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## CYPSELA MORPHOLOGY OF THE GENUS *ANAPHALIS* DC. (GNAPHALIEAE-ASTERACEAE) FROM PAKISTAN

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### Abstract

Cypselae morphology of 17 taxa of the genus *Anaphalis* DC., was examined using light and scanning electron microscopy. On the basis of cypselae surface all the taxa are divided into two main groups and most of the species are delimited due to their distinct micromorphological characters of cypselae.

### Introduction

The genus *Anaphalis* DC., belongs to the tribe Gnaphalieae of the family Asteraceae and comprises 15 species in Pakistan (Qaiser & Abid, 2003). The cypselae micromorphological characters have played an important role of systematic significance in the family Astraceae (Kynclova, 1970; Merxmuller & Grau, 1977; Haque & Godward, 1984; Mateu & Guemes, 1993; Abid & Qaiser, 2002; Ritter & Miotlo, 2006; Abid & Qaiser, 2007a, 2007b; Abid & Zehra, 2007). Although in earlier reports on cypselae morphology of the family Astraceae from Pakistan only the tribes Inuleae and Plucheeae (Abid & Qaiser, 2002, 2007a,b; Abid & Zehra, 2007) have been studied. Cypselae characters in the genus *Anaphalis* have not received due attention (Anderberg, 1991; Qaiser & Abid, 2003). The present studies of cypselae morphology are carried out to strengthen the recognition of taxa in the genus *Anaphalis* from Pakistan.

### Materials and Methods

Seventeen taxa of the genus *Anaphalis* DC., were studied for cypselae characters under stereomicroscope (Nikon XN Model), compound microscope (Nikon Type 102) and scanning electron microscope (JSM-6380A). For scanning electron microscopy mature cypselae were directly mounted on metallic stub using double adhesive tape and coated with gold for a period of 6 minutes in sputtering chamber and observed under SEM.

The following characters were studied under light microscope.

**Cypselae:** Shape, colour, size, surface.

**Pappus:** Bristles shape, series, number, size, colour.

**Corpopodium:** Shape, position, diameter of carpopodium and diameter of foramen of carpopodium were observed under scanning electron microscope.

### Observations

**General cypselae characters of *Anaphalis*:** Cypselae oblong, or ellipsoid or oblong-ellipsoid, slightly angular or non angular, 0.5-1.5 x 0.25-1.0mm, colour varies from

yellowish brown to reddish brown or dark brown, non-ribbed, sparsely or densely papillate-clavate hairy. Pappus uniseriate, bristly, white or off white or golden, 8-18, 1.5-5.0mm long. Carpopodium broad disc like or narrow circular ring or slightly angular, without any interruption, subbasal or basal in position, 85-152 $\mu\text{m}$  in diameter. Foramen of carpopodium 58.5-100 $\mu\text{m}$  in diameter (Table 1; Figs. 1-3).

#### Key to the species

- 1 +Cypselas sparsely papillate-clavate hairy ..... 2  
-Cypselas densely papillate-clavate hairy ..... 4
- 2 +Cypselas reddish brown. Pappus bristles 16-20. Carpopodium broad circular disc like ..... 3  
-Cypselas dark brown. Pappus bristles 10-12. Carpopodium narrow circular ring like ..... *A. adnata, A. busua*
- 3 +Cypselas oblong-ellipsoid. Carpopodium basal in position, 150 $\mu\text{m}$  in diameter, foramen of carpopodium 90 $\mu\text{m}$  in diameter ..... *A. chitralensis*  
-Cypselas ellipsoid. Carpopodium subbasal in position, 130 $\mu\text{m}$  in diameter, foramen of carpopodium 68 $\mu\text{m}$  in diameter ..... *A. virgata*
- 4 +Cypselas and carpopodium slightly angular ..... 5  
-Cypselas and carpopodium non angular ..... 6
- 5 +Pappus bristles golden. Carpopodium 148 $\mu\text{m}$  in diameter ..... *A. boissieri*  
-Pappus bristles white. Carpopodium 106-111 $\mu\text{m}$  in diameter ..... *A. royleana* var. *royleana*, var. *cana*, var. *concolor*
- 6 +Carpodium broad circular disc like, 88 $\mu\text{m}$  in diameter ..... *A. patentifolia*  
-Carpodium narrow circular ring like 100 - 152 $\mu\text{m}$  in diameter ..... 7
- 7 +Pappus bristles 16-18. Carpopodium 152 $\mu\text{m}$  in diameter, foramen of carpopodium 100 $\mu\text{m}$  in diameter ..... *A. scapulosa*  
-Pappus bristles 8-12. Carpopodium 100-130 $\mu\text{m}$  in diameter, foramen of carpopodium 75-90 $\mu\text{m}$  in diameter ..... 8
- 8 +Cypselas yellowish brown ..... *A. stantonii*  
-Cypselas dark brown or reddish brown ..... 9
- 9 +Pappus bristles off white ..... *A. contorta*  
-Pappus bristles white ..... 10
- 10 +Cypselas dark brown ..... *A. margaritacea, A. triplinervis*  
-Cypselas reddish brown ..... 11
- 11 +Cypselas 0.5 x 0.25mm ..... *A. kashmiriana*  
-Cypselas 1.0 x 0.5mm ..... *A. nepalensis* var. *nepalensis*, var. *monocephala*

