Comparative achene morphology of *Persicaria* sect. *Cephalophilon* and related taxa (Polygonaceae)

Min-Jung KONG and Suk-Pyo HONG*

Laboratory of Plant Systematics, Department of Biology, Kyung Hee University, Seoul 02447, Korea
(Received 4 June 2018; Revised 22 June 2018; Accepted 27 June 2018)

ABSTRACT: The achene morphology of 21 taxa of *Persicaria* sect. *Cephalophilon* including the related taxa of *Koenigia* (Polygonaceae) was examined using stereo microscopy and scanning electron microscopy to evaluate its diagnostic values and taxonomic implications. The outlines of achene were narrowly ovoid to broadly ovoid in general, and elliptic achenes were observed in *Koenigia* taxa, with the achene shape being biconvex or trigonous. The size of the *P.* sect. *Cephalophilon* taxa ranged from $0.93\text{--}3.55 \times 0.80\text{--}2.31$ mm and the size of examined *Koenigia* taxa ranged from $1.10\text{--}1.84 \times 0.79\text{--}1.02$ mm. The smallest achenes in *P.* sect. *Cephalophilon* were observed in *P. humilis* and the largest achenes were found in *P. chinensis* var. *chinensis*. Furthermore, infra-specific taxa of *P. chinensis* had relatively large achenes compared to other taxa within the section. Most taxa of *P.* sect. *Cephalophilon* had dull achenes, while four taxa had glossy achenes (*P. capitata*, *P. runcinata* var. *runcinata*, *P. runcinata* var. *sinensis*, and *P. sinuata*). Four types of surface patterns (smooth, tubercles, small pits, and irregular ridge) were observed, and the tubercles were divided into two subtypes according to the grouping and distribution pattern. The achene morphology of *P.* sect. *Cephalophilon* is described and compared, and its taxonomic implications are also discussed.

Keywords: achene morphology, micromorphology, SEM, pericarp, *Persicaria* section *Cephalophilon*

Persicaria (L.) Mill., a genus within the tribe Persicarieae Dumort., consists of approximate 100 species. It is characterized by many-flowered spicate, capitate or panicle inflorescence, a simple leaf, chartaceous ochrea entire or ciliate, 4–5 tepals, and 4–8 stamens (Haraldson, 1978; Freeman and Reveal, 2005). *Persicaria* has often been suggested as a member of *Polygonum* L. s.l. (Li et al., 1998, 2003; Zhu et al., 2007), while various morphological and molecular data support the independence of the genus *Persicaria* from *Polygonum* s.l. based on the inflorescence, ochrea type, tepal venation, or filament shape (Haraldson, 1978; Ronse Decraene and Akeroyd, 1988; Ronse Decraene et al., 2000; Lamb Frye and Kron, 2003).

Persicaria are divided into four to five sections based on the presence of an exerted style and prickles, the type of inflorescence, and the degree of tepal connation (sections *Echinocaulon*, *Cephalophilon*, *Persicaria*, *Tovara*, and *Rubrivena*) (Freeman and Reveal, 2005). *P.* sect.

Cephalophilon (Meisn.) H. Gross, consisting of 19 taxa (12 species 7 varieties) of herbaceous plants, is distinguished from other sections within *Persicaria* by a petiole usually winged, stem without prickles and inflorescence capitate (Meisner, 1832; Li et al., 2003; Freeman and Reveal, 2005). These morphological features have supported this section since it was initially suggested as a single section within *Polygonum* by Meisner (1832) (Haraldson, 1978; Li et al., 1998, 2003; Freeman and Reveal, 2005). Moreover, *P.* sect. *Cephalophilon* is also a monophyletic group based on highly supported values according to recent molecular studies (Kim and Donoghue, 2008; Galasso et al., 2009; Schuster et al., 2015). *P.* sect. *Cephalophilon* is sister to the rest of sections in *Persicaria*, and the *Persicaria* which include section *Cephalophilon* altogether is sister to AKB clade (*Aconogonon* – *Koenigia* – *Bistorta* clade); thus, a detailed study of *P.* sect. *Cephalophilon* would enhance our understanding of the relationships among these taxa.

*Author for correspondence: sphong@khu.ac.kr

Nonetheless, the sectional delimitation and infrasectional classification of *P. sect. Cephalophilon* have remained unclear. In the earliest treatments, some taxa (*P. capitata* (Buch.-Ham. ex D. Don) H. Gross, *P. microcephala* (D. Don) H. Gross and *P. runcinata* (Buch.-Ham. ex D. Don) H. Gross) belonged to *Polygonum* sect. *Polygonum* (Don, 1825), and *P. sect. Cephalophilon* was treated as a genus *Cephalophilon* (Meisn.) Spach, *Ampelygonum* Lindl. or *Truellum* Houtt. (Lindley, 1838; Spach, 1841; Sojak, 1974). In addition, some taxa of *Koenigia* belonged to *P. sect. Cephalophilon* due to the floral characters (e.g., inflorescence, tepal, and style) (Li et al., 1998, 2003), although various morphological characters have indicated that these taxa (*P. sect. Cephalophilon* and *Koenigia*) were separated (Haraldson, 1978; Hedberg, 1997; Ronse Decraene et al., 2000). Meisner (1832) suggested two subsections, *Didymocephalon* and *Corymbocephalon*, to be included in the *Polygonum* sect. *Cephalophilon*, but more detailed research to evaluate the infrasectional classification has not been conducted. Subsequently, Zhu et al. (2007) also suggested new series (*Palmatifolia*, *Perennes*, *Criopolitana*, and *Annu*) within *Polygonum* sect. *Cephalophilon* based on the habit and stomata type on the abaxial side of the leaf. It is noteworthy that this suggestion still includes some *Koenigia* taxa. Therefore, more detailed study of *Persicaria* sect. *Cephalophilon* is necessary to re-evaluate the suggested infrasectional classification.

