taxa, *Oxyria digyna* had the highest use value (1.08), and it also had the highest relative frequency citation (0.88). The study's findings proved to be highly effective in identifying and delimiting Polygonaceae species at the tribe and species levels. Finally, these qualities are key taxonomic features for correctly identifying complex taxa. More phylogenetic and anatomical studies are recommended to highlight the relationship between different genera and species of the Polygonaceae. The findings highlight the urgent need for sustainable practices in medicinal plant research and conservation actions in the Kashmir Himalayas, involving concerted efforts to ensure their preservation and long-term viability.

Ethics Approval and Consent to Participate

All the experiments were performed in accordance with relevant guidelines and regulations.

Author Contributions

Syed Waseem Gillani: Field Collection, Writing-Original draft preparation, Mushtaq Ahmad: Supervision and Review Editing, Muhammad Zafar: Supervision and Reviewing the first draft, Muhammad Manzoor: Statistical analysis, Syed Waseem Gillani: Methodology and Data analysis, Javeed Hussain: Methodology and English editing, Shazia Sultana: Investigation, Syed Waseem Gillani, Muhammad Manzoor, Abdul Basit Mehmood and Bushra Ali: Interpretation of Results, Abd El-Zaher M.A. Mustafa, Mohamed S Elshikh, and Mohamed Ragab AbdelGawwad: Funding acquisition & data curation.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

All the data has been presented in this article.

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