Table 1. Cypselae morphological features of the genus *Anaphalis*.

Name of species	Cypselae				Size (mm)
	Shape	Surface (hairs)	Colour		
<i>Anaphalis adnata</i>	Ellipsoid, non-angular	Sparingly papillose- Clavate	Dark brown		0.5 x 0.25
<i>A. boissieri</i>	Oblong, slightly angular	Densely papillose- Clavate	Yellowish brown		1-1.5 x 0.5
<i>A. busua</i>	Ellipsoid, non-angular	Sparingly papillose- Clavate	Dark brown		0.5 x 0.25
<i>A. chiraleensis</i>	Oblong-ellipsoid, non-angular	Sparingly papillose- Clavate	Reddish brown		1.5 x 0.5
<i>A. contorta</i>	Oblong, non-angular	Densely papillose- Clavate	Reddish brown		1.0 x 0.5
<i>A. kashmiriana</i>	Oblong, non-angular	Densely papillose- Clavate	Reddish brown		0.5 x 0.25
<i>A. margaritacea</i>	Oblong, non-angular	Densely papillose- Clavate	Dark brown		1.0 x 0.5
<i>A. nepalensis</i> var. <i>nepalensis</i>	Oblong, non-angular	Densely papillose- Clavate	Reddish brown		1.0 x 0.5
<i>A. nepalensis</i> var. <i>monocephala</i>	Oblong, non-angular	Densely papillose- Clavate	Reddish brown		1.0 x 0.5
<i>A. patentifolia</i>	Oblong, non-angular	Densely papillose- Clavate	Yellowish brown		0.75-1.0 x 0.5
<i>A. royleana</i> var. <i>royleana</i>	Oblong, slightly angular	Densely papillose- Clavate	Yellowish brown		1.0 x 0.5
<i>A. royleana</i> var. <i>cana</i>	Oblong, slightly-angular	Densely papillose- Clavate	Yellowish brown		1.0 x 0.5
<i>A. royleana</i> var. <i>concolor</i>	Oblong, slightly angular	Densely papillose- Clavate	Yellowish brown		1.0 x 0.5
<i>A. scapulosa</i>	Oblong, non angular	Densely papillose- Clavate	Dark brown		1.0 x 0.25
<i>A. staintonii</i>	Oblong, non -angular	Densely papillose- Clavate	Yellowish brown		1.0 x 0.5
<i>A. triplinervis</i>	Oblong, non-angular	Densely papillose- Clavate	Dark brown		1.0 x 0.5
<i>A. virgata</i>	Ellipsoid, non-angular	Sparingly papillose- Clavate	Reddish brown		1.5 x 0.5

Table 1. (Cont'd.).

