

Foliar anatomy of the genus *Potentilla* L. (ROSACEAE) in Iran and its taxonomic implication

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Abstract

Leaf anatomy of 27 species of *Potentilla* L. (Rosaceae), representing 9 sections and four related genera, especially *Tylosperma* Botsch., *Schistophyllidium* (Juz. ex Fed.) Ikonn., *Drymocallis* Fourr. ex Rydb., and *Sibbaldia* L. from Iran were studied using light microscopy. The most outstanding characters of blade and midrib were measured. The result revealed a number of taxonomically important characters e.g., trichome types, stomata positions, mesophyll types, crystals location and vascular bundle shape. Three types of stomata, two main types of mesophyll and two main types of vascular bundles were identified. Leaf anatomical evidence is of taxonomic value and reflect the ecological differences in the studied species.

Keywords: leaf anatomy; *Potentilla* L.; Rosaceae; Iran

1. Introduction

The genus *Potentilla* L. is a major representative of the tribe *Potentilleae*, in the sub family Rosoideae and family Rosaceae [1]. The species of *Potentilla* are herbaceous and woody perennials, widely distributed in the temperate regions of the northern hemisphere [2, 3]. Iranian species of *Potentilla* grow under different environmental conditions in Irano-Turanian, Hyrcanian (Caspian) and Zagros floristic provinces and in different altitudes ranging from 100 m to 4600 m [4-6].

The genus *Potentilla* includes 2 sections (*Fragariastrum* Koch. and *Eupotentilla* Koch), including (Annuals and Perennials) and 68 species in *Flora Orientalis* [7], 4 groups, 4 subgenera, 14 sections and 53 species in *Flora of Turkey* [8], 6 species in *Flora of Iraq* [9] and 220 species (including: 193 species of *Potentilla* and 27 species of 11 related genera) in flora of the former USSR. [5, 10, 11]. Rechinger reported 51 species of the genus in the area covered by *flora Iranica* (including: Iran, Afghanistan, parts of western Pakistan, northern Iraq, and Azerbaijan [4]. Based on Khatamsaz (1993), this genus includes 4 subgenera: 1) Subgen. *Trichothalamus* (Lehm.) Reichenb (including: *P.* sect. *Xylorrhiza* (Th.Wolf) Schiman-Czeika and *P. lignosa* Wild), 2) Subgen.

Schistophyllidium Juz. (including: *P.* sect. *Schistophyllidium* Juz. and *P. bifurca* L.), 3) Subgen. *Fragariastrum* (Heister ex Faber.) Reichb. (including: two sections *P.* sect. *Speciosae* (Th.Wolf) Juz. and *P.* sect. *Campestres* (Poevrl.) Juz and 3 species), 4) Subgen. *Potentilla* (including: 10 sections, ca. 37 species) [12] and 14 endemic to Iran [13].

Ertter and Attar (2007) reported 58 species of *Potentilla* L., 3 species of *Drymocallis* Fourr. ex Rydb., two species of *Tylosperma* Botsch., and one species each of *Farinopsis* Chrtek & Soják., *Schistophyllidium* (Juz. ex Fed.) Ikonn., *Dasiphora* Raf., *Duchesnea* J. E. Smith. and *Sibbaldia* L. in the area covered by *Flora Iranica* [14].

In the most recent study of the genus in Iran, 39 species of *Potentilla* (including 22 endemic species and two inter sectional hybrids (*P. szovitsii* Th. Wolf.: *P. divaricata* sect. *Rectae* × *P. thuringiaca* sect. *Chrysanthae*) and *P. radiata* Lehm.: *P. argentea* .sect. *Terminales*) × *P. thuringiaca* sect. *Chrysanthae*) were identified [6, 15].

The genus *Potentilla* is well-known for its taxonomic complexity and high degree of morphological variations. The most important studies of the genus were based on macro [4, 12, 16-18] and micro morphological surveys [19-27], especially those of Soják (1998-2010) [1, 5, 11, 27-30]. The phylogentic studies of the genus were carried out especially by Eriksson et al. (1998, 2003) [2, 31] and Dobeš and Paule (2010) [32].

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Metcalf and Chalk (1957) described the general anatomical features of some genera in family *Rosaceae* [33]. However, the anatomical features of the genus *Potentilla* have not yet been studied in detail. Therefore this work aims to characterize the anatomical data of this genus and to evaluate the implication of anatomical characters in taxonomic surveys.

2. Material and Method

In this study, 27 species representing 9 sections of *Potentilla* and four related genera were selected from numerous specimens collected from different parts of Iran (during 2005 to 2007). The voucher specimens were deposited in Tehran University Herbarium (TUH). *Flora Iranica* [4] was the primary reference for identification, with some nomenclatural updates as per Erter and Attar (2007) [14]. The species sampled are listed in Appendix 1. Materials were fixed in alcohol-glycerin (60:40), then the leaf and midrib transverse section were prepared by hand cutting. Foliar cross sections were prepared from the central leaflet of palmate leaves and the most upper leaflet of the pinnate leaves by means of the blade of a razor. Transverse sections were stained by methyl green and Bismarck brown. The observations were carried out by light microscope (Nikon 200 M using a $\times 100$ eyepiece), and photographed by Leitz light microscope (Wetzlar, Nikon camera model Coolpix S10).

3. Results

Leaf anatomical analysis revealed interesting features. The most outstanding characters of blade and midrib were measured (Tables 1 and 2).

3.1. Lamina transverse sections

3.1.1. Trichome

Among the examined species, four different types of trichomes were recognized: Straight (pilose, spinose), crispate, floccose-crispate and glandular hairs (Figs. 1-12).

Glandular trichome with unicellular stalk and single head cell, are present in the majority of the studied taxa (except *Drymocallis* species). In the studied species of *Drymocallis*, glandular trichomes consist of a long multicellular stalk and single head cell. The tallest glands were observed in *Drymocallis* species e.g. *D. poterifolia* (Boiss.) Soják (with 110.4 μm) (Figs. 10 and 11) and *D. rupestris* (L.) Soják (with 130.8 μm) (Figs. 10-12).

3.1.2. Cuticle layer

The minimum cuticular thicknesses of adaxial and abaxial surface were identified in *P. pedata* Nestl. (4.5-6 μm) (Fig. 13) and *P. anserina* L. (3.5-4 μm) (Fig. 14). The maximum cuticular thicknesses of adaxial and abaxial surface were observed in *P. radiata* Lehm. (12-14 μm) and *P. reptans* L. (10-11 μm) (Figs. 15).

3.1.3. Number of epidermal cells

In the transverse sections, epidermal cells are isodiametric, rectangular and in a few cases triangular to polygonal in shape. The lower epidermal cells are smaller than the upper ones. In the majority of the studied taxa e.g. *P. elvendensis* Boiss., single epidermal layer was observed. In contrast, in some species, especially *P. radiata* (with 2) two epidermal layers were present.

