POLLEN MORPHOLOGY OF SOME *BLUMEA* SPECIES DISTRIBUTED IN MANDALAY AREA

Aye Aye Thin¹ & Soe Myint Aye²

Abstract

The pollen morphology of some *Blumea* species distributed in Mandalay Area was undertaken. The samples of 8 species belonging to 1 genus under 1Tribes were collected from January to June during the year 2018. The morphological characters of pollen grains of each species were also studied. The types of pollen grains were tricolpate, triporate, tricolporate and tetracolporate. Three species were tricolpate and triporate. Only one species were tricolporate and tetracolporate. The exine ornamentation in pollen was found to be all echinate. The shapes of pollen were spheroidal, prolate spheroidal and subprolate. Four species were spheroidal, two species were subprolate, only one species were medium and three spheroidal. The size of pollen were small or medium. Five species were medium and three species were small. An artificial key to the species were also constructed. The pollen photomicrographs of each species were presented by polar view and equatorial view.

Keywords: Blumea, Tricolpate, Triporate, Tricolporate, Tetracolporate, Exine, Echinate.

Introduction

Blumea DC. is one of the largest genera in the Tribe Inuleae which includes approximately 100 species worldwide. *Blumea* is primarily distributed in tropical Asia and Africa while its highest diversity is in tropical Asia (Peng *et al.* 2020). Seventeen species were recorded in Myanmar (Kress *et al.* 2003).

Palynology is the study of spores and pollen grains. Spores and pollen grains have a number of morphological and ultrastructural features. Pollen wall structure refers to the internal form of the pollen grain wall. Mature pollen walls almost always consists of two major layers; intine and exine. The exine is the outermost layer which is composed of primarily of cellulose and pectins. The exine is the hard, outermost desiccation-resistant wall layer that provides the major structural support for the cytoplasm (Simpson 2006). Two basic types of pollen grains are the porate and colpate. Furrows are elongate and boat shaped and ends are more or less sharp. Pollen grains that have one long furrow are called monocolpate. Tricolpate pollen grains have three long furrows (Yamada & Iwanmai 1988).

Pollen grains carry the male gametes of seed plants. Pollen biology has a strong impact in agriculture, biotechnology forestry and genetics (Meo 2009). There are many disciplines associated with plant taxonomy which are used by taxonomists to improve the identification, classification and systematic position of plant taxa. Among these disciplines palynology is one of the most significant tools used by modern taxonomist to identify and differentiate closely related taxa. The study of pollen has direct relevance in agriculture, horticulture, foresty, plant breeding and biotechnology (Zafar *et al.* 2007). Dinis and Pereira (2007) described pollen morphology of Inuleae Tribes are porilolongate, sculpture echinate. Pornpanggrueng (2016) stated that shape of *Blumea* species were prolate spheroidal, oblate spheroidal and subprolate, spine length 1.3-6µm long.

The aim and objectives of this research to investigate on type, shape, size and sculpture of pollen and to provide the valuable information of pollen morphology that will be useful in plant identification.

¹ Lecturer, Department of Botany, Monywa University

² Deputy Director General, Department of Higher Education, Ministry of Education

Materials and Methods

The specimens were collected in Mandalay Area from January to June during the year 2018. Identification of genera and species were carried out by referring to Dassanayake (1980), Backer (1965). Myanmar and English names were checked by referring Hundley and Chit Ko Ko (1989), Kress *et al.* (2003). All the fresh pollen were collected from the anther of mature flowers and stored in glass vial with glacial acetic acid.

The pollen samples were acetolysed by the method of Erdtman (1960). The samples in glass vial were put into a test tube then crushed with a glass rod. Acetolysis solution was mixed by 9 parts of glacial acetic acid and 1 part of concentrated sulphuric acid. The acid was dropped gently down the side of the tube. The 1cc of acetolysis mixture was poured into the test tube containing the pollen sample and stirred with a glass rod. The test tube was heated in a water bath 70°-80°C for 25-30 minutes. The test tube was allowed to cool, and the sample diluted with distilled water and centrifuged for 20-30 minutes at 3000 rpm. This was repeated twice decanting the water each time. Dilute glycerin solution was added to the residue then transferred and stored in air tight glass vial and labeled.