Fruit and seed morphology have provided useful characters to identify in the flowering plants, and especially, the surface sculpturing and ornamentation have been considered to have systematic importance (Roth 1977; Olowokudejo, 1985; Barthlott, 1990; Zhang et al., 2005). Achene morphologies have been studied by many researchers within Polygonaceae Juss. (Martin, 1954; Oh and Hong, 1999; Ayodele and Zhou, 2010; Ghimire et al., 2016; Kanwal et al., 2016), and the achene micromorphology and anatomy have already been described as useful taxonomic features in certain sectional and generic delimitations (Ronse Decraene et al., 2000). Although the detailed achene morphology of *P. sect. Cephalophilon* has been studied (e.g., *P. capitatum*, *P. chinensis*, *P. nepalensis* or *P. runcinata*) (Ronse Decraene et al., 2000; Kantachot et al., 2010; Ghimire et al., 2016; Kanwal et al., 2016), the papers were limited to only certain taxa. In addition, a comprehensive achene morphological study of *P. sect. Cephalophilon* is still lacking despite the character has been used in many systematic studies.

Thus, the present study aims (1) to document and illustrate the achene morphology of *P. sect. Cephalophilon* and (2) to evaluate the systematic values of the obtained achene characteristics of *P. sect. Cephalophilon*.

Materials and Methods

The achene morphologies of 17 taxa of *P. sect. Cephalophilon* except two taxa (*P. criopolitana* and *P. umbrosa*) and related taxa whose positions are unclear between this section and *Koenigia* (Table 1, Appendix 1) were investigated. The achenes were collected from herbarium specimens of K, KHUS, KUN, PE, and WU (Thiers, 2018).

Fully matured fruits were selected and observed under a stereomicroscope (SteREO Discovery.V8, Carl Zeiss Microscopy GmbH, Jena, Germany). To observe the detailed sculpturing patterns of achenes, dry fruits were rehydrated overnight in the Agepon wetting agent (Agepon:DW = 1:200) and fresh fruits were fixed in FAA (formaldehyde: acetic acid: alcohol) for 24 h and stored in 70% ethanol. The prepared achenes were dehydrated through an ethanol series (in 50, 70, 90, and 95% ethanol for 10 min each and in absolute ethanol for about 20 min) after which the absolute acetone was replaced with carbon dioxide to reach the critical point drying (SPI-13200J-AB, SPI Supplies, West Chester, PA, USA). All samples for the scanning electron microscopy (SEM) analysis were coated with platinum using an ion-sputtering device (E-1045, Hitachi, Tokyo, Japan). The samples were examined under a field emission SEM (FE-SEM, S-4700, Hitachi) device operating at 10 kV with a working distance of 10–13 mm. For the achene length and width 5–10 samples per specimen of each taxon were measured using software (Macnification 2.0 version, Orbicule, Leuven, Belgium). To confirm the consistency of morphological characteristics, we compared at least two specimens for each taxon when possible. Only one achene each of *P. chinensis* var. *paradoxa* 1, *P. glacialis* var. *glacialis* 3, *P. microcephala* var. *microcephala*, *P. strindbergii* was included for the comparison between taxa. The terminology for fruit characters was mainly adopted from Ronse Decraene et al. (2000).

Results and Discussion

External morphs: size, color and shape

The size of the achenes ranged from 0.93 to 3.55 mm in length and from 0.79 to 2.31 mm in width (Table 1). The achene size and shape were similar to those recorded in previous studies (Ronse Decraene et al., 2000; Kantachot and Chantaranothai, 2011; Ghimire et al., 2016; Kanwal et al., 2016). The largest achenes were found in *P. chinensis* var. *chinensis* (3.28–3.41 × 2.30–2.31 mm; investigated specimen, *Togash* 533), while the smallest achenes of *P. sect. Cephalophilon* were found in *P. humilis* (0.93–1.08 × 0.80–1.01 mm; investigated specimen, *Peng et al.* 9347). Among

the studied taxa, the *P. chinensis* group has the largest achene in the section ($2.34\text{--}3.55 \times 1.77\text{--}2.31$ mm), and relatively large achenes are also observed in *P. microcephala* var. *sphaerocephala* and *P. strindbergii*. The sizes of the *Koenigia*

taxa were $1.10\text{--}1.84 \times 0.79\text{--}1.02$ mm (Table 1). The *Koenigia* achenes are smaller than those of *P. sect. Cephalophilon*, while the almost taxa within the section have similar achenes size except for the smallest or the largest achenes.

Table 1. Details of achene characteristics in *Persicaria* sect. *Cephalophilon* and related taxa within the *Koenigia*.

