Taxonomic Studies on the Genus *Miscanthus* II. Anatomical Patterns of Leaves*

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Recently, many anatomical works on grasses have been carried out by many botanists such as Prat^{1,2)}, Metcalfe³⁾, Tateoka⁴⁾, Brown⁵⁾ and Lee⁶⁾.

In 1960, Metcalfe described anatomical patterns of epidermis and cross section of leaves in *Miscanthus condensatus* and *M. sacchariflorus* in his book, Anatomy of Monocotyledons, Gramineae Part I.

In the present study of leaf anatomy, the diagnostic characters in delimiting among the species, section and subsection of *Miscanthus* have been investigated in as many Asiatic species as possible. The material for the study mainly consists of dry leaf blades preserved in herbaria of University of Tokyo, National Science Museum, and Kyoto University. Some fresh materials were also collected from different localities in Japan. The third leaf blade from the top was chosen, and softened by boiling water for about thirty minutes. The epidermis were then peeled from the leaf blade with cellulose acetate film. The detailed peeling procedure is as follows: The water was first removed from the leaf surface by blotting, and acetone was placed on the cellulose acetate film. The materials were then placed abaxially on the cellulose acetate film, and were pressed slightly with a finger for about two minutes. The film was finally mounted and the material was taken away by tweezers. Some softened materials were placed in 30% hydrofluoric acid for 5 hours, and were washed with fresh water for about an hour. The material was cut with a razor blade by hand, or sliding microtome. The sections were placed on a slide glass, and stained with safranin and fast green, and then dehydrated with alcohol and mounted on a slide glass.

Anatomical patterns of leaf blades in principal species

Miscanthus sinensis Andersson

Epidermis of leaf blade: Papillae 21 to 31 μ long, 18 to 21 μ wide, over the abaxial surface, costal and intercostal zones. Usually the papillae alternate with stomata. Stomata 26 μ long, 15 μ wide. Subsidiary cells dome-shaped. Bicellular microhairs 57(47-60) μ , basal cell 30(26 to 40) μ long, on side of the costal and intercostal zones of the abaxial surface. Cushion hairs on the abaxial surface. Silica cells dumbbell-shaped over the veins. Long cells deeply waved on the adaxial surface, and near the marginal portion of the abaxial surface (Fig. 1-2).

T. S.† Leaves: Midrib: M.d. bundles 152 μ long, 136 μ wide. The M.d., four to six M.j. and M.n. bundles rounded, or egg-shaped, and enclosed with a single bundle

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† T. S., cross section; M.d., median bundle; M.j., major bundle; M.n., minor bundle; G.d., girder bundle.

Fig. 1. Epidermis of leaf blade. 1. *M. floridulus*, 2. *M. sinensis*, 3. *M. sacchariflorus*, 4. *M. condensatus*, 5. *M. oligostachyus* var. *longiberbis*, 6. *M. nepalensis*.

B, Bicellular microhairs; C, Cushion hairs; P, Papillae; S, Stoma.

sheath interrupted adaxially, and abaxially. The G.d. bundles usually enclosed with a round and perfect single sheath. Sclerenchyma 2 to 3 layers on the abaxial and 7 to 9 layers from bundles to epidermis on the abaxial surface (Fig. 2-2).

Miscanthus floridulus (Labill.) Warburg

Epidermis of leaf blade: Papillae none on both surfaces. Stomata $18(15-21)\mu$ long, and $14(13-16)\mu$ wide. Subsidiary cells triangular. Bicellular microhairs $68(63-76)\mu$ long. The basal cells $44(38-47)\mu$ long, and distal cell tapering toward the apex. Silica cells present with cork cells on the costal zone. Long cells slightly waved (Fig. 1-1).

T. S. Leaves: Midrib: M.d.,* M.j., and M.n. bundles egg-shaped or rounded, enclosed by a bundle sheath except abaxial, and adaxial. G.d. bundles surrounded with a round bundle sheath. Sclerenchyma of bundle cap of M.d. bundle as broad as the bundle or nearly so, but the other bundles usually narrower than the bundles (Fig. 2-4).

