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Short Communication

Studies on floral characters and pollen behaviour at different levels of ploidy in mulberry (*Morus* spp.)

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Mulberry (*Morus* spp.) is generally a diploid, mono-genous, heterozygous tree belonging to the family Moraceae having 28 chromosomes (2n = 28). It has 24 species and one sub-species and distributed in wide area of tropical, sub-tropical, temperate and sub-artic zones [1]. It is rich in ploidy and a lot of triploid varieties have been found especially among *Morus bombysis* Koidz. In *M. cathayana* Hemsl. tetraploid, pentaploid and hexaploid varieties are present. *M. serrata* Roxb., indigenous to India, and *M. tiliaefolia* Makino, originally from Japan and Korea, are known to be hexaploids. *M. nigra* L. is a decosaploid (2n = 22x = 308), with the largest number of chromosomes among phanerogams indicating a lot of variation in chromosome numbers as well as level of ploidy in mulberry [2]. These variations in ploidy and geographical diversity have resulted in plasticity of mulberry for many characters. Distingnishing ploidy by pollen fertility, pollen size, stigma length and other characters has been well correlated with the cytological confirmation [3]. The present communication deals with floral morphology and pollen behaviour related to ploidy.

Nine dioecious, male germplasm accessions three each of diploid (Mandalaya, Kosen and Burma-8), triploid (Tr-4, Tr-9 and Tr-10) and tetraploid (T-4, T-8 and T-11) were studied during the flowering season i.e., January to April in the years 2000-2002. Measurements of well-grown catkin of all the accessions were recorded for catkin length, breadth and florets/catkin. Fresh pollen was collected in the Petri dish from the single catkin during 10.30-11.0 hours of the day. Pollen diameter, giant pollen and viability were tested with 2% aceto-carmine solution.

More than 500 pollen grains per plant were scored. Only well filled, stained pollen grains were used for diameter measurement. The greater diameter of each pollen grain was measured at a magnification of 600x with the aid of an ocular micrometer. For germination study, pollen grains were allowed to germinate in 10% sucrose solution in hanging drop method in the cavity slides. Pollens 1.5 times larger than the normal pollen grain were considered as giant pollen. Data were statistically analysed.

ANOVA showed significant difference among the accessions of different ploidy on different characters (Table 1 and 2). Data on catkin length and width of triploids were larger than the diploid and tetraploid accessions (Table 1) ranging from 28.30-30.14 mm in triploids followed by diploid (25.94-29.89 mm) and tetraploid (19.95-21.66 mm). Similar trends were observed in catkin width also. Among the accessions at different levels of ploidy, number of florets in a single catkin was highest in diploids (35.81), which was significantly higher ($P < 0.01$) than the triploids and tetraploids except in Mandalaya (22.65). The flowers in triploids did not show any morphological changes other than superiority in size. The inheritance of bigger size of catkin but less number of florets in the catkins in triploid accessions indicated genetical advance over the diploids and tetraploids where triploids were originated either through crossing of diploid (2n = 28) and tetraploid (2n = 4x = 56) or a natural triploid (2n = 3x = 42). The reduced number of florets per catkin in triploids and tetraploids did not follow the earlier observation [4].

Average size of pollen grain of the accessions significantly varied with the difference in ploidy level. The diameter of pollen ranged from 18.76 to 22.46 µm. Triploids showed highest values (20.61-22.46 µm) followed by tetraploid (19.20-20.43 µm) and diploid (18.76-19.03 µm) accessions. The accessions, which were diploid and tetraploid showed uniformity in pollen size with an average size of 18.89 µm in diploid and 19.59 µm in tetraploids but for triploids (21.67 µm) the size varied significantly. The number of germ pores in the pollen also influenced the size of the pollen. Generally, two germpores were recorded in the pollen of diploid, triploid and tetraploid genotypes except three germpores in rare occasions in triploids. The occurrence of different sizes of pollen in the triploids placed them in between the tetraploids and diploids. Stanley and Linskens [5] reported that in many crop plants, pollen size has positive correlation with the chromosome numbers and ploidy level except few herbaceous plants, where pollen size has negative correlation [6]. The pollen size either increased or decreased with the

Table 1. Variation of catkin size and pollen characters at different ploidy levels in mulberry