Taxa	Length (mm)	Width (mm)	Outline	Shape	Beak	Stipitate	Glossy	ST	Figure
<i>P. capitata</i> 1	1.99–2.13	1.20–1.32	nov	tri	+	+	+	I	1A, 2A
<i>P. capitata</i> 2	1.54–1.60	0.96–0.98	nov	tri	+	+	+	I	-
<i>P. capitata</i> 3	1.72–1.78	0.88–1.10	nov	tri	+	+	+	I	-
<i>P. capitata</i> 4	1.64–1.78	0.91–0.97	nov	tri	+	+	+	I	-
<i>P. chinensis</i> var. <i>chinensis</i> 1	3.28–3.41	2.30–2.31	ovo	tri	+	+	-	II-2	1B, 2B
<i>P. chinensis</i> var. <i>chinensis</i> 2	2.57–2.70	1.99–2.17	bov	tri	+	+	-	II-2	-
<i>P. chinensis</i> var. <i>hispida</i> 1	2.72–2.73	1.95–2.01	ovo	tri	+	+	-	II-2	-
<i>P. chinensis</i> var. <i>hispida</i> 2	2.34–2.73	1.77–1.92	ovo	tri	+	+	-	II-2	1C
<i>P. chinensis</i> var. <i>ovalifolia</i>	2.46–2.86	1.78–2.02	ovo	tri	+	+	-	II-2	1D
<i>P. chinensis</i> var. <i>paradoxa</i> 1	2.42	1.87	bov	tri	+	+	-	II-2	-
<i>P. chinensis</i> var. <i>paradoxa</i> 2	2.89–3.55	2.08–2.18	ovo	tri	+	+	-	II-2	1E, 2C
<i>P. glacialis</i> var. <i>glacialis</i> 1	1.30–1.31	0.96–0.97	ovo	tri	+	+	-	II-1	1F
<i>P. glacialis</i> var. <i>glacialis</i> 2	1.59–1.68	1.25–1.32	bov	tri	+	+	-	II-1	-
<i>P. glacialis</i> var. <i>glacialis</i> 3	1.27	0.85	nov	tri	+	+	-	II-1	-
<i>P. glacialis</i> var. <i>przewalskii</i> 1	1.77–1.86	1.55–1.83	bov	tri	+	-/+	-	II-1	1G, 2D
<i>P. glacialis</i> var. <i>przewalskii</i> 2	1.82–2.02	1.61–2.00	bov	tri	+	+	-	II-1	-
<i>P. humilis</i> 1	1.03–1.15	0.87–0.97	bov	bic	+	+	-	IV	1H, 2E
<i>P. humilis</i> 2	1.13–1.15	1.03–1.04	bov	bic	+	+	-	IV	-
<i>P. humilis</i> 3	0.93–1.08	0.80–1.01	bov	bic	+	+	-	IV	-
<i>P. microcephala</i> var. <i>microcephala</i>	1.81	1.13	nov	tri	+	+	-	II-2	1I, 2F
<i>P. microcephala</i> var. <i>sphaerocephala</i>	3.15–3.28	2.25–2.27	ovo	tri	+	+	-	II-2	-
<i>P. microcephala</i> var. <i>wallichii</i>	2.07–2.10	1.23–1.30	nov	tri	+	+	-	II-2	1J
<i>P. nepalensis</i> 1	2.11–2.33	1.86–2.04	bov	bic	+	+	-	II-1, IV	1K, 2G
<i>P. nepalensis</i> 2	1.91–1.95	1.50–1.58	bov	bic	+	+	-	II-1	-
<i>P. nepalensis</i> 3	2.23–2.27	1.80–1.82	bov	bic	+	+	-	II-1	-
<i>P. palmata</i>	2.17–2.71	1.50–1.88	bov	tri	+	+	-	II-1	1L, 2H
<i>P. runcinata</i> var. <i>runcinata</i>	2.47–2.50	1.22–1.38	nov	tri	+	+	+	III	1M, 2I
<i>P. runcinata</i> var. <i>sinensis</i>	1.80–1.97	1.54–1.59	bov	tri	-	+	+	I, III	1N, 2J
<i>P. sinuata</i>	1.32–1.97	1.01–1.37	bov	tri	+	+	+	I, III	1O
<i>P. strindbergii</i>	2.79	1.78	ovo	tri	+	+	-	IV	1P
<i>K. delicatula</i>	1.62–1.75	0.79–0.96	ell	tri	-	-	-	I	1Q, 2K
<i>K. nepalensis</i> 1	1.33–1.84	0.80–1.02	ell	tri	-	-/+	-	I	-
<i>K. nepalensis</i> 2	1.35–1.67	0.83–1.01	ell	tri	-	-	-	I	1R
<i>K. pilosa</i>	1.31–1.45	0.80–1.01	bov	tri	-	+	-	I	1S
<i>K. islandica</i>	1.10–1.33	0.84–1.01	ovo	bic	-	-	-	I	1T, 2L

ST, surface sculpture; bov, broadly ovoid; bic, biconvex; ell, elliptic; nov, narrowly ovoid; ovo, ovoid; tri, trigonous; +, present; -, absent.