3.1.3.1. Epidermal thickness

Minimum adaxial epidermal thickness (10-12 μm) was observed e.g. in *P. elvendensis*, while maximum adaxial epidermal thickness (25-27 μm) was found in *P. reptans* L. (Fig. 15).

3.1.4. Stomata

Based on stomata position on the epidermal level, 3 main type classes were recognized (Table 3):

I type class) Superficial stomata, which includes the following species: *P. micrantha* Ramond ex DC., *P. adschatica* Sommier & Levier ex Keller, *P. hirta* L., *P. pedata* Nestl (Fig. 13), *P. recta* L., *P. szovitsii* Th. Wolf, *P. inclinata* Villars., *P. reptans* and *P. supina* L.

II type class) Superficial-prominent, this type class is observed in the following species: *P. pannosa* Boiss. & Hausskn. (Fig. 16), *D. poterifolia*, *D. rupestris*, *P. anserina* (Fig. 14), *P. argentea* L., *P. meyeri* Boiss., *P. crantzii* (Crantz) Beck ex Fritsch., *P. aucheriana* Th. Wolf, *P. bungei* Boiss., *P. kurdica* Boiss. & Hohen., *P. elvendensis*, *P. nuda* Boiss., *P. petraea* (Willd) and *P. speciosa*. III type class) superficial-hidden. This type class is found in the following species: *P. radiata* (Fig. 17), *Schi. bifurcum* (L.) Ikonn. (Fig. 18), *S. parviflora* L. (Fig. 19) and *T. lignosa* (Willd.) Botsch. (Fig. 20).

2.1.3.1.5. Mesophyll

Two main type classes were identified (Table 4):

I type class) Dorsiventral

II type class) Isobilateral, poorly differentiated palisade and spongy layers (compact tissues).

I type class is further divided into 3 categories:

Table 1. Important and useful anatomical characters of blade for distinguishing species

Species	Blade characters									
	Ad. E. Th (μm)	Ab. E. Th (μm)	P. L/P. Th (μm)	S. L/S. Th (μm)	H/H.K	St.T	A ($^{\circ}$)	No. E.L.	Ad, Ab.C. Th (μm)	L.T
1. <i>Schistophyllidium</i> (Juz. Ex Fed.) Ikonn. 1.1. <i>Schi. bifurcum</i> (L.) Ikonn.	18-20	10-16	3/56-59	-	G, short hair, S.G, 1 celled, 31 μm h, 10 μm H	III	180	1	8-10, 5-6.5	Isobi. LS
2. <i>Tyloperma</i> Botsch. 2. 1. <i>T. lignosa</i> (Willd.) Botsch.	11-14	8-10	2/24-30	2/8-10	S.G, 1 celled 12 μm h, 6 μm H	III	60	1	6-8, 4.5-5	Dorsi. LS
3. <i>Drymocallis</i> Fourr. ex Rydb. 3.1. <i>D. poteriifolia</i> (Boiss.)	10-12	7.5-9	2/18-20	2-3/16.5-18	S.G, 2-3 celled, 208 μm h, 110 μm H.	II	63	1	6-8.5, 4-5.5	Dorsi. LS
3.2. <i>D. rupestris</i> (L.) Soják	18-20	10-12	2/40-42	3-4/40-42	S.G, 3-4 celled, 130 μm h; 20 μm H	II	90	1-2	7.5-9, 5-6	Dorsi. LS
4. <i>Sibbaldia</i> L. 4.1. <i>S. parviflora</i> Willd.	10-12	8-10	2-2.5/28-31	2-3/20-22	S.C, 1 celled 10 μm h; round 6-7 μm H	III	117	1	8-10, 6-7	Dorsi. LS
5. <i>Potentilla</i> L. 5.1. <i>P. sect. Pentaphylloides</i> Tausch 5.1.1. <i>P. anserina</i> L.	18	11	2/50-54	2/(30-34)	S; long crispate	II	63	1	6.5-9, 3.5-4	Dorsi. LS
5.2. <i>P. sect. Terminales</i> (Döll) Gren. & Godr 5.2.1. <i>P. argentea</i> L.	18	12-14	2-3/33-35	2/(19-22)	long crispate	II	85	1	8-10, 3.5-4	Dorsi. LS
5.2.2. <i>P. meyeri</i> Boiss	10-14	6-8	3/28-38	2/14-16	S.G, long crispate	II	59	1	8-10, 6-8	Dorsi. CS
5.2.3. <i>P. inclinata</i> Vill.	16-21	6-9	2/32-36	2/16-18	S short & long	I	82	1-2	10-12, 4.5-6	Dorsi. CS
5.3. <i>P. sect. Aureae</i> (Rydb.) Juz, 5.3.1. <i>P. crantzii</i> (Crantz) Beck ex Fritsch	12-17	8-10	3-4/60-66	2/12-16	S.G, 1 celled, 9h μm , 5-6 μm H.	II	78	1	10-12, 6-8	Dorsi. CS
5.4. <i>P. sect. Rectae</i> (Th. Wolf) Juz. 5.4.1. <i>P. adscharica</i> Sommier & Levier ex R. Keller	16	10	2 / 26-28	3(32-35)	S.G, 1 celled 16-18 μm h, 12-13 μm H	I	50	1	8-10, 4.5-6	Dorsi. LS.
5.4. 2. <i>P. hirta</i> L.	18-21	10-12	4-5/38-40	2/15-18	S-long hair 1 celled	I	83	1-2	8-10, 4.5-5	Dorsi. CS
5.4. 3. <i>P. pedata</i> Nestl.	11-14	8-10	2/35-38	2/20-22	S.G. long hair, 1 celled gland	I	36	1	4-6, 4.5-6	Dorsi. CS
5.4.4. <i>P. recta</i> L.	20-23	10-14	3-4/38-44	2-4/25-37	S, long -short	I	65	1-2	10-12, 8-9.5	Isobi. LS
5.5. <i>P. sect. Rivales</i> Poeverl. 5.5. 1. <i>P. supina</i> L.	14-16	11-14	2/28-32	2/22-24	S.G, 1 celled small 15 μm h, short hair	I	74	1	8.5-11, 5-7.5	Dorsi. CS
5.6. <i>P. sect. Persicae</i> (Th. Wolf) 5. 6. 1. <i>P. aucheriana</i> Th. Wolf	10-14	6.5-8	2/24-27	2/20-24	S.G, 1 celled 14 μm h, 9 μm H	II		1	8-10, 78.5	Dorsi. LS
5.6.2. <i>P. bungei</i> Boiss.	12-16	8-11	2-3/26-30	222-26	S.G, 1 celled with 1 short stalk cell. 26 μm h, 10 μm H	II	85	1	8-10, 5-6.4	Dorsi. CS

Table 1. (Continued)