Name of species	Bristles	Pappus		
		Number	Size (mm)	Colour
<i>Anaphalis adhatda</i>	Barbellate, basally with patent cilia	10-12	1.5	White
<i>A. boissieri</i>	Barbellate, basally with patent cilia	14-16	3-4	Golden
<i>A. busua</i>	Barbellate, basally with patent cilia	10-12	2.5-3.0	White
<i>A. chiraleensis</i>	Barbellate, basally with patent cilia	18-20	3.5-4.0	Off white
<i>A. contorta</i>	Barbellate, basally with patent cilia	10-12	3-4.0	Off white
<i>A. kashmiriana</i>	Barbellate, basally with patent cilia	8-10	3-4.0	White
<i>A. margaritacea</i>	Barbellate, basally with patent cilia	8-10	3.5-5.0	White
<i>A. nepalensis</i> var. <i>nepalensis</i>	Barbellate, basally with patent cilia	10-12	3-4	White
<i>A. nepalensis</i> var. <i>monocephala</i>	Barbellate, basally with patent cilia	10-12	3-4	White
<i>A. patenifolia</i>	Barbellate, basally with patent cilia	10-12	3-4	White
<i>A. royleana</i> var. <i>royleana</i>	Barbellate, basally with patent cilia	14-16	3-4	White
<i>A. royleana</i> var. <i>cana</i>	Barbellate, basally with patent cilia	10-12	3.0	White
<i>A. royleana</i> var. <i>concolor</i>	Barbellate, basally with patent cilia	10-12	3.0	White
<i>A. scapulosa</i>	Barbellate, basally with patent cilia	16-18	3.0	White
<i>A. stainionii</i>	Barbellate, basally with patent cilia	10-12	3-3.5	White
<i>A. triplinervis</i>	Barbellate, basally with patent cilia	10-12	3-3.5	White
<i>A. virgata</i>	Barbellate, basally with patent cilia	16-18	3-4	Off white

Table 1. (Cont'd.).

Name of species	Shape	Carpopodium		
		Position	Diameter of carpopodium ( $\mu\text{m}$ )	Diameter of foramen of carpopodium ( $\mu\text{m}$ )
<i>Anaphalis adnata</i>	Narrow circular ring without any interruption	Sub basal	97	63
<i>A. boissieri</i>	Slightly angular without any interruption	Sub basal	148	82
<i>A. busua</i>	Narrow circular ring without any interruption	Sub basal	85	58.5
<i>A. chitralensis</i>	Broad circular disc without any interruption	Basal	150	90
<i>A. contorta</i>	Narrow circular ring without any interruption	Sub basal	110	80
<i>A. kashmiriana</i>	Narrow circular ring without any interruption	Sub basal	100	80
<i>A. margaritacea</i>	Narrow circular ring without any interruption	Sub basal	130	78
<i>A. nepalensis</i> var. <i>nepalensis</i>	Narrow circular ring without any interruption	Sub basal	112	75
<i>A. nepalensis</i> var. <i>monocephala</i>	Narrow circular ring without any interruption	Sub basal	110	76
<i>A. patenifolia</i>	Broad circular disc without any interruption	Sub basal	88	60
<i>A. royleana</i> var. <i>royleana</i>	Slightly angular without any interruption	Sub basal	106	64
<i>A. royleana</i> var. <i>cana</i>	Slightly angular without any interruption	Sub basal	111	62
<i>A. royleana</i> var. <i>concolor</i>	Slightly angular without any interruption	Sub basal	110	63
<i>A. scapulosa</i>	Narrow circular ring without any interruption	Sub basal	152	100
<i>A. staintonii</i>	Narrow circular ring without any interruption	Sub basal	100	78
<i>A. triplinervis</i>	Narrow circular ring without any interruption	Sub basal	130	90
<i>A. virgata</i>	Broad circular disc without any interruption	Sub basal	130	68

Fig. 1. Scanning Electron micrographs. *Anaphalis adnata*: A, cypselae; B, surface; C, carpopodium. *A. boissieri*: D, cypselae; E, surface; F, carpopodium. *A. busua*: G, cypselae; H, surface; I, carpopodium. *A. chitralensis*: J, cypselae; K, surface; L, carpopodium. *A. contorta*: M, cypselae; N, surface; O, carpopodium (Scale bar: A, D, G= 100 $\mu$ m ; J = 200 $\mu$ m; M = 50 $\mu$ m; B,C,F, K, L, O = 20 $\mu$ m; E = 30 $\mu$ m; H, I, N = 10 $\mu$ m).