Fig. 1. Scanning electron microscopy micrographs of the achenes of section *Cephalophilon* and taxa related to *Koenigia*. **A.** *Persicaria capitata* (Expedition to Sichuan 268). **B.** *P. chinensis* var. *chinensis* (Togash 533). **C.** *P. chinensis* var. *hispida* (Zhou et al. 017). **D.** *P. chinensis* var. *ovalifolia* (Iwatsuki et al. S1493). **E.** *P. chinensis* var. *paradoxa* (Li et al. 1192). **F.** *P. glacialis* var. *glacialis* (Wang 69071). **G.** *P. glacialis* var. *przewalskii* (Xiao 738). **H.** *P. humilis* (Gamble s.n.). **I.** *P. microcephala* var. *microcephala* (Grierson and Long 3874). **J.** *P. microcephala* var. *wallichii* (Qinghai-Xizang Expedition 6676). **K.** *P. nepalensis* (Kong 7). **L.** *P. palmata* (Wang 79687). **M.** *P. runcinata* var. *runcinata* (Johns et al. 9642). **N.** *P. runcinata* var. *sinensis* (Li and Huang 2849). **O.** *P. sinuata* (Strachey and Winterbottom 40). **P.** *P. strindbergii* (Henry 10499A). **Q.** *K. delicatula* (Wang 68561). **R.** *K. nepalensis* (Sinclair and Long 5041). **S.** *K. pilosa* (Zhao 259). **T.** *K. islandica* (no detailed collect information). BK, beak; ST, stipitate.

The achenes in this study are brown to black, and noticeable colors such as green or yellow were not found in the present study. In *P.* sect. *Cephalophilon*, the achenes of most taxa were partially enclosed by the perianth, while *P. chinensis* has an accrescent perianth, such as a perianth being berry-like in appearance (Li et al., 2003; Ghimire et al., 2016). This perianth

makes it easy to distinguish *P. chinensis* from the other taxa of *P.* sect. *Cephalophilon*. Most taxa had dull achenes, while four taxa had glossy achenes (*P. capitata*, *P. runcinata* var. *runcinata*, *P. runcinata* var. *sinensis*, and *P. sinuata*). Thus, the presence of glossiness with other achene characters can be helpful to identify certain taxa.

The outlines of achenes were found to be elliptic to broadly ovoid (Fig. 1), and the most taxa have ovoid achenes. The taxa of *P. sect. Cephalophilon* had narrowly ovoid to broadly ovoid achenes (Fig. 1A–P, S, T), while elliptic achenes were found in *Koenigia* (Fig. 1Q–R). Elliptic achenes with both ends narrow are observed in *K. delicatula* and *K. nepalensis*. The achene shapes in the studied taxa were trigonous or biconvex (Fig. 1, Table 1). Most taxa had trigonous achenes, while biconvex achenes were observed in *P. humilis*, *P. nepalensis*, and *K. islandica*. These taxa, which had biconvex achenes, are clearly distinguished from other taxa. The achene dimorphism in the shape is often found in many taxa in Polygonaceae, as well as in *Polygonum* s.l. (Yurtseva, 2001; Li et al., 2003; Freeman and Reveal, 2005; Kantachot and Chantaranothai, 2011). In *P. sect. Cephalophilon*, *P. glacialis* var. *przewalskii* is known to have heteromorphic achenes (e.g., biconvex or trigonous) (Li et al., 2003); however, the achene shapes were rather consistent for the taxa in this study. Therefore, the achene shape can be used when setting limits on certain taxa.

The existence of a beak, which is positioned at style base, was also an investigated character. All taxa of *P. sect. Cephalophilon* had a beak structure on the apex (Fig. 1A–M, O, P), except for *P. runcinata* var. *sinensis* (Fig. 1N), while the *Koenigia* taxa had achenes without a beak (Fig. 1Q–T). A stipitate was also observed in most of the taxa studied here, except some *Koenigia* taxa (Table 1).

Achene micromorphology: surface sculpturing

Four types of achene surfaces were recognized: smooth (Type I), tubercles (type II), small pitted (Type III), and irregularly ridged (Type IV), and the tubercles were divided into two subtypes according to the grouping and distribution pattern (Fig. 2, Table 1). The surface sculpture type is consistent with each taxa except *P. nepalensis* (Type II-1 and IV), *P. runcinata* var. *sinensis* (Type I and III), and *P. sinuata* (Type I and III) (Table 1).

1. Type I

The whole fruit surface is smooth (Fig. 2A, K, L). The surface is uneven and anticlinal cell walls were observed; however, characterized structures such as tubercles were not observed: *P. capitata*, *P. microcephala* var. *microcephala*, *P. runcinata* var. *sinensis*, *P. sinuata*, *K. delicatula*, *K. nepalensis*, *K. pilosa*, and *K. islandica*.

2. Type II

Tubercles are distributed on the achene surface. This type is divided into two subtypes based on the grouping and distributed pattern.

2a. Type II-1

Tubercles are distributed along the anticlinal wall singly (discontinued), and the anticlinal wall forms a complete reticulate (Fig. 2D, G, H): *P. glacialis* var. *glacialis*, *P. glacialis* var. *przewalskii*, *P. nepalensis*, and *P. palmata*.