5.6.3. <i>P. elvendensis</i> Boiss	10-12	6-8	2/22-24	/20-22	S-long short; crispate	II	63		10-12, 5-7.5	Dorsi. LS
5.6.4. <i>P. kurdica</i> Boiss. & Hohen	12-16	6-10	3/48-50	2-3/25-34	S-straight hair	I	58	1	10-12, 8.5-10	Dors. LS
5.6.5. <i>P. nuda</i> Boiss	10-12	6-8	3/34-38	2/22-26	S-1celled	II	62	1	10-12, 6-7	Dorsi. CS
5.6.6. <i>P. pannosa</i> Boiss. Hausskn.	19-22	8-12	3/61-66	--	S, 1 celled either side G, very rare 9-10µm h, 5 µm H	II	40	1-2	8-10, 6.5-8	Isobil. CP
5.6.7. <i>P. petraea</i> Willd.	18-20	8-10	3/50-53	3/36-38	S.G, 1celled, 25µm h, 10 µm H, Crispate-straight		61	1-2	8-10, 7-8.59	Dorsi . LS
5.7. <i>P. sect. Potentilla</i>										
5.7.1. <i>P. reptans</i>										
Accession No. 366391	15-18	12-16	2-2.5/(34-37)	3-3.5/30-35)	S	I	50	1	6-7.5, 6.5-7	Dorsi. CS
36604	18-20	16-19	3/(31-33.5)	4-5/(38-41)	S-very random	I	65	1	6-7.7, 6.5-7	
36626	16-20	2-24	2-2/(36-38)	3-4/(32-35)	S	I	60	1	7-8.5, 6.5-7	
36617	22-25	16-19	2/(23-25)	3/(35-37)	S	I	96	1	6-8, 6.5-7	
36618	25-27	16-18	2/(25-28)	3/(30-40)	S	I	58	1	10-12, 10-11	
5.8. <i>Speciosae</i> (Th. Wolf) Juz.										
5.8. 1. <i>P. speciosa</i> (Th. Wolf) Juz.	10-12	6.5-8.7	3/27-30	2/20-24	S.G crispate floccose either side	II	114	1	12.5-14, 8-9.5	Dorsi. LS
5.9. <i>P. sect. Micranthae</i> Soják.										
5.9. 1. <i>P. micrantha</i> Ramond ex DC.	12-16	9-11	2-3/25-28	2/15-18	S.G. 2 celled, 54 µm 12 µm H, 26 µm h	I	110	1	6-8, 4.5-6	Dorsi. LS
Intersectional Hybrids										
5.10.1. <i>P. radiata</i> Lehm	18-20	8-10	3/44-48	2/20-25	S short-long hair	III	47	2	12-14, 8-10	Dorsi . CS
5.10.2. <i>P. szovitsii</i> Th.Wolf	20-22	15-18	3/54-56	2/12-14	S.G. 1celled, 38 µm h, 18 µm H	I	114	1-2	8-10, 8-10	Dorsi. CS

Abbreviations: Ad. E. Th: Adaxial Epidermis Thickness; Ab. E. Th: Abaxial Thickness; P. L/P. Th: Palisade Parenchyma Layers/Palisade Thickness; S. L/S. Th: Spongy Parenchyma Layers/Spongy Thickness; H/H. K: Hair/Hair kind (S: Simple; G: Glandular; H: Head; h: Height); A: Angle between two halves of the blade; Ad -Ab. C: Adaxial and Abaxial Cuticle Thickness; No. E. L: Number of Epidermal Layers; St. T: Stomata Types; L.T: Leaf type (Iso: Isobilateral; Dorsi: Dorsiventral; LS: Loose spongy; CS: Compact spongy; CP: Compact palsied)

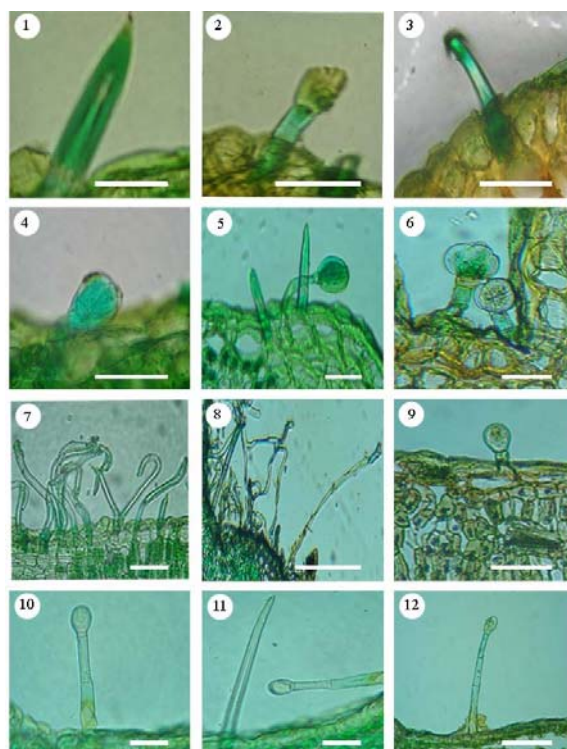
Table 2. Important and useful anatomical characters of midrib