The mounted slides were observed under light microscope to study the pollen morphology. For each species more than 10 pollen grains were measured and recorded. The terminology used in the accordance with Erdtman (1971) and Hesse *et al.* (2009).

Results

Pollen morphology of 8 species of *Blumea* has been studied. The lists of collected species were as shown in Table 1.

Tribe	No	Scientific Name	Myanmar Name
Inuleae	1	Blumea barbata DC.	Ya nat
	2	Blumea hieraciifolia (D. Don) DC.	Unknown
	3	Blumeajun ghuhniana (Miq.) Boerl.	Unknown
	4	Blumea laciniata (Wall. ex Roxb.) DC.	Mmo
	5	Blumea megacephala (Randeria) C. C.	Pme lan
		Chang & Y. Q. Tseng	
	6	Blumea pterodonta DC.	Unknown
	7	Blumea sericans (Kurz) Hook. f.	Kadu
	8	Blumea tenella DC. ex Decne	Unknown

Table 1 List of Collected Species

1. Blumea barbata DC. in Wight, Contrib, Bot. India 14. 1834.

Myanmar name	:	Ya nat
English name	:	Unknown
Flowering period	:	December to June

Pollen morphology

Triporate, spheroidal, medium, $30.0 - 32.4 \mu m$ in diameter; amb rounded triangular: pori lolongate, $15.6 - 18.0 \times 1.8 - 2.4 \mu m$ in length and breadth; exine $6.0 - 8.4 \mu m$ thick, sexine $4.8 - 6.0 \mu m$ thick, sexine thicker than nexine; sculpturing echinate; spine $3.6 - 6.0 \mu m$ long, basal cushion about $6.0 \mu m$ in width, about $3.6 \mu m$ in height, interspinal space about $1.8 \mu m$ wide.

Specimens examined: Madaya Township, Sedawgyi; 22° 28' N and 96° 20' E; 9 March, 2018; Aye Aye Thin, collection no 4.

2. Blumea hieraciifolia (D.Don) DC., in Wight, Contrib. 15. 1834.

Erigeon hieraciifolius D.Don, Prodr. Fl. Nepal. 172. 1825

Blumea flexuosa, C. B. Clarke, Comp. Ind. 86. 1876.

Myanmar name : Unknown

English name : Unknown

Flowering period : October to April

Pollen morphology

Tetracolporate, spheroidal, medium, $31.2 - 32.4 \mu m$ in diameter; amb rounded quadrangular; colpi ¹/₂ way up to the pole, $15.6 - 16.2 \times 3.6 - 4.8 \mu m$ in length and breadth; pori circular, $2.4 - 3.6 \mu m$ in diameter; exine $4.8 - 5.8 \mu m$ thick, sexine about $4.6 \mu m$ thick, sexine thicker than nexine; sculpturing echinate; spine $3.6 - 4.8 \mu m$ long, basal cushion $4.8 - 6.0 \mu m$ in width, about $3.6 \mu m$ in height, interspinal space about $6.0 \mu m$ wide.

Specimens examined: Amarapura Township, Shanlaykyun; 22° 55' N and 96° 03'E; 13 March, 2018; Aye Aye Thin, collection no. 5.

3. Blumea junghuhniana (Miq.) Boerl., Fl. Ind. Bat. 1858.

Conyza junghuhniana Miq., Fl. Ind. Bat. 2: 55. 1856.

Conyza dasycoma var. pinnatifida (Miq) Boerl. Handl. Fl. Ned. Ind. 1:239. 1891.