The M.d. bundle 182μ long and 137μ wide: Lamina: 182μ thick. The pattern of the cross section is similar to *M. sinensis*, and it differs in three G.d. bundles present under bulliform cells, and well-developed colorless parenchyma. Papillae are not significant on the abaxial surface.

Miscanthus sacchariflorus (Maxim.) Benth

Epidermis of leaf blade: Papillae usually none on both surfaces. Bicellular microhairs $79(78-80)\mu$, basal cell $40(36-47)\mu$ long, and distal cell strongly tapered. Stomata $32(31-34)\mu$ long, $15(13-18)\mu$ wide. Subsidiary cells triangular on both surfaces. Silica cells dumbbell-shaped on costal zone. Long cell deeply sinuous (Fig. 1-3).

T. S. Leaves: Midrib: M.d., M.j., and G.d. bundles almost round. M.d. bundle 182μ long, 197μ wide. G.d. bundles usually enclosed by a single bundle sheath. Sclerenchyma, bundle caps of the M.d., and M.j. bundles 5 to 7 layers cell thickness, and not broader than the bundles.

Lamina: 197μ thick. Mn., and G.d. bundles completely enclosed by a bundle sheath. All patterns of the cross section are similar to those of *M. sinensis*, but no papillae have been seen on the abaxial surface (Fig. 2-4).

Miscanthus oligostachyus Stapf

Epidermis of Leaves: Large papillae 41μ long, 26μ wide, and alternate with stomata in the same row on the intercostal zone. Stomata 23 to 28μ long, 13 to 18μ wide. Bicellular microhairs $65(60-71)\mu$, basal cell 34 to 44μ long. Cushion hairs on the abaxial surface (Fig. 1-5).

T. S. Leaves: Midrib: M.d., M.n., and G.d. bundles are rounded. The length of M.d. bundles, and M.d. bundle cap is a half of thickness of the midrib or nearly so. The M.d. bundles 150μ thick, and 150μ wide. The two lateral bundles are much smaller than the M.d. bundles (Fig. 2-5).

Miscanthus condensatus Hackel

Epidermis of leaf blade: Papillae present over the abaxial surface. Large papillae 81μ long and 47μ wide on the costal zone of the abaxial surface. Small papillae alternate with stomata in the same row. Stomata 26μ long, 15μ wide. Subsidiary cells dome-shaped. Bicellular microhairs $70(55-81)\mu$ long, and basal cells $48(44-52)\mu$ long. Silica cell dumbbell-shaped over the veins. Long cells deeply waved on the adaxial surface (Fig. 1-4).

Fig. 2. T.S. Midrib. 1. *M. condensatus*, 2. *M. sinensis*, 3. *M. nepalensis*, 4. *M. floridulus*, 5. *M. oligostachyus* var. *longiberbis*.

M.d., median bundle; M.j., major bundle; M.n., minor bundle; G.d., girder bundle.

T. S. Leaves: Midrib: M.d., M.j., and M.n. bundles clave- or bottle-shaped. The bundle sheath interrupted adaxially, and also some G.d. bundle sheath interrupted on the abaxially or adaxially. M.d. bundles $182\ \mu$ long, and $152\ \mu$ wide. Sclerenchyma 5 to 6 layers on the adaxial.

Lamina: $255\ \mu$ thick. M.j. bundles triangular or nearly so. M.n. bundles elliptical and enclosed by a bundle sheath completely, or except abaxially and adaxially. G.d. bundles rounded under the colorless parenchyma and bulliform cells (Fig. 2-1).