Species	Blade characters												
	D.VB. Ad- D.VB. Ab	L/W M	L/W X	L/W Ph	Ad. C. Th (μ m)	Ad. C. Th (μ m)	Ab. P. Th. (μ m)	I. Ph. Th. (μ m)	T.V.B	Ad. E. Th. (μ m)	Ab. E. Th. (μ m)	H. K	M. Sh & No.
1. <i>Schistophyllidium</i> (Juz. Ex Fed.) Ikonn. 1. <i>Schi. bifurcum</i> (L.) Ikonn.	12-18	0.93	2.7	2.5	12-16	34	16-20	10-12	Col	18-21	10-12	S	Sub.1
2. <i>Tylosperma</i> Botsch. 2. 1. <i>T. lignosa</i> (Willd.) Botsch.	10-15	0.96	118	1.2	18-21	18-25	20-25	8-10	col	12-16	8-11	S.G	Sub.1
3. <i>Drymocallis</i> Fourr. ex Rydb. 3.1. <i>D. poteriifolia</i> (Boiss.)	6-19	1.04	1.02	1.05	6-8.5	17.5-20	10-12	8-19	Amph	6-8	4-5.5	S.G	Tri. 1
3.2. <i>D. rupestris</i> (L.) Soják	8-22	086	1.66	1.44	10-12	20-23	10-12	10-12	Amph	12-14	5-8	S.G	Tri. 1
4. <i>Sibbaldia</i> L. 4.1. <i>S. parviflora</i> Willd.	8-15	1.47	1.04	1.15	4-6	10-14	10-12	6-4	Amph	5-7	3.5-5	S	Tri. 1
5. <i>Potentilla</i> L. 5.1. <i>P. sect. Pentaphylloides</i> Tausch 5.1.1. <i>P. anserina</i> L.	12-30	1.2	1.58	1.23	16-19	16-18	35-38	18	Amph	16	8	S.G	Tri. 1
5.2. <i>P. sect. Terminales</i> (Döll) Gren. & Godr 5.2.1. <i>P. argentea</i> L.	15-31	1.35	1.6	1.13	10	26-28	40-12	4-8	Amph	14-16	10-12	S	Sub.1
5.2.2. <i>P. meyeri</i> Boiss	8-17	3(1.57)	1.53	1.23	10	20-28	12-16	5-8	Amph	8-10	6-9.5	S	Sub.-Tri 1-3
5.2.3. <i>P. inclinata</i> Vill.	7-15	1.71	1.66	1.7	12	18-22	10-18	6.5-8	Amph	8-12	6-8.5	S	Sub. 1
5.3. <i>P. sect. Aureae</i> (Rydb.) Juz, 5.3.1. <i>P. crantzii</i> (Crantz) Beck ex Fritsch	6-12	1.01	1.5	1.18	10	18-20	10	6.5-8	Amph	12-16	6.5-8.5	S.G	Sub. 1
5.4. <i>P. sect. Rectae</i> (Th. Wolf) Juz. 5.4.1. <i>P. adscharica</i> Sommier & Levier ex R. Keller	16-35	1.17	1.8	1.16	8-10	20-22	24-26	8-12	Amph	12-14	10-11.5	S.G	Sub. 1
5.4.2. <i>P. hirta</i> L.	6-18	1.31	1.63	1.49	12-15	35-40	16-20	14-18	Amph	14-16	8-10	S	Sub. 1
5.4.3. <i>P. pedata</i> Nestl	9-29	1.51	1.31	1.41	12-16	24-28	18-20	8-10	Amph	14-16	10-14	S	Sub. 1
5.4.4. <i>P. recta</i> L.	6-18	1.31	1.63	1.49	12-15	35-40	16-20	14-18	Amph	14-16	8-10	S	Sub. 1
5.5. <i>P. sect. Rivales</i> Poeverl. 5.5.1. <i>P. supina</i> L.	8-22	1.6	1.52	1.8	12-16	20-24	21-27	8-10	Col	14-18	12-15	S.G	Sub.1
5.6. <i>P. sect. Persicae</i> (Th. Wolf) 5.6.1. <i>P. aucheriana</i> Th. Wolf.	8-18	1.43	1.5	1.73	14	26	16-20	14-18	Amph	10-14	10-12	S.G.	Sub.1
5.6.2. <i>P. bungei</i> Boiss.	8-20	1.8	1.42	1.43	8-10	10-12	10	8-10	Amph	16-19	11-14	S.G	Sub.1

Table 2. (Continued)

5.6.3. <i>P. elvendensis</i> Boiss.	8-30	1.09	1.41	1.32	9-12	30-35	28-32	10-14	Amph	10-14	5-856	S.G	Sub.1
5.6.4. <i>P. kurdica</i> Boiss. & Hohen	10-23	1.23	1.12	1.25	8-10	25-29	22-27	8-10	Col	12-14	8-10	S.G	Sub.1
5.6.5. <i>P. nuda</i> Boiss	14-30	0.88	1.23	1.25	32-28	42-45	20-24	10-12	Col	12-16	12-14	S.G	Tri.1-3
5.6.6. <i>P. pannosa</i> Boiss. & Hausskn.	8-18	1.15	1.44	1.88	8-10	16-20	8-12	4-6	Col-	18-21	8-10	S.G	Tri.1
5.6.7. <i>P. petraea</i> Willd.	8-12	0.69	1.5	1.25	16-18	34-36	16-20	8-10	Col	12-14	8-12	S.G	Tri.1-3
5.7. <i>P. sect. Potentilla</i>													
5.7.1. <i>P. reptans</i>	16-39	1.08	1.35	2.59	14	58	41.5	8-11	Amp	20-32.5	20-22	S	Sub.1
5.8. <i>P. sect. Speciosae</i> (Th. Wolf)													
5.8.1. <i>P. speciosa</i> (Th.Wolf) Juz	14-36	0.91	1.07	1.6	4-6	10-14	4-8	2-4	Col	8-10	3.5-5	S	Tri.1
5.9. <i>P. sect. Micranthae</i> Soják.													
5.9.1. <i>P. micrantha</i> Ramond ex	9-19	1.56	1.42	1.16	20-25	21-24	21-25	14-17	Col	12-16	9.5-11	S.G	Sub.1
Intersectional Hybrids													
5.10.1. <i>P. radiata</i> Lehm	5-22	1.35	1.47	1.31	8-10	23-28	18-24	8-11	Amph	10-15	7-9.5	S.G	Sub.1
5.10.2. <i>P. szovitsii</i> Th.Wolf	14-34	1.2	1.35	1.28	13-15	40-43	30-33	7-10	Col	22-25	16-18	S.G	Sub.1

Abbreviations: D.VB. Ad. and D.VB. Ab: Distance of Vascular Bundle from Adaxial and Abaxial Epidermis; L/W M: Length/Width of Midrib; L/W X: L/W Ph: Length / Width of Phloem; Ad. C. Th: Adaxial Collenchyma Thickness; Ab. C. Th: Abaxial Collenchyma Thickness; Ad. P. Th: Adaxial Parenchyma Thickness; Ab. P. Th: Abaxial Parenchyma Thickness; I. Ph. Th: Internal Phloem Thickness; T.V.B: Type of Vascular Bundle (Col: Colateral; Amph: Amphicribal); Ad. E. Th: Adaxial Epidermis Thickness; Ab. E. Th: Abaxial Epidermis Thickness; H.K: Hair/Hair Kind (S: Simple; G: Glandular); M. Sh & No: Midrib Shape; number (Sub: Suborbiculare; Tri: Triangular)



Figs. 1-6. Straight trichomes and glands of different species of *Potentilla*: 1-2. *P. micrantha*; 3-4. *P. supina*; 5. *P. adsharica*; 6. *P. szovitsii*; 7. crispate trichome of *P. meyeri*; 8. floccose-crispate trichome of *P. speciosa*; 9. gland of *Schi. bifurcum*; 10. gland and stomata of *T. lignosa*; 10-11. multicellular trichome and glands of *D. poteriiifolia*; 12. *D. rupestris*, Scale bar = 0.05 μm

Category I a) Dorsiventral with 2-2.5 palisade parenchyma layers. This category was observed, especially in *P. micrantha*. (Fig. 21), *S. parviflora* (Fig. 22) and *T. lignosa* (Willd.) Botsch (Fig. 23).

Category I b) Dorsiventral, with 3-4 layers of palisade parenchyma. This category was recognized in some species e.g. *P. crantzii* (Fig. 24), *P. petraea* (Fig. 25) and *P. speciosa* (Fig. 26).