Myanmar name : Unknown English name : Unknown Flowering period : September to March

Pollen morphology

Tricolporate, subprolate, small, $20 - 24 \times 18.0 - 19.2 \,\mu\text{m}$ in length and breadth; amb rounded triangular; colpi ³/₄ way up to the pole, $14.4 - 15.6 \times 1.2 - 2.4 \,\mu\text{m}$ in length and breadth; pori lolongate, $4.8 - 7.2 \times 1.8 - 2.4 \,\mu\text{m}$ length and breadth; exine about 5.4 μm thick, sexine about 3.0 μm thick, sexine thicker than nexine; sculpturing echinate; spine about 4.8 μm long, basal cushion about 2.4 μm in width, about 1.8 μm in height, interspinal space about 3.6 μm wide.

Specimens examined: Mahaaungmyay Township, Mandalay University Campus; 96° 03' N and 96° 08' E; 28 February, 2018; Aye Aye Thin, collection no. 2.

4.Blumea laciniata (Wall. ex Roxb.) DC., Prodr. 5: 436. 1836.

Conyza laciniata Wall.ex Roxb., Fl. Ind. 3:427. 1832.

Blumea glandulosa Benth., Fl. Hongk. 177.1861.

Myanmar name	:	Unknown
English name	:	Cut leaf Blumea
Flowering period	:	October to March

Pollen morphology

Tricolpate, spheroidal, small, 22.8 - 24.0 μ m diameter; amb rounded triangular; colpi ³/₄ way up to the pole, 15.6 - 18.0 × 2.4 - 4.8 μ m in length and breadth; exine 3.6 μ m thick, sexine about 2.4 μ m thick, sexine thicker than nexine; sculpturing echinate; spine 2.4 - 3.6 μ m long, basal cushion 2.4 - 3.6 μ m in width, about 2.4 μ m in height, interspinal space about 6.0 μ m wide.

Specimens examined:Chanmyathazi Township, Myothit; 21° 55' N and 96° 5' E; 1 March, 2018; Aye Aye Thin, collection no.3.

5. Blumea megacephala (Randeria) C. C. Chang & Y. Q. Tseng, Acta

Phytotax. Sin 12 (3): 310. 1974. *Blume ariparia* DC.var. *megacephala* Randeria, Blumea 10 (1): 215. 1960. Myanmar name : Pme lan English name : Unknown Flowering period : January to May

Pollen morphology

Triporate, prolate spheroidal, medium, $28.8 - 30.0 \times 25.2 - 28.0 \mu m$ in length and breadth; amb rounded triangular; pori lolongate, $6.0 - 7.2 \times 4.8 - 5.4 \mu m$ in length and breadth; exine 6.6 μm thick, sexine about 5.16 μm thick, sexine thicker than nexine; sculpturing echinate; spine about 6.0 μm long, basal cushion about 2.4 μm in width, about 2.4 μm in height, interspinal space 6.0 μm wide.

Specimens examined: Chanmyathazi Township, Myothit; 21° 55' N and 96° 5' E; 13 March, 2018; Aye Aye Thin, collection no. 8.

6. Blumea pterodonta DC. in Wight, Contrib. 16. 1834.

Laggera pterodont	a (DC.) Benth. & Hook.f., Gen. Pl. 2:290.1873.
Myanmar name	:	Unknown
English name	:	Unknown
Flowering period	:	February to May

Pollen morphology

Triporate, oblate spheroidal, medium, $25 - 32 \times 30 - 31 \mu m$ in length and breadth; amb rounded triangular; pori circular, $6 - 7 \mu m$ in diameter; exine $4.2 - 4.8 \mu m$ thick, sexine about 3.6 μm thick, sexine thicker than nexine; sculpturing echinate; spine $2.4 - 4.8 \mu m$ long, basal cushion about 3.6 μm wide, about 3.6 μm in height, interspinal space about 2.4 μm wide.