Fig. 2. Scanning Electron micrographs. *Anaphalis kashmiriana*: A, cypselae; B, surface; C, carpopodium. *A. margaritacea*: D, cypselae; E, surface; F, carpopodium. *A. nepalensis* var. *nepalensis*: G, cypselae; H, surface; I, carpopodium. *A. patentifolia*: J, cypselae; K, surface; L, carpopodium. *A. royleana* var. *royleana*: M, cypselae; N, surface; O, carpopodium (Scale bar: A, D, G, M = 100 $\mu$ m; J = 50 $\mu$ m; B, E, K, L, N = 10 $\mu$ m; C, F, H, I, O = 20 $\mu$ m).

Fig. 3. Scanning Electron micrographs. *Anaphalis royleana* var. *concolor*: A, cypsela; B, surface; C, carpodium. *A. scapulosa*: D, cypsela; E, surface; F, carpodium. *A. staintonii*: G, cypsela; H, surface; I, carpodium. *A. triplinervis*: J, cypsela; K, surface; L, carpodium. *A. virgata*: M, cypsela; N, surface; O, carpodium (Scale bar: A,G= 100 $\mu$ m; D,M = 200 $\mu$ m; J, N = 50 $\mu$ m; B, C, E, F, H, K, L, O = 20 $\mu$ m; I = 10 $\mu$ m).

### Results and Discussion

The genus *Anaphalis* DC., is distinguished amongst all of the genera of Gnaphalieae due to the presence of short clavate hairs on cypsela surface (Anderberg, 1991, Bremer, 1994; Qaiser & Abid, 2003). Presently it is observed that cypsela surface is papillate-clavate hairy rather than the clavate hairy only and secondly their density was not considered previously. On the basis of density of these hairs all the taxa of *Anaphalis* are divided into two groups i.e. cypselas sparsely or densely papillate-clavate hairy. Sparsely papillate-clavate cypselas are found in *A. adanata* Wall. ex DC., *A. busua* (Buch.-Ham. ex D.Don) DC., *A. chitralensis* Qaiser & R. Abid and *A. virgata* Thomson ex C.B.Clarke (Figs. 1B, H, K; 3N). Species like *A. adanata* and *A. busua* can be delimited by the presence of dark brown cypselas having 10-12 pappus bristles and narrow circular ring like carpodium (Fig. 1A, C, G, I). Furthermore both the species can not be separated due to similar cypsela characters while, in *A. chitralensis* and *A. virgata* cypselas are reddish brown with 16-20 pappus bristles and broad circular disc like carpodium (Figs. 1J,L;3M, O) and both the species remain distinct from each other due to differences in cypsela shape, position and diameter of carpodium. Taxa having densely papillate-clavate cypselas may be further divided into two groups one group with slightly angular cypselas and carpodium, including *A.boissieri* E. Georgiadou (Fig. 1D, F) and *A. royleana* DC. var. *royleana*, var. *cana* Hook. f. and var. *cancolor* Hook.f. (Figs. 2M, O; 3A, C). *A. boissieri* is further separated from *A. royleana* by the presence of golden pappus bristles and larger carpodium, while in *A. royleana* pappus bristles are white. Another group of densely papillate-clavate cypselas having non-angular cypselas and carpodium, from which *A. patentifolia* Rech.f. is distinguished by the presence of broad circular disc like carpodium (Fig. 2J, L). While in other species carpodium is narrow circular ring-like, amongst them *A. scapulosa* Boriss., remains distinct from rest of the species due to 16-18 pappus bristles and comparatively larger carpodium. Another species, *A. staintonii* E. Georgiadou (Fig. 3G, I) is distinct by having yellowish brown cypselas. While in *A. contorta* (D.Don) Hook.f., *A. kashmiriana* P.C. Pant and *A. nepalensis* (Spreng.) Hand.-Mazz. cypselas are reddish brown but *A. contorta* still remains distinct from both the species due to off white pappus bristles. *A. kashmiriana* and *A. nepalensis* have white pappus bristles. On the other hand the two remaining species *A. margaritacea* (L.) Benth. and *A. triplinervis* (Sims.) C.B. Clarke have dark brown cypselas and both the species could not be separated due to indistinct cypsela features. Therefore for most of the species of *Anaphalis* the micromorphological characters of cypsela are quite stable and characteristic and have systematic importance.

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