Category I c) Dorsiventral, with compact palisade and spongy parenchyma layers. This category was observed in some species e.g. *P. hirta* (Fig. 27), *P. reptans* (Fig. 28) and *P. szovitsii* (Fig. 29) with 3-4 compact palisade and 2-3 spongy parenchyma layers.

II type class is identified in *Schi. bifurcum* (Fig. 18) and *P. pannosa* Boiss. & Hausskn.

3.1.6. Crystals

Calcium oxalate crystals are found in different parts of the lamina. In the majority of species e.g., *P. anserina*, *P. argentea*, *P. adscharica*, *P. crantzii*, *P. hirta*, *P. meyeri*, *P. elvendensis* (Fig. 30), *P. pedata*, *P. petraea*, *P. recta*, *P. szovitsii*, *P. reptans*, *Schi. bifurcum* and *T. lignosa* clustered calcium oxalate crystals were present in the palisade parenchyma exactly under the epidermis layer, while in *P. bungei* and *P. kurdica* (Fig. 31) crystals

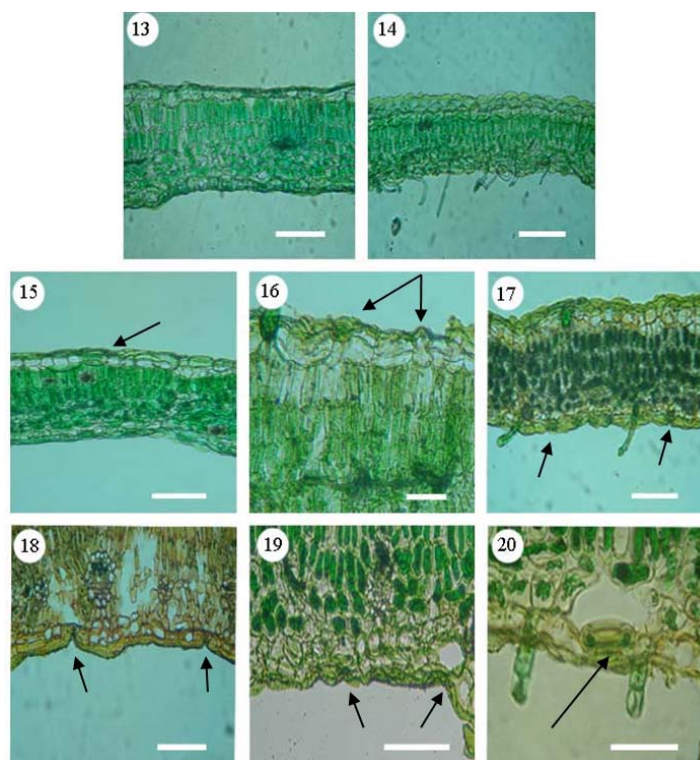
were identified in palisade and spongy layers. In *P. radiata* large numbers of small scattered sand crystals were observed. In contrast, crystals were absent in *P. pannosa*.

3.1.7. Leaf angle

Leaf angles between two halves of the blade vary from species to species e.g. *P. recta* (39°, minimum among the studied species) (Fig. 32), *P. radiata* (42°) (Fig. 33), *P. anserina* (63°) (Fig. 34), *P. argentea* (85°) (Fig. 35), *P. micrantha* (110°) (Fig. 36), *P. crantzii* (78°) (Fig. 37), *P. petraea* (61°) (Fig. 38), *D. rupestris* (90°) (Fig. 39), *D. poteriiifolia* (63°) (Fig. 40), *T. lignosa* (60°) (Fig. 41), *S. parviflora* (117°) (Fig. 42), and *Schi. bifurcum* (180°, maximum among the studied species) (Fig. 43).

3.2. Midrib transverse sections

The midrib is suborbicular to triangular in transactional outline. The epidermal layer of midrib is covered by four trichome types on the abaxial and adaxial surface. The shape of midrib changes from suborbiculare to triangular. The epidermis and vascular bundle are the two main parts of the midrib.



Figs. 13-20. *P. pedata*; 14. *anserina* (minimum adaxial and abaxial cuticular thicknesses); 15. *P. reptans* (maximum adaxial and abaxial cuticular thicknesses and superficial-stomata); 16. *P. pannosa* (superficial-prominent stomata); 17. *P. radiata*; 18. *Schi. bifurcum*; 19. *S. parviflora*; 20. *T. lignosa* (superficial-hidden stomata) scale bar = 0.05 μm

3.2.1. Epidermis

In the majority of the species single epidermal layer was observed. Minimum adaxial epidermal cell (5-7 μm) was observed in *S. parviflora*, while maximum abaxial epidermal cell (20.7-32.5 μm) was measured in *P. reptans*.

3.2.2. Vascular bundle

Two main types of vascular bundles: amphicribal e.g., *P. recta* (Figs. 44-45), *P. aucheriana* (Fig. 46), *P. meyeri* (Figs. 47-48), *P. szovitsii* (Fig. 49) and collateral e.g., *P. petraea* (Figs. 50-51), *P. micrantha* (Fig. 52), *P. nuda* (Fig. 53-54) and *Schi. Bifurcum* (Fig. 55), were recognized. The distance of the vascular bundle from the adaxial epidermal layer varies from a minimum 5 μm (in *P. radiata* and *P. hirta*) to a maximum 25 μm (in *Schi. bifurcum*). The distance of the vascular bundle from the abaxial epidermal layer ranges from a minimum 12 μm (in *P. crantzii* and *P. petraea*) to a maximum 45 μm (in *P. szovitsii*). The minimum length/width ratio (0.86 μm) was observed in *D. rupestris* and maximum length/width ratio (1.51 μm) was identified in *P. micrantha*. Xylem length/width ratio is measured. The xylem length/width ratio was

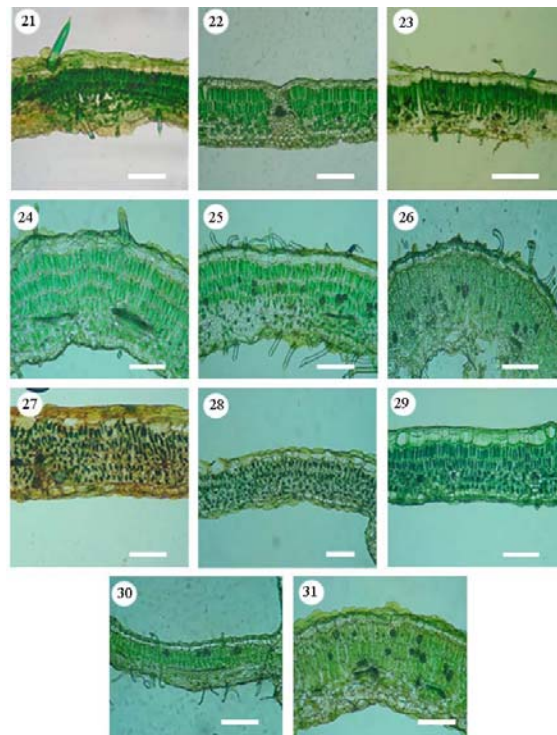
minimum (1.03 μm) in *P. argentea* and maximum (2.7 μm) in *Schi. bifurcum*. Phloem length/width ratio was minimum (1.00 μm) in *P. reptans* and maximum (2.5 μm) in *Schi. Bifurcum*.