Miscanthus nepalensis (Trin.) Hackel

Epidermis of leaf blade: Papillae 13 to $28\ \mu$ long, 13 to $24\ \mu$ wide. Large papillae on the costal zone. Stomata 26 to $28\ \mu$ long, 13 to $15\ \mu$ wide. Subsidiary cells dome-shaped. Bicellular microhairs $44\ \mu$ long, basal cell $26\ \mu$ long. Cushion hairs on the abaxial surface. Silica cells dumbbell-shaped on the costal zone (Fig. 1-6).

T. S. Leaves: Midrib: M.d., M.n., and G.d. bundles are round. The bundles enclosed by a sheath except the abaxial. The M.d. bundles $120\ \mu$ long, $105\ \mu$ wide. Sclerenchyma of bundle cap 5 to 7 layers cell thickness, and as broad as the bundles.

Lamina: $120\ \mu$ thick. The M.j. bundle surrounded by a sheath except the abaxial, and adaxial. The M.n. and G.d. bundles are round, and enclosed with a bundle sheath. Fan-shaped large bulliform cells on the adaxial surface. The cell wall of bulliform cells sinuous. Colorless parenchyma is not well developed under bulliform cells (Fig. 2-3).

Anatomical characters of leaf blades in hybrids

In the present study, leaf anatomy was also carefully examined for some hybrids^{*7,8)}. The hybrids, *Miscanthus sinensis*×*M. floridulus* and *M. sinensis*×*M. sacchariflorus* show papillae and dome-shaped stomata on the abaxial surface of epidermis as in *Miscanthus sinensis*. The hybrid, *Miscanthus sinensis*×*M. condensatus* also has papillae as *Miscanthus sinensis*, but it shows Co type of leaf cross section. The modes of patterns of several characters in the hybrids are shown as follows.

Table 1.

	<i>M. sinensis</i>	Hybrid**	<i>M. floridulus</i>
Papillae	present	present	none
Stomata	dome-shaped	dome-shaped	triangular
Bicellular microhairs	$57-58\ \mu$	$76-86\ \mu$	$71-83\ \mu$
Cushion hairs	present	none	none

(to be continued)

* Hybrids (*M. sinensis*×*M. floridulus*, *M. sinensis*×*M. sacchariflorus*) from Dr. Adati of Fac. Agr., Mie University. Hybrid (*M. sinensis*×*M. condensatus*) from Dr. Nishikawa of Fac. Agr., Gifu University.

** Fig. 3-1.

Table 1.

(continued)

	<i>M. sinensis</i>	Hybrid*	<i>M. sacchariflorus</i>
Papillae	present	present	none
Stomata	dome-shaped	dome-shaped	triangular
Bicellular microhair	44-47 μ	63-65 μ	78-80 μ
	<i>M. sinensis</i>	Hybrid**	<i>M. condensatus</i>
Papillae	present (small)	present (small)	present (large)
Stomata	dome-shaped	dome-shaped	dome-shaped
Bicellular microhair	55-58 μ	58-61 μ	60-70 μ
T. S. Leaves	S type	Co type	Co type

* Fig. 3-4. ** Fig. 3-5.

Summary and Conclusion

1. The similarities on the gross morphology have been observed between *M. sinensis* and *M. condensatus*, or *M. sinensis* and *M. floridulus*. It is, therefore, rather difficult to classify the species of *Miscanthus* by the gross morphology. The species of *Miscanthus*, however, can easily be classified by the epidermal patterns. The epidermis of *Miscanthus* is grouped into the following three principal types.

a. The *sinensis*-type: Papillae present on the abaxial surface. Subsidiary cells dome-shaped. Cushion hairs usually on the abaxial surface; i.e. *Miscanthus sinensis*, *M. oligostachyus* var. *longiberbis*, *M. nepalensis* (Fig. 1-5, 6).

b. The *condensatus*-type: Papillae present on the abaxial surface. Large papillae present in the costal zone of the abaxial surface. Subsidiary cells dome-shaped. No. cushion hairs on the abaxial surface; i.e. *Miscanthus condensatus* (Fig. 1-4).

c. The *floridulus*-type: Papillae usually absent on the abaxial surface. Subsidiary cells triangular. Numerous stomata present on both surfaces of leaf blade; i.e. *M. floridulus*, *M. sacchariflorus* (Fig. 1-1, 3).