Genotypes	Ploidy	Chromosome No.	Catkin length (mm)	Catkin breadth (mm)	No. of florets/ catkin	Pollen diameter (μm)	Pollen germination (%)	Giant pollen (%)
T-4	4x	56	19.95(1.091)	6.86(0.120)	27.12(0.489)	19.20(0.922)	57.50(0.922)	1.49(0.247)
T-8	4x	56	21.56(0.943)	6.16(0.092)	25.78(0.214)	19.34(1.077)	57.80(1.077)	1.68(0.098)
T-11	4x	56	21.66(0.681)	6.57(0.249)	22.87(0.323)	20.43(0.327)	55.40(1.281)	1.65(0.186)
Mean			**21.05**	**6.55**	**25.26**	**19.59**	**56.70**	**1.60**
Tr-4	3x	42	29.92(0.631)	9.02(0.117)	28.07(0.870)	20.61(0.466)	45.60(0.917)	2.63(0.450)
Tr-9	3x	42	28.30(0.856)	8.70(0.126)	25.99(1.057)	22.46(0.543)	46.63(1.114)	2.96(0.835)
Tr-10	3x	42	30.14(0.594)	8.92(0.140)	25.52(0.802)	21.94(0.934)	46.50(1.688)	2.63(0.390)
Mean			**29.45**	**8.88**	**19.86**	**21.67**	**46.24**	**2.74**
Kosen	2x	28	29.89(0.608)	7.95(0.287)	50.83(0.902)	19.03(0.469)	89.40(1.020)	-
Mandalaya	2x	28	25.94(0.626)	7.91(0.104)	22.65(0.951)	18.89(0.587)	90.10(1.044)	-
Burma-8	2x	28	26.42(0.721)	7.55(0.143)	33.96(1.017)	18.76(0.335)	88.90(1.221)	-
Mean			**27.41**	**7.80**	**35.81**	**18.89**	**89.46**	-
CD at 5%			0.74**	0.13**	0.73**	0.55**	1.11**	0.35**

Data in parenthesis are Standard Deviation of Mean

Table 2. Estimates of genetic parameters for catkin and pollen characters in mulberry

Source	Catkin length (mm)	Catkin breadth (mm)	No. of florets/catkin	Pollen diameter (μm)	Pollen germination (%)	Giant pollen (%)
Mean ± SE	25.97±0.41	7.75±0.11	29.19±0.83	20.07±0.15	64.20±1.95	1.45±0.12
Range	19.8-30.94	6.0-9.2	23.1-52.1	18.3-22.9	44.0-91.0	0-4.6
F	**	**	**	**	**	**
PCV	15.428	13.461	30.019	6.846	30.404	82.630
h^2	0.997	0.9989	0.9995	0.9899	0.9997	0.994
Expected genetic gain %	31.72	27.703	61.8127	13.959	62.6214	169.40

increase or decrease of ploidy level. Increase in average size of pollen in triploid was due to wide variation with more number of bigger sized pollens. The size variation of pollen in triploid genotypes indicated their hybridity through crossing of diploid and tetraploid genotypes. The tendency of occasional occurrence of giant pollen with good fertility in the diploid genotypes help to explain the natural occurrence of polyploid cultivars though incidentally no giant pollen were observed in the diploid genotypes.

Results on pollen germination showed that pollen viability varied significantly with difference of ploidy. Pollen germination was highest in diploid (89.46%) followed by tetraploid (56.90%) and triploid (46.24%). The reduction of pollen fertility coupled with presence of giant pollen (>1%) in triploid and tetraploid corroborates the observations of earlier report in mulberry [4]. The less germination of pollen in triploids may be due to genetical imbalance and also due to variation in chromosomes [7].

Catkin size, number of florets per catkin, pollen germination and giant pollen number showed a wider range. Maximum value of phenotypic co-efficient of variation was observed in occurrence of giant pollen (82.63%) followed by pollen germination (30.40%) and number of florets per catkin (30.02%). The traits giant pollen, pollen germination and number of florets per catkin indicate high heritability while catkin size as moderate heritability. Therefore, for using the mulberry genetic resources as male parent in breeding, pollen germination, occurrence of giant pollen, pollen diameter, catkin size and also frequency of florets per catkin may be studied for preliminary screening of ploidy before cytological confirmation.

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