2b. Type II-2

Numerous tubercles are distributed along the reticulated anticlinal wall. Connected or fused tubercles are also observed, and single tubercles are occasionally observed on the lumen surface (Fig. 2B, C, F): *P. chinensis* var. *chinensis*, *P. chinensis* var. *hispidum*, *P. chinensis* var. *ovalifolia*, *P. chinensis* var. *paradoxa*, *P. microcephala* var. *microcephala*, *P. microcephala* var. *sphaerocephala*, and *P. microcephala* var. *wallichii*.

3. Type III

The achene surface with small pits is laying radially around the cell (Fig. 2I, J): *P. runcinata* var. *runcinata*, *P. runcinata* var. *sinensis*, and *P. sinuata*.

4. Type IV

Irregular ridges are laying on anticlinal cell walls, creating a reticulate pattern. This reticulate pattern is similar to that of Type II, however, tubercles were not observed on the ridge (Fig. 2E): *P. humilis*, *P. nepalensis* (partial), and *P. strindbergii*.

The anticlinal cell type on achene surface of *P. sect. Cephalophilon* are either undulate or straight-curved (Fig. 2). The anticlinal cell wall was difficult to observe on a Type I surface (Fig. 2A); however, *Koenigia* taxa, which have Type I surfaces, show easily distinguishable anticlinal cell walls (Fig. 2K, L, Table 1). The anticlinal cell walls of *Koenigia* are puzzle-like arrangement of undulate anticlinal walls (Fig. 2K, L, Table 1), but *K. islandica* has relatively weak undulated anticlinal cell walls (Fig. 2L). The remarkable anticlinal walls of achene surface with the absence of a beak is also help to clarify *Koenigia* taxa from *P. sect. Cephalophilon*. The *Koenigia* has been considered related taxa with *Aconogonon*, which was treated as a section of *Koenigia*, based on molecular phylogeny (Schuster et al., 2015). The *Aconogonon* is known to have ovate or ellipsoid achenes with smooth surface, and moreover, beak and stipitate are also found (Hong, 1992). Thus, achenes outline and surface sculpture of these two group are similar, but absence of beak is distinguished *Koenigia* taxa from *Aconogonon*.

Most of taxa in present study have Type II surface (11 taxa) (Fig. 2B–D, F–H, Table 1), and Type II-2 is more abundant

Fig. 2. The surface of the achene of section *Cephalophilon* and taxa related to *Koenigia*. **A.** *Persicaria capitata* (Type I; Expedition to Sichuan 268). **B.** *P. chinensis* var. *chinensis* (Type II-2; Togash 533). **C.** *P. chinensis* var. *paradoxa* (Type II-2; Li et al. 1192). **D.** *P. glacialis* var. *przewalskii* (Type II-1; Xiao 738). **E.** *P. humilis* (Type IV; Gamble s.n.). **F.** *P. microcephala* var. *microcephala* (Type II-2; Grierson and Long 3874). **G.** *P. nepalensis* (Type II-1; Kong 7). **H.** *P. palmata* (Type II-1; Wang 79687). **I.** *P. runcinata* var. *runcinata* (Type III; Johns et al. 9642). **J.** *P. runcinata* var. *sinensis* (Type III; Li and Huang 2849). **K.** *K. delicatula* (Type I; Wang 68561). **L.** *K. islandica* (Type I; no detailed collect information).

among taxa. Type II-1 is observed in four taxa, *P. glacialis* var. *glacialis*, *P. glacialis* var. *przewalskii*, *P. nepalensis*, and *P. palmata*. Among these taxa, *P. nepalensis* could be distinguished from other taxa based on biconvex and Type II-1 surfaces achene although part of surface has Type IV. The

existence of tubercles in achene micromorphology is considered to be an important feature. The functions of tubercles are not clear, but a protruding structure, such as tubercles or verrucae, may be related to the habitat environment or the dispersal mechanisms (Oh and Hong, 1999; Yurtseva,

2001; Kantachot and Chantaranothai, 2011). For example, Oh and Hong (1999) suggested that an achene with tubercles had a more developed morph than a smooth achene; thus, an achene with tubercles is motivating modifications to reach a better fit with the possibilities of myrmecochory or zoochory. On the other hand, this structure is generally found on the achene surfaces of plants in arid habitats and thus may serve to absorb water for germination and/or help to move water to the inside of the cells (Yurtseva, 2001; Kantachot and Chantaranothai, 2011). However, the *P. sect. Cephalophilon* taxa have similar habitat environments; moreover, this feature is consistent in each species (e.g., mountain slopes, grassy slopes, shaded places, thickets in valleys) (Li et al., 2003; Freeman and Reveal, 2005).

Types III and IV represent useful features by which to classify certain species of the *P. sect. Cephalophilon*. The small pits of Type III appeared to be scattered throughout the surface, and this type is observed in the intra-species taxa of *P. runcinata* and *P. sinuata* in the present study (Table 1). The *P. sinuata* has been considered to be close relationship with *P. runcinata* based on its morphology (Babington, 1838), and surface micromorphology of achene is supported this suggestion. However, these two taxa were belonged to different series (e.g., series *Annui* and *Perennes*) according to infrasectional classification proposed by Zhu et al. (2007), thus more study is needed to clarify the taxonomic relationship between two taxa.