2. By the patterns of the cross section of leaves, the following three types are recognized.

a. The *sinensis*-type: Vascular bundles in midrib and lamina are round or egg-shaped, and enclosed more or less with a sheath throughout. Bundle cap is usually narrower than the bundles; e.g. *Miscanthus sinensis*, *M. floridulus*, and *M. sacchariflorus* (Fig. 3-2, 4).

b. The *oligostachyus*-type: Vascular bundles in midrib and lamina are rounded, and enclosed with a well-developed sheath. M.j. bundles not more than two in number. M.d. bundle cap usually wider than the bundles. Bulliform cells well-developed in the lamina; e.g. *M. oligostachyus*, *M. tinctorius*, *M. nepalensis*, *M. changii* (Fig. 2-3, 5).

c. The *condensatus*-type: Vascular bundles in midrib are bottle-shaped, and usually enclosed with a sheath except the abaxial side. Lamina is thicker than the others. Large papillae present on the costal zone of the abaxial side; e.g. *Miscanthus condensatus* (Fig. 2-1). The *M. condensatus* in the strict sense is very distinct from *M. sinensis* in the anatomical pattern.

3. The length of bicellular microhairs seems to vary according to environments

Fig. 3. Epidermis of leaf blade (1-4) and T.S. Midrib (5). 1. Hybrid (*M. sinensis* × *M. floridulus*), 2. Hybrid (*M. sinensis* × *M. condensatus*), 3. *M. floridulus* var. *papillatus*, 4. Hybrid (*M. sinensis* × *M. sacchariflorus*), 5. T.S. Midrib of hybrid (*M. sinensis* × *M. condensatus*).

B, Bicellular microhairs; P, Papillae; S, Stoma.

where the plants grow. The bicellular microhairs of *Miscanthus sacchariflorus*, *M. floridulus*, *M. floridulus* var. *taiwanensis*, and *M. condensatus* are long, while bicellular microhairs of *M. sinensis* and *M. oligostachyus* are shorter than the formers. Particularly *Miscanthus nepalensis* has very short bicellular microhairs (Fig. 4 and Table 2).

4. Similarities in epidermis and vascular bundles among the groups of *Kariyasua* and *Miscanthus nepalensis* are observed. The detailed discussion will be presented in the forthcoming paper (Part 3).

5. The epidermis of *Miscanthus sinensis* and *M. oligostachyus* can be regarded as the same type, but there are minor differences in the size and shape of stomata (Fig. 1-2, 5, 6).

6. The presence of papillae in hybrids between species of *Miscanthus* seems to be a dominant character than the absence of those.

7. In the hybrid between *M. sinensis* and *M. condensatus*, the papillae of *M. sinensis* type appeared, while the vascular bundles of *M. condensatus*-type were dominantly presented (Figs. 2-1, 3-5).

8. The leaf anatomy suggests that the several strains of *Miscanthus* are derived from natural hybridization of the two different species possibly growing intermingled; i.e. *M. sacchariflorus* var. *breviberbis* (*M. sinensis* × *M. sacchariflorus*), *M. floridulus* var. *papillatus* (*M. sinensis* × *M. floridulus*). *M. floridulus* var. *taiwanensis* (some taxa of *M. sinensis* × *M. floridulus*), *Miscanthus oligostachyus* var. *shinanoensis* (certain taxa of *Miscanthus* × *M. oligostachyus*) (Fig. 3-1, 2, 3, 4, 5).

9. Table 2 shows the outline of the leaf anatomies.

Fig. 4. Bicellular microhairs on leaf blade.

