Cytology of Some W. Himalayan Species of *Polygonatum* and *Disporum*

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The genera *Polygonatum* and *Disporum* belong to the tribe Polygonatae of the family Liliaceae (Hutchinson 1959). Important cytotaxonomical investigations on this tribe have been made by Hasegawa (1932, 1933), Matsuura and Suto (1935), Satô (1942), Eigsti (1942), Ownbey (1944), Suomalainen (1947), Therman (1950, 1953 a, b, 1956), Jones (1951) and Mehra and Pathania (1960).

According to Airy Shaw (1966) the genus *Polygonatum* comprises 50 species which are chiefly met with in North temperate regions. Seven species are represented in the W. Himalayas (Hooker 1894). Four species have been studied presently.

The genus *Disporum* constitutes 20 species (Airy Shaw l. c.) which are Asiatic and North American in distribution. Only one species, *D. cantoniense* occurs in the W. Himalayas and has been