Type IV is observed in *P. humilis*, *P. strindbergii*, and a small portion of *P. nepalensis*. Among these taxa, *P. humilis* is easily distinguished from the others based on achene size and shape with surface sculpturing. Though the *P. humilis* and *P. nepalensis* have similar achene morph (Fig. 1H, K), there are differences of size and micromorphology. These two taxa are known to have similar gross morphologies as well as achene morphs, indicating that a detailed phylogenetic study of these two taxa is needed.

In conclusion, a systematic evaluation of the achene morphology of *P. sect. Cephalophilon* revealed that a comprehensive approach of studied characters, such as the size, shape, and sculpturing type, is useful for identifying certain taxa at the species level. Among the characters studied here, the sculpturing type is important to identify certain taxa of *P. sect. Cephalophilon*. Moreover, the achene size, existence of a beak, and surface sculpturing serve to separate *Koenigia* taxa from *P. sect. Cephalophilon*. However, achene characters are considered insufficient if used to clarify its taxonomic delimitation at the sectional or infrasectional level as suggested in early studies because the achene characters are recurring

patterns within the *Persicaria*. But the achene morphological data contribute to our understanding of *P. sect. Cephalophilon*. Further studies are necessary to analyze the evolutionary tendency of the achene morphology in comparison with the molecular phylogeny within the *Persicaria*, and to investigate potential correlations with ecological factors such as dispersal mechanisms or habitats.

Acknowledgments

We are grateful to our colleagues at Kyung Hee University and to the directors of the herbaria of K, KHUS, KUN, PE, and WU. We also thank the anonymous reviewers for their valuable comments and suggestions, which substantially improved our manuscript. This study is part of the Ph.D. dissertation of M.-J. Kong. The work was supported by grants from the Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Education, Sciences and Technology [Grant numbers NRF-2015R1D1A1A01058462] to S.-P. Hong.

Conflict of Interest

Authors declare that there is no conflict of interest.

Literature Cited

- Ayodele, A. E. and Z.-K. Zhou. 2010. Scanning electron microscopy of fruits in the West African Polygonaceae. *Journal of Systematics and Evolution* 48: 336–343.
- Babington, C. C. 1838. IV. Descriptions of those species of *Polygonum* and *Fagopyrum* which are contained in the Indian Herbarium of J. Forbes Royle, Esq., F.L.S., &c., late Superintendent of the H.E.I. Botanical Garden at Saharunpore, and now Professor of Materia Medica in King's College, London. *Transactions of the Linnean Society of London* 18: 93–119.
- Barthlott, W. 1990. Scanning electron microscopy of the epidermal surface in plants. In *Scanning Electron Microscopy in Taxonomy and Functional Morphology*. Claugher, D. (ed.), Clarendon, Oxford. Pp. 64–94.
- Don, D. 1825. *Prodromus Florae Nepalensis*. J. Gale, Londini, 256 pp.
- Freeman, C. C. and J. L. Reveal. 2005. Polygonaceae. In *Flora of North America*. Vol. 5. *Flora of North America Editorial Committee* (ed.), Oxford University Press, New York. Pp. 479–601.
- Galasso, G., E. Banfi, F. De Mattia, F. Grassi, S. Sgorbati and M.