1. *M. condensatus*, 2. *M. sinensis* var. *longiaxis*, 3. *M. sinensis*, 4. *M. sacchariflorus*, 5. *M. sacchariflorus* var. *gracilis*, 6. *M. floridulus*, 7. *M. floridulus* var. *papillatus*, 9. *M. floridulus* var. *malayanus*, 10. *M. floridulus* var. *ryukyuensis*, 11. *M. floridulus* var. *taiwanensis*, 12. *M. tinctorius*, 13. *M. changii*, 14. *M. oligostachyus*, 15. *M. nepalensis*, 16. *M. oligostachyus* var. *shinanoensis*.

Table 2. Leaf anatomy in *Miscanthus*.*

	T.S.	E.T.	Papillae	stomata			B.C.	C.H.
				length	width	s.c.		
<i>M. sacchariflorus</i>	S	F	—	32 μ	15 μ	T	78 μ	—
<i>M. sac.</i> var. <i>breviberbis</i>	S	FS	few	30	12	TD	70	—
<i>M. sinensis</i>	S	S	+s	26	15	D	57	+
<i>M. floridulus</i>	S	F	—	18	14	T	68	—
<i>M. flor.</i> var. <i>papillatus</i>	S	S	+s	16	8	D	65	—
<i>M. flor.</i> var. <i>taiwanensis</i>	S	S	+s	20	15	D	88	—
<i>M. condensatus</i>	Co	Co	+1	26	15	D	70	—
<i>M. cond.</i> var. <i>intermedius</i>	Co	Co	+1	21	10	D	60	—
<i>M. oligostachyus</i>	O	S	+s	26	15	D	65	+
<i>M. oligost.</i> var. <i>shinanoensis</i>	SO	S	+s	26	17	D	77	+
<i>M. oligost.</i> var. <i>longiberbis</i>	O	S	+s	32	19	D	63	+
<i>M. tinctorius</i>	O	S	+s	24	19	D	47	+
<i>M. changii</i>	O	S	+s	20	10	D	49	—
<i>M. nepalensis</i>	O	S	+s	27	14	D	36	+

* T.S., type of leaf cross section; E.T., type of leaf epidermis; s.c., subsidiary cell; B.C., bicellular microhairs; C.H., cushion hairs; S, *sinensis*-type; F, *floridulus*-type; T, triangular; D, dome-shaped; —, absent; +, present; +s, present and small; +1, present and large; Co, *condensatus*-type; O, *oligostachyus*-type.

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摘 要

李 永 魯: ススキ属の分類学的研究 II. 葉の解剖型

本報は第1報につづくもので、東京大学理学部の原 寛教授のもとでなされたものである。ススキ類は外形上互いに類似しているため、分類上種々の困難がある。これをまず葉の解剖学的特徴にもとづいて分類したいと考えた。アジア全体にわたって産する多くの種類をとりあつかうとともに、足立氏および西川氏から送っていただいた交配雑種も調査し、解剖型および雑種における遺伝因子の表現を検討した。また自然雑種とおもわれるものを解剖学の見地から発見しようとした。葉の表皮型によって、ススキ型、ハチジョウススキ型およびトキワススキ型の3つに容易にわけられた。また葉の横断面の型によってこれを3つの型に分けた。ススキ型、ハチジョウススキ型およびカリヤス型がそれである。2細胞性微毛では長さに相当変異があることを認めたが、一般に、トキワススキ、オギ、ハチジョウススキでは長くススキ類では短かく、カリヤス類ではもっとも短かいことを認めた。カリヤス類と *M. nepalensis* は葉の解剖型が非常によく似ている。雑種において表皮上にパピラ (Papillae) のある形質は常に遺伝学的に優性であり、そのない形質は劣性であるように見える。ススキとハチジョウススキとの雑種ではススキのパピラが出現し、維管束はハチジョウススキ型で、あたかもススキの皮をかぶったハチジョウススキのようである。以上のような葉の解剖学の見地から、これらの雑種性をおびた変種とおもわれるものの数群を九州、信州および台湾などで見いだすことができた。

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