3.2.3. Collenchyma

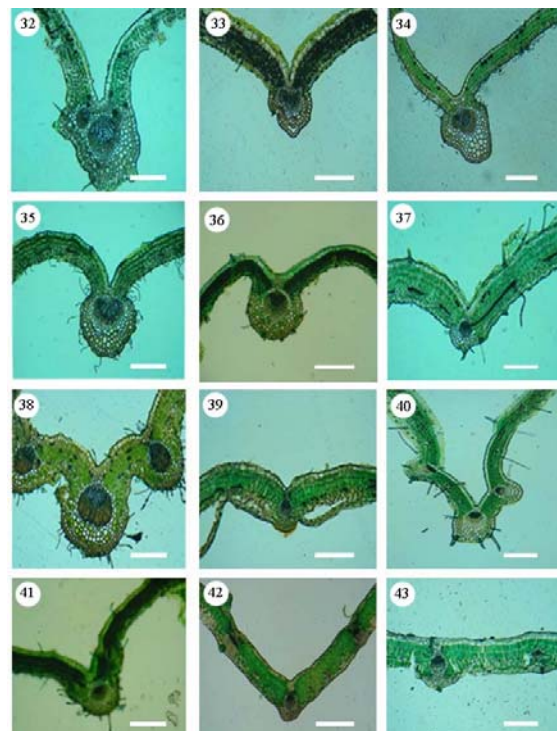
Distribution of the collenchyma in the adaxial and abaxial levels and their thickness were measured. Adaxial collenchyma thickness ranges from a minimum 4-6 μm in *P. speciosa* to a maximum 32-28 μm in *P. nuda*. Abaxial collenchyma thickness ranges from a minimum 10 μm (in *P. bungei*) to a maximum 58 μm in *P. reptans*. Parenchyma tissue includes only abaxial tissue. Minimum abaxial parenchyma thickness (4-6 μm) was found in *P. pannosa* and the maximum (14-18 μm) in *P. recta*.

4. Discussion

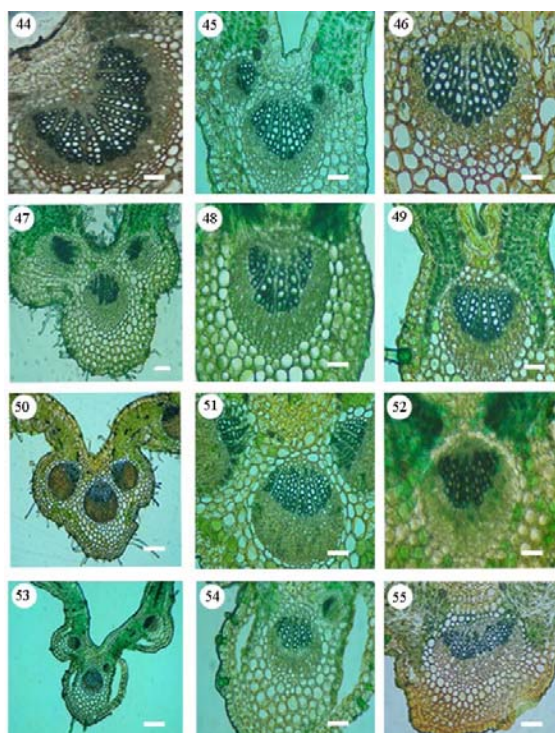
Trichomes types, stomata positions, mesophyll types (dorsiventral-isobilateral), crystals location and vascular bundle shape are the most important anatomical characteristics of the genus *Potentilla* L. Trichome types were used at sectional and species levels in previous taxonomic studies [10, 27-30].



Figs. 21-31. Mesophyll of I type class category I a: 21. *P. micrantha*; 22. *S. parviflora*; 23. *T. lignosa*; category I b: 24. *P. crantzii*; 25. *P. petraea*; 26. *P. speciosa*; category I c: 27. *P. hirta*; 28. *P. reptans*; 29. *P. szovitzii*; 30. *P. elvendensis*; 31. *P. kurdica*; scale bar = 0.05 μm



Figs. 32-43. Leaf angles of different species 32. *P. recta*; 33. *P. radiata*; 34. *P. anserina*; 35. *P. argentea*; 36. *P. micrantha*; 37. *P. crantzii*; 38. *P. petraea*; 39. *D. rupestris*; 40. *D. poterifolia*; 41. *T. lignosa*; 42. *S. parviflora*; 43. *Schi. Bifurcum*; scale bar = 0.05 μm



Figs. 44-55. Amphicribal vascular bundle: 44-45. *P. recta*; 46. *P. aucheriana*; 47-48. *P. meyeri*; 49. *P. szovitsii*; collateral vascular bundle: 50-51. three vascular bundles of *P. petraea*; 52. *P. micrantha*; 53-54. *P. nuda*; 55. *Schi. bifurcum*; scale bar = 0.05μm

Table 3. Leaf type class based on Stomata

Species	Genera & Sections	Stomata type class
1. <i>P. adscharica</i>	<i>Rectae</i> (Th. Wolf) Juz.	I Superficial
2. <i>P. hirta</i>	<i>Rectae</i> (Th. Wolf) Juz.	
3. <i>P. pedata</i>	<i>Rectae</i> (Th. Wolf) Juz.	
4. <i>P. recta</i>	<i>Rectae</i> (Th. Wolf) Juz.	
5. <i>P. inclinata</i>	<i>Terminales</i> (Döll) Gren. & Godr	
6. <i>P. micrantha</i>	<i>Micranthae</i> Soják.	
7. <i>P. reptans</i>	<i>Potentilla</i>	
8. <i>P. supina</i>	<i>Rivales</i> Poeverl.a L.	
9. <i>P. szovitsii</i>	<i>Rectae</i> x <i>Chrysanthae</i>	
10. <i>D. poterifolia</i>	<i>Drymocallis</i> Fourr. ex Rydb	II Superficial -prominent
11. <i>D. rupestris</i>	<i>Drymocallis</i> Fourr. ex Rydb	
12. <i>P. anserina</i>	<i>Pentaphylloides</i> Tausch	
13. <i>P. argentea</i>	<i>Terminales</i> (Döll) Gren. & Godr	
14. <i>P. meyeri</i>	<i>Terminales</i> (Döll) Gren. & Godr	
15. <i>P. crantzii</i>	<i>Aureae</i> (Rydb.) Juz.	
16. <i>P. aucheriana</i>	<i>Persicae</i> (Th. Wolf) Juz.	
17. <i>P. bungei</i>	<i>Persicae</i> (Th. Wolf) Juz.	
18. <i>P. elvendensis</i>	<i>Persicae</i> (Th. Wolf) Juz.	
19. <i>P. kurdica</i>	<i>Persicae</i> (Th. Wolf) Juz.	
20. <i>P. nuda</i>	<i>Persicae</i> (Th. Wolf) Juz.	
21. <i>P. pannosa</i>	<i>Persicae</i> (Th. Wolf) Juz.	
22. <i>P. petraea</i>	<i>Persicae</i> (Th. Wolf) Juz.	
23. <i>P. speciosa</i>	<i>Speciosae</i> (Th. Wolf) Juz.	
24. <i>Sch. bifurcum</i>	<i>Schistophyllidium</i> (Juz. Ex Fed.) Ikonn	III Superficial-Hidden
25. <i>S. parviflora</i>	<i>Sibbaldia</i> L.	
26. <i>T. lignosa.</i>	<i>Tyloperma</i> Botsch.	
27. <i>P. radiata</i>	<i>Terminales</i> x <i>Chysanthae</i>	