Specimens examined: Amarapura Township, Shanlaykyun; 22° 55' N and 96° 03' E; 13 May, 2018; Aye Aye Thin, collection no. 6.

7. Blumea sericans (Kurz) Hook. f., Fl. Brit. India.3: 262. 1881.

B. barbatavar. sericans Kurz, As. Soc. Bengal.46:188.1877.

Myanmar name	:	Kadu
--------------	---	------

English name	:	Unknown
--------------	---	---------

Flowering period : January to March

Pollen morphology

Tricoplate, spheroidal, small, 19.2 - 24.0 μ m in diameter; amb rounded triangular; colpi ³/₄ way up to the pole, 15.6 - 16.8 × 2.4 - 4.8 μ m in length and breadth; exine about 3.6 μ m thick, sexine about 2.4 μ m thick, sexine thicker than nexine; sculpturing echinate; spine 1.2 - 2.4 μ m long, basal cushion about 1.8 μ m in width, about 1.2 μ m in height, interspinal space about 1.2 μ m wide.

Specimens examined: Madaya Township, Sedawgyi; 22° 28' N and 96° 20' E; 13 March, 2018; Aye Aye Thin, collection no.7.

8. Blumea tenella DC. ex Decne in Wight, Contrib. Bot. Ind: 411. 1834.

Conyza tenella (Decne) DC.ex Miq., Fl. Ned. Ind. 240. 1856.

Myanmar name	:	Unknown
English name	:	Unknown
Flowering period	:	January to March

Pollen morphology

Tricolpate, subprolate, medium, $36.0 - 38.4 \times 31.2 - 32.4 \mu m$ in length and breadth; amb rounded triangular; colpi ½ way up to the pole, $18.0 - 19.2 \times 2.4 - 3.6 \mu m$ in length and breadth; exine $4.8 - 6.0 \mu m$ thick, sexine about $4.8 \mu m$ thick, sexine thicker than nexine; sculpturing echinate; spine about $3.6 \mu m$ long, basal cushion about $2.4 \mu m$ in width, about $2.4 \mu m$ in height, interspial space about $6.0 \mu m$ wide.

Specimens examined: Chanmyathazi Township, Myothit; 21° 55' N and 96° 5' E; 10 January, 2018; Aye Aye Thin, collection no. 1



Figure 2 Blumea hieraciifolia (D. Don) DC. Scale bar = 10 µm



A. HabitB. Polar viewC. Equatorial viewFigure 3Blumea junghuhniana (Miq.) Boerl. Scale bar = 10 μm







A. HabitB. Polar viewFigure 4 Blumea laciniata (Wall. ex Roxb.) DC.Scale bar = 10 μm







A. HabitB. Polar viewC. Equatorial viewFigure 5Blumea megacephala(Randeria) C. C. Chang & Y. Q. Tseng Scale bar = 10 μ m



A. Habit





bit B. Polar view C. Equatorial view **Figure 6** Blumea pterodonta DC. Scale bar = $10 \,\mu m$



A. HabitB. Polar viewC. Equatorial viewFigure 7Blumea sericans (Kurz) Hook. f.Scale bar = $10 \ \mu m$



Figure 8 Blumea tenella DC. ex Decne Scale bar = $10 \ \mu m$

Table 2	Pollen	Morphology	of 8	Species	of	Blumea
---------	--------	------------	------	---------	----	--------