- Labra. 2009. Molecular phylogeny of *Polygonum* L. s.l. (Polygonoideae, Polygonaceae), focusing on European taxa: preliminary results and systematic considerations based on *rbcL* plastidial sequence data. *Atti della Società italiana di scienze naturali e del Museo civico di storia naturale di Milano* 150: 113–148.
- Ghimire, B., G. E. Choi, M. J. Jeong, H. Lee, K. M. Lee, S. W. Son, C. H. Lee and G. U. Suh. 2016. Fruit morphology and anatomy of *Persicaria chinensis* (Polygonaceae). *Journal of Japanese Botany* 91: 218–225.
- Haraldson, K. 1978. Anatomy and taxonomy in Polygonaceae subfam. Polygonoideae Meissn. emend. Jaretsky. *Symbolae Botanicae Upsaliensis* 22: 1–95.
- Hedberg, O. 1997. The genus *Koenigia* L. emend. Hedberg (Polygonaceae). *Botanical Journal of the Linnean Society* 124: 295–330.
- Hong, S.-P. 1992. Taxonomy of the genus *Aconogonon* (Polygonaceae) in Himalaya and adjacent regions. *Symbolae Botanicae Upsalienses*. Vol. 30. Uppsala University, Uppsala, 118 pp.
- Kantachot, C. and P. Chantaranothai. 2011. Achene morphology of *Polygonum* s.l. (Polygonaceae) in Thailand. *Tropical Natural History* 11: 21–28.
- Kantachot, C., P. Chantaranothai and D. A. Simpson. 2010. A synopsis of the genus *Persicaria* (Polygonaceae) in Thailand. *Thai Forest Bulletin (Botany)* 38: 128–149.
- Kanwal, D., R. Abid and M. Qaiser. 2016. The seed atlas of Pakistan – XIV. Polygonaceae. *Pakistan Journal of Botany* 48: 1833–1848.
- Kim, M. H., M. Kwak, J. Song, S. J. Lee, M. J. Yoo and C. W. Park. 2001. Morphology of hairs, flowers, and achenes of *Polygonum* section *Echinocaulon* Meisn. (Polygonaceae). *Korean Journal of Plant Taxonomy* 31: 143–160. (in Korean)
- Kim, S. T. and M. J. Donoghue. 2008. Molecular phylogeny of *Persicaria* (Persicarieae, Polygonaceae). *Systematic Botany* 33: 77–86.
- Lamb Frye, A. S. and K. A. Kron. 2003. *rbcL* Phylogeny and character evolution in Polygonaceae. *Systematic Botany* 28: 326–332.
- Li, A. J., T. Kao, Z. Mao and Y. Liu. 1998. Polygonaceae. In *Flora Republicae Popularis Sinicae*. Vol. 25(1). Li, A. J. (ed.), Science Press, Beijing. Pp. 1–209.
- Li, A. J., B. Bao, A. E. Grabovskaya-Borodina, S. P. Hong, J. McNeill, S. L. Mosyakin, H. Ohba and C. W. Park. 2003. Polygonaceae. In *Flora of China*. Vol. 5. Ulmaceae-Basellaceae. Wu, Z. Y., P. H. Raven and D. Y. Hong (eds.), Science Press, Beijing and Missouri Botanical Garden Press, St. Louis, MO. Pp. 277–350.
- Lindley, J. 1838. Edwards's Botanical Register. James Ridgway, London, Pp. 62–63.
- Martin, A. C. 1954. Identifying *Polygonum* seeds. *The Journal of Wildlife Management* 18: 514–520.
- Meisner, C. F. 1832. Synopsis Polygonearum. In *Plantae Asiaticae Rariores*. Vol. 3. Wallich, N. (ed.), Treuttel and Würtz, London. Pp. 53–65.
- Oh, I. C. and S. P. Hong. 1999. A taxonomic study of achene microstructure in the genera *Polygonum* L. s. str. and *Polygonella* Michx. (Polygoneae-Polygonaceae). *Korean Journal of Plant Taxonomy* 29: 123–134. (in Korean)
- Olowokudejo, J. D. 1985. Scanning electron microscopy of fruits in the genus *Biscutella* L. (Cruciferae). *Phytomorphology* 35: 273–286.
- Ronse Decraene, L. P. and J. R. Akeroyd. 1988. Generic limits in *Polygonum* and related genera (Polygonaceae) on the basis of flora characters. *Botanical Journal of the Linnean Society* 98: 321–371.
- Ronse Decraene, L. P., S. P. Hong and E. Smets. 2000. Systematic significance of fruit morphology and anatomy in tribes Persicarieae and Polygoneae (Polygonaceae). *Botanical Journal of the Linnean Society* 134: 301–337.
- Roth, I. 1977. Fruits of Angiosperms. Gebrüder Borntraeger, Berlin, 675 pp.
- Schuster, T. M., J. L. Reveal, M. J. Bayly and K. A. Kron. 2015. An updated molecular phylogeny of Polygonoideae (Polygonaceae): relationships of *Oxygonum*, *Pteroxygonum*, and *Rumex*, and a new circumscription of *Koenigia*. *Taxon* 64: 1188–1208.
- Soják, J. 1974. Observations on the genus *Truelleum* Houtt. (Polygonaceae). *Preslia* 46: 139–156.
- Spach, M. E. 1841. Histoire Naturelle des Vegetaux-Phanerogames. Librairie Encyclopedique De Port, Paris.
- Thiers, B. 2018. [continuously updated] Index Herbariorum: a global directory of public herbaria and associated staff. Retrieved Sep. 1, 2018, available from <http://sweetgum.nybg.org/ih/>.
- Yurtseva, O. V. 2001. Ultrasculpture of achene surface in *Polygonum* section *Polygonum* (Polygonaceae) in Russia. *Nordic Journal of Botany* 21: 513–528.
- Zhang, Z.-Y., D.-Z. Yang, A.-M. Lu and S. Knapp. 2005. Seed morphology of the tribe Hyoscyameae (Solanaceae). *Taxon* 54: 71–83.
- Zhu, L.-T., F.-J. Lu, Y.-T. Hou and F.-Z. Li. 2007. Micro-morphology of leaf epidermis and its taxonomic significance of *Polygonum* section *Cephalophilon* in China. *Journal of Wuhan Botanical Research* 25: 136–142.

Appendix 1. Specimens examined for the comparative study of achene morphology of *Persicaria* sect. *Cephalophilon* and related taxa.