Table 4. Leaf type class based on mesophyll

Species	Genera & Sections	mesophyll type class	
1. <i>P. adschatica</i>	<i>Rectae</i> (Th. Wolf) Juz.	I Category a 2-2.5 Palisade parenchyma/ Loose spongy parenchyma	I Dorsiventral
2. <i>P. anserina</i>	<i>Pentaphylloides</i> Tausch		
3. <i>P. aucheriana</i>	<i>Persicae</i> (Th. Wolf) Juz		
4. <i>P. bungei</i>	<i>Persicae</i> (Th. Wolf) Juz		
5. <i>P. elvendensis</i>	<i>Persicae</i> (Th. Wolf) Juz		
6. <i>P. inclinata</i>	<i>Terminales</i> (Döll) Gren. & Godr		
7. <i>P. micrantha</i>	<i>Micranthae</i> Soják.		
8. <i>P. pedata</i>	<i>Rectae</i> (Th. Wolf) Juz.		
9. <i>P. supina</i>	<i>Rivales</i> Poeverl. a L.		
10. <i>D. poteriiifolia</i>	<i>Drymocallis</i> Fourr. ex Rydb		
11. <i>D. rupestris</i>	<i>Drymocallis</i> Fourr. ex Rydb		
12. <i>S. parviflora</i>	<i>Sibbaldia</i>		
13. <i>P. argentea</i>	<i>Terminales</i> (Döll) Gren. & Godr		
14. <i>P. crantzii</i>	<i>Aureae</i> (Rydb.) Juz,		
15. <i>P. kurdica</i>	<i>Persicae</i> (Th. Wolf) Juz.		
16. <i>P. meyeri</i>	<i>Terminales</i> (Döll) Gren. & Godr		
17. <i>P. nuda</i>	<i>Persicae</i> (Th. Wolf) Juz.		
18. <i>P. pannosa</i>	<i>Persicae</i> (Th. Wolf) Juz		
19. <i>P. petraea</i>	<i>Persicae</i> (Th. Wolf) Juz.		
20. <i>P. speciosa</i>	<i>Speciosae</i> (Th. Wolf) Juz.		
21. <i>P. hirta</i>	<i>Rectae</i> (Th. Wolf) Juz.	I Category c 3-5 Palisade parenchyma/ Compact spongy parenchyma	
22. <i>P. recta</i>	<i>Rectae</i> (Th. Wolf) Juz.		
23. <i>P. reptans</i>	<i>Potentilla</i>		
24. <i>P. radiata</i>	<i>Terminales</i> x <i>Chrysanthae</i>		
25. <i>P. szovitsii</i>	<i>Rectae</i> x <i>Chrysanthae</i>		
26. <i>Sch. bifurcum</i>	<i>Schistophyllidium</i> (Juz. Ex Fed.) Ikonn	II Isobilateral	
27. <i>T. lignosa</i>	<i>Tylosperma</i> Botsch.		

The presence of calcium oxalate crystals in Rosaceae was reported by Metcalfe and Chalk (1957) [33] and Ontivero et al. (2000) [34]. In the present study all the studied species of *P. Sect. Persicae* (except *P. pannosa*) possess randomly scattered crystals in palisade and spongy parenchyma layers. In contrast, all the studied species of *P. sect. Rectae* have scattered crystals only in palisade layers. In *P. anserina* large numbers of crystals are present in the palisade parenchyma layer very close to the upper epidermis. However, the variation in the crystals localities was noticed in four populations of *P. argentea*. In three populations (collected from Guilan Siyahkal, Ardabil Sabalan, Mazandaran Chaloos road) [Appendix 1] only a few crystals were observed, while in the fourth population (collected from Guilan, Spili) [Appendix 1], a large number of calcium oxalate crystals were identified in both the palisade and spongy parenchyma.

According to previous authors [33 and 35] in *Potentilla*, stomata are ranunculaceous type. In the present study, based on stomata position 3 main type classes were recognized (Table 3). All studied species of *P. sect. Rectae* (Th. Wolf) Juz. and *P. sect. Micranthae* Soják (*P. micrantha*) have superficial stomata, while all studied species of *P. sect. Persicae* (Th. Wolf) Juz and *P. sect. Speciosae* (Th. Wolf) Juz have superficial-prominent stomata. Some taxa e.g. *Schi. bifurcum*, *T. lignosa*, *P. radiata* and *Sibbaldia* are adapted to semi-arid

conditions and possess hidden stomata and heavy cuticle coverage.

Based on the present result two main type classes of mesophyll were identified (Table 4). In the I type class (including 3 categories), the studied species of I and II categories are adapted to a wet ecological climate and humid condition have wide spongy layers, while all studied species III category prefer a semiarid condition and have compact mesophyll with limited spongy tissue. According to Ivanova et al. (2002) the proportion of palisade to spongy mesophyll tissues varies with plant species and growth conditions [36].

Many species of *Potentilla* grow in the alpine regions. Based on P'yankov and Kondrachuk (2003), the types of changes in mesophyll structure are associated with plant adaptation in mountainous conditions. These changes manifested themselves in different numbers of palisade layers [37]. This is observed in different species and among different taxa of the same species. Foliar anatomy of 5 populations of *P. reptans*, (the most commonly distributed species of *Potentilla* in the country) revealed variation in the proportion of palisade to spongy mesophyll tissues (Fig 40-42). Foliar flexibility and adaptation of leaf internal structures to different environmental conditions (climatic factors and altitudes) are highly expressed in *P. reptans*. According to Metcalfe and Chalk [33], in different species of Rosaceae and *Potentilla* L. a principal solitary crescent-shaped bundle, with smaller or very small subsidiary ones in or towards

the wings are present. In different species of *potentilla* L. e.g. *P. adscharica*, *P. anserina*, and *P. aucheriana* (Fig. 51) a solitary crescent-shaped bundle was observed. While some studied species such as *P. meyeri* (Fig. 53) and *P. petraea* (52 and 55), have three vascular bundles.