No.	Scientific Name	Type of Aperture	Shape of EV	Size of EV	Colpi	Pori	Exine thickness	Size of pollen grains	Sculpture	Spine length
1	Blumea barbata DC.	P ₃	S	30.0-32.4	-	Lo	6.0-8.4	Me	echi	3.6-6.0
2	Blumea hieraciifolia (D. Don) DC	C_4P_4	S	31.2-32.4	1⁄2	Cir	4.8-5.8	Me	echi	3.6-4.8
3	<i>Blumea junghuhniana</i> (Miq.) Boerl.	C_3P_3	Subpro	20- 24×18.0- 19.2	3⁄4	Lo	5.4	Sm	echi	4.8
4	Blumea laciniata (Wall. ex Roxb.) DC.	C ₃	S	22.8-24.0	3⁄4	-	3.6	Sm	echi	2.4-3.6
5	<i>Blumea megacephala</i> (Randeria) C. C. Chang & Y. Q. Tseng	P ₃	Pros	28.8- 30.0×25.2- 28.0	-	Lo	6.6	Me	echi	6.0
6	<i>Blumea pterodonta</i> DC.	P ₃	Obls	25-32×30- 31	-	Cir	4.2-4.8	Me	echi	2.4-4.8
7	<i>Blumea sericans</i> (Kurz) Hook. f.	C ₃	S	19.2-24.0	3⁄4	-	3.6	Sm	echi	1.2-2.4
8	<i>Blumea tenella</i> DC. ex Decne	C ₃	Subpro	36.0- 38.4×31.2- 32.4	1⁄2	-	4.8-6.0	Me	echi	3.6
С	$=$ Colpate $P_3 =$	Triporate	3⁄4 =	3⁄4 way up t	o the po	ole	Pros = Prol	late Spher	oidal	
Р	= Porate Cir =	Circular	Pro =	Prolate			Obls = Obl	ate Spher	oidal	
Subp	oro = Subprolate Lo =	Lolongate	S =	Spheroida	1		$\frac{1}{2} = \frac{1}{2}$ wa	y up to th	e pole	

Artificial key to the species

1.	Aperture colporate or porate 2
1.	Aperture colpate 6
	2. Tricolporate or tetracolporate 3
	2. Triporate 4
3.	Pollen shape spheroidal, colpi ¹ / ₂ way up to the pole, pori circular
	2. Blumea hieraciifolia
3.	Pollen shape subprolate, colpi ³ / ₄ way up to the pole, pori lolongate
	3. Blumea junghuhniana
	4. Pori circular, exine 4.2 - 4.8µm thick 6. Blumea pterodonta
	4. Pori lolongate, exine 6.0-8.4- 4.8µm thick5
5.	Pollen shape spheroidal 1. Blumea barbata
5.	Pollen shape prolate spheroidal5. Blumea megacephala

	6. Colpi ¹ / ₂ way up to the pole8. <i>Blumea tenella</i>
	6. Colpi ³ / ₄ way up to the pole7
7.	Spine length more than 2.4 4. Blumea laciniata
7.	Spine length less than 2.47. Blumea sericans

Discussion

Pollen morphology was classified on the basic of aperture type, shape, size and sculpture pattern of the pollen. In this research 8 species belonging to 1 genus under 1 tribes were studied. In this study, apertures of pollen grain were tricolpate, triporate, tricolporate and tetracolporate, 3 species were tricolpate and triporate and only one species were tricolporate and tetracolporate and subprolate. The shape of pollen grains were spheroidal, prolate spheroidal, oblate spheroidal and subprolate, 4 species were spheroidal, 2 species were subprolate, only species were prolate spheroidal and oblate spheroidal. The sizes of pollen grains were found to be small and medium, 3 species were small and 5 species were medium. The sculptures of pollen grains were all echinate.

In the present study, the pollen grains of Tribes Inuleae are echinate and the pollen grains of *Blumea. barbata* DC., *B. junghuhniana* (Miq.) Boerl., *B. megacephala* (Randeria) C. C. Chang & Y. Q. Tsengare pori lolongate. Dinis and Pereira (2007) reported that the pollen grains of Tribes Inuleae were echinate and porilolongate. Therefore, the resulting character is agreed with previous finding. In the present study, the spine lengths of *Blumea* specieswere 1.3-6.0 μ m long and the shape of *Blumea junghuhniana* (Miq.) Boerl., *B. megacephala* (Randeria) C. C. Chang & Y. Q. Tsengand *B. pterodonta* DC. were prolate spheroidal, oblate spheroidal and subprolate. Pornponggrungrueng (2016) described that the pollen grains of this species were prolate spheroidal, oblate spheroidal and subprolate and the spine length of *Blumea* species were 1.3-6.0 μ m long which character is agreed with (Pornponggrungrueng 2016).