Taxa	Specimen information
<i>Persicaria capitata</i> (Buch.-Ham. ex D. Don) H. Gross 1	China Sichuan, Sep 1988, <i>Expedition to Sichuan</i> 268 (K)
<i>P. capitata</i> (Buch.-Ham. ex D. Don) H. Gross 2	Bhutan Tashigang, Jun 1979, <i>Grierson and Long</i> 2061 (K)
<i>P. capitata</i> (Buch.-Ham. ex D. Don) H. Gross 3	Bhutan Tschilingor, Jun 1979, <i>Grierson and Long</i> 2276 (K)
<i>P. capitata</i> (Buch.-Ham. ex D. Don) H. Gross 4	New Zealand Waitemata Country, Sep 1946, <i>Bangerter</i> 5316 (K)
<i>P. chinensis</i> (L.) H. Gross var. <i>chinensis</i> 1	Japan, –, <i>Togash</i> 533 (WU)
<i>P. chinensis</i> (L.) H. Gross var. <i>chinensis</i> 2	Himalaya, Sep 1977, <i>Schilling s.n.</i> (K)
<i>P. chinensis</i> var. <i>hispida</i> (Hook.f.) Kantachot 1	Thailand Phukrandung, Dec 1971, <i>van Beuseko et al.</i> 4512 (K)
<i>P. chinensis</i> var. <i>hispida</i> (Hook. f.) Kantachot 2	China Yunnan, Oct 1999, <i>Zhou et al.</i> 017 (PE)
<i>P. chinensis</i> var. <i>ovalifolia</i> (Meisn.) Hara	Indonesia Sumatra, Sep 1971, <i>Iwatsuki et al.</i> S1493 (K)
<i>P. chinensis</i> var. <i>paradoxa</i> (H. Lév.) B. Li 1	China Yunnan, Oct 1940, <i>Qin</i> 25317 (PE)
<i>P. chinensis</i> var. <i>paradoxa</i> (H. Lév.) B. Li 2	China Yunnan, Nov 2002, <i>Li et al.</i> 1192 (KUN)
<i>P. glacialis</i> (Meisn.) H. Hara var. <i>glacialis</i> 1	China Yunnan, Aug 1935, <i>Wang</i> 69071 (PE)
<i>P. glacialis</i> (Meisn.) H. Hara var. <i>glacialis</i> 2	China Sichuan, Aug 2005, <i>Boufford et al.</i> 34635 (KUN)
<i>P. glacialis</i> (Meisn.) H. Hara var. <i>glacialis</i> 3	–, <i>Handel-Mazzetti</i> 7087 (WU)
<i>P. glacialis</i> var. <i>przewalskii</i> (A. K. Skvortsov & Borodina) M. J. Kong and S. P. Hong (nom. ined.) 1	China Shaanxi, Sep 1958, <i>Xiao</i> 738 (PE)
<i>P. glacialis</i> var. <i>przewalskii</i> (A. K. Skvortsov & Borodina) M. J. Kong and S. P. Hong (nom. ined.) 2	China Yunnan, Sep 1986, <i>Sun and Qian</i> 715 (KUN)
<i>P. humilis</i> (Meisn.) H. Hara 1	–, Sep 1875, <i>Gamble s.n.</i> (K)
<i>P. humilis</i> (Meisn.) H. Hara 2	China Poshang, Sep 1938, <i>Yu</i> 18038 (KUN)
<i>P. humilis</i> (Meisn.) H. Hara 3	China Yunnan, Aug 2008, <i>Peng et al.</i> 9347 (KUN)
<i>P. microcephala</i> (D. Don) H. Gross var. <i>microcephala</i>	Bhutan Sarbhang, Mar 1982, <i>Grierson and Long</i> 3874 (K)
<i>P. microcephala</i> var. <i>sphaerocephala</i> (Wall. ex Meisn.) Hara	India Chennai, May 1889, <i>Gamble</i> 20727 (K)
<i>P. microcephala</i> var. <i>wallichii</i> (Meisn.) Hara	Tibet Autonomous Region, Jun 1975, <i>Qinghai-Xizang Expedition</i> 6676 (PE)
<i>P. nepalensis</i> (Meisn.) H. Gross 1	R.O.K. Gyeonggi-do, Sep 2011, <i>Kong</i> 7 (KHUS)
<i>P. nepalensis</i> (Meisn.) H. Gross 2	China Sichuan, Jul 1989, <i>Zhao</i> 186 (K)
<i>P. nepalensis</i> (Meisn.) H. Gross 3	Indonesia Java, Feb 1926, <i>Danser s.n.</i> (K)
<i>P. palmata</i> (Dunn) Yonekura & H. Ohashi	China Yunnan, Oct 1936, <i>Wang</i> 79687 (PE)
<i>P. runcinata</i> (Buch.-Ham. ex D. Don) H. Gross var. <i>runcinata</i>	Indonesia Papua, Aug 1998, <i>Johns et al.</i> 9642 (K)
<i>P. runcinata</i> var. <i>sinensis</i> (Hemsley) B. Li	China Yunnan, Aug 1995, <i>Li and Huang</i> 2849 (KUN)
<i>P. sinuata</i> (Royle ex Bab.) Rajbh. & R. Joshi	Indonesia Kumaon, –, <i>Strachey and Winterbottom</i> 40 (K)
<i>P. strindbergii</i> (J. Schust.) Galasso	China Yunnan, –, <i>Henry</i> 10499A (K)
<i>Koenigia delicatula</i> (Meisn.) H. Hara	China Yunnan, Aug 1935, <i>Wang</i> 68561 (PE)
<i>K. nepalensis</i> D. Don 1	Tibet Yatung, 1897, <i>Hobson s.n.</i> (K)
<i>K. nepalensis</i> D. Don 2	Bhutan, Sep 1984, <i>Sinclair and Long</i> 5041 (K)
<i>K. pilosa</i> Maxim.	China Sichuan, Jul 1989, <i>Zhao</i> 259 (K)
<i>K. islandica</i> L.	–, Sep 1876, – (K)

Herbarium acronyms are in accordance with Thiers (2018) [continuously updated].

–, no detailed collect information.