5. Conclusion

The result of the present survey revealed a number of taxonomically important characters (e.g. trichome types, stomata positions, mesophyll types, crystals location and vascular bundle shape) that reflect the ecological differences in the studied species. The presence of compact mesophyll; hidden stomata and heavy cuticle coverage are the major anatomic characters of the species growing under semiarid condition. In contrast, species adapted to a wet climate have larger mesophyll, superficial, superficial-prominent stomata.

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APPENDIX

	IRAN: Province, Collector, Date	Accession No.
1. Schistophyllidium (Juz. ex Fed.) Ikonn.	Guilan, Asalem-KhalKhal road, Faghir. 6. 6. 06	36600 (TUH)
1.1. <i>Schi. bifurcum</i> (L.) Ikonn.		
2. Tylosperma Botsch	Tehran, Tehran-Shemshak road, Ghahreman & Mozaffarian. 20.7. 88	6212 (TUH)
2.1. <i>T. lignosa</i> (Willd.) Botsch.		
3. Drymocallis Fourr. ex Rydb.	Mazandaran, Firoozkooh, Attar. 12. 6. 06	39000 (TUH)
3.1. <i>D. rupestris</i> (L.) Soják.		
3.2. <i>D. poteriifolia</i> (Boiss.) Soják.	Fars, Abadeh, Termeh & Izadyar.7. 6. 1969	36855 (IRAN)
4. Sibbaldia L.		
4.1. <i>S. parviflora</i> Willd.	Guilan, Asalem-KhalKhal road, Termeh & Mousavi. 19. 5. 74	36839 (IRAN)
5. Potentilla L.		
5.1. P. sect. Pentaphylloides Tausch.		
5.1.1. <i>P. anserina</i> L.	Mazandaran, Firooskooh, Attar .27. 6. 05	39001 (TUH)
5.2. P. sect. Terminales (Döll) Gren. & Godr.		
5.2.1. <i>P. argentea</i> L.	Guilan, Sipili, Faghir.16 .6. 06	36538 (TUH)
<i>P. argentea</i> L.	Guilan, Siyahkal, Faghir.16. 6. 06	36585 (TUH)
<i>P. argentea</i> L.	Ardabil, Sabalan mount, Faghir. 14. 5. 06	36614 (TUH)
<i>P. argentea</i> L.	Mazandaran, Chlous road, Faghir. 16. 7. 06	36595 (TUH)
5.2.2. <i>P. meyeri</i> Boiss.	Guilan, Siyahkal, Spili, Larikhani, Saidi. 13. 5. 93	18855 (TUH)
5.2.3. <i>P. inclinata</i> Vill.	Guilan, Siyahkal, Spili, Larikhani, Saidi. 13. 5. 93	33466 (TUH)
5.3. P. sect. Aureae (Rydb.) Juz.		
5.3.1. <i>P. crantzii</i> (Crantz) Beck ex Fritsch.	Mazandaran, Ramsar, Samamous mountain, Faghir. 22. 7. 06	36631 (TUH)
5.4. P. sect. Rectae (Th.Wolf) Juz.		
5.4.1. <i>P. recta</i> L.	Guilan, Damash-Jirandeh, Faghir. 16. 6. 06	36598 (TUH)
5.4.2. <i>P. pedata</i> Nestl.	Guilan, Siyahkal, Faghir . 16. 6. 06	36588 (TUH)
5.4.3. <i>P. hirta</i> L.	Mazandaran, Chlous road, Faghir. 16. 7. 06	36627 (TUH)
5.4.4. <i>P. adscharica</i> Sommier & Levier ex R. Keller	Azarbaijan, Kalibar, Attar & Dadjou . 17. 7. 93	17184 (TUH)
5.5. P. sect. Rivales Poeverl.		
5.5.1. <i>P. supina</i> L.	Qazvin, Alamout Ghahreman & Mozaffarian. 11. 8. 91	9963 (TUH)
5.6. P. sect. Persicae (Th. Wolf) Juz.		
5.6.1. <i>P. bungei</i> Boiss.	Kordestan, Marivnan-Paveh, Maroufi & Mohamadi. 21.3. 03	6494
5.6.2. <i>P. elvendensis</i> Boiss.	Fars, Abadeh, Termeh. 5.7.79	36763 (IRAN)
5.6.3. <i>P. kurdica</i> Boiss. & Hohen.	Kordestan, Sanandaj, Saral, Hanaganvillage, Maroufi . 1.6. 07	8060
5.6.4. <i>P. pannosa</i> Boiss. & Hausskn.	Hamedan, Alvand southern slope, Mozaffarian. 8. 7. 88	64940 (TUH)
5.6.5. <i>P. nuda</i> Boiss.	Yazd, Chenaran, Ardakan-Hezarmasjed, Termeh & Mousavi. 18. 8. 77	36827 (IRAN)
5.6.6. <i>P. aucheriana</i> Th.Wolf.	Mazandaran, Kelardasht, Khoramdasht, 3400m-3900m, Faghir.12. 8. 06	36630 (TUH)
5.6.7. <i>P. petraea</i> Willd.	Mazandaran, Ramsar, Samamous mount., Faghir. 22 .7. 06	36632 (TUH)
5.7. P. sect. Potentilla		
5.7.1. <i>P. reptans</i> L.	Guilan, Siahkal, Faghir.16. 6. 06	36639 (TUH)
	Mazandaran, youshbaladeh. 18. 5. 06	36626 (TUH)
	Guilan, Asalem-Khalkal, Faghir .6. 6. 06	36617 (TUH)
	Guilan, Asalem-Khalkal, Faghir .6. 6. 06	36618 (TUH)
	Mazandaran, Javaherdeh, Faghir. 26.7. 09	36604 (TUH)
5.8. P. sect. Speciosae (Th.Wolf) Juz.		
5.8.1. <i>P. speciosa</i> Willd.	East Azarbaijan, Ormiyeh , Ghahreman & Mozaffarian. 29. 6. 97	20056 (TUH)
5.9. <i>P. sect. Micranthae</i> Soják		
5.9.1. <i>P. micrantha</i> Ramond ex DC.	Guilin, Asalem-Khalkal, Faghir .6. 6. 06	36602 (TUH)
Intersectional Hybrids		
Terminales × Chysanthae		
5.10.1. <i>P. radiata</i> (Th.Wolf) Juz.	Guilan, Damash-Jirandeh, Faghir.16. 6. 06	36620 (TUH)
Rectae × Chysanthae		
5.10.2. <i>P. szovitsii</i> Th.Wolf.	Guilan, Damash-Jirandeh, Faghir 16. 6. 06	36593 (TUH)