According to resulting data, spines, sculpture, shape and size are useful taxonomical character in systematic study of *Blumea* species.

Conclusion

It was concluded that the size, shape, sculpture and aperture of pollen grains were varied from species to another. The results showed that the form of pollen were useful as any other characters in the identification of plants.

By studying on pollen morphology of *Blumea*, it was observed that the members of *Blumea* possess the peculiar sculpture of pollens among species.

Thus, the present study will be useful information for the palynological characters which can be used in identification of some species of *Blumea*.

Acknowledgements

We would like to thank the Rector Dr Thura Oo, Monywa University for his permission this research. We would like to express our gratitude to Dr Tin Tin Nyunt, Professor and Head, Department of Botany, Monywa University for her encouragement, reviewing and providing necessary suggestions in this research. We are also thankful to Dr Theingi Htay, Professor, Department of Botany, Monywa University, for her invaluable advice. We would like to express my heartfelt gratitude and thank to Dr Nu Nu Yee, Professor and Head, Department of Botany, University of Mandalay, for her very kind help.

References

- Backer, B.A. (1965). "Flora of Java VolII." Nordboff. groningen, The Netherlands.
- Dassanayake, M. D. (1980). "A Revised Handbook of Flora of Ceylon Vol I". Amerind Publishing Co., Ltd New Delhi.
- Dinis and Pereira. (2007). A Contribution to the Ultrastructural Knowledge of the Pollen Exine in Tribe Inuleae. Department of Botany. University of Coimbra. Combra, Portugal.
- Erdtman, G. (1960). "The Acetolysis Method." A Revised Description, Sevensu. Bot. Tidsur, S4: S61-564
- Erdtman, G. (1971)."Pollen Morphology and Plant Taxonomy, An Introduction to Palynology I." Hafner Publishing Company., New York
- Hesse, M., Halbritter. H., Weber. M., Buchner. R., frosch- Radivo.A. and Ulrich. S. (2009)."Pollen Terminologg an Illustrated Handbook". University of Vienna, Austria
- Hundley, H. G. and Chit KoKo. (1989). "List of Tress, Shrubs, Herbs, and Principle Climbers, etc." Fourth Revised Edition, Mayangon, Rangoon, Burma.
- Kress, J. W., R. A. Defilipps and Daw Yin Yin Kyi. (2003). A Checuljist of the Trees, Shrubs, Herbs and Climbers of Myanmar." Department of Systematic Biology-Botany, National Meseum of natural history, Washington, DC: USA
- Meo, A. A. (2009). Palynological studies of Compositae from Pakistan. Boys Hostels, Quaid-i-Azan University, Islamabad, Pakistan.
- Peng, Y. C. Yang and Y. Luo. (2020). *Blumeahtamanthi* (Asteraceae) a New species from Myanmar. Gardening and Horiticulture Department. Tropical Botanical Garden. Yunnan 666303, China.
- Pornponggrungrueng, P. (2016). Pollen Morphology Tribes Inuleae (Compositae in Thailand), Department of Biology, Faculty of Science, Khon Koen University, Thailand.
- Simpson, M. G. (2006). Plant systematic. Elsevier academic press. California, USA.
- Yamada, Y. and Y. Iwanmai. (1988). Pollen illustrations and scanning electron micrographs. Kodansha. Tokyo.
- Zafar, M., M. Ahmad and M. A. Khan. (2007). Palynology of family asteraceae from Flora of Rawalpindi-Pakistan. Int. J. Agric. Biol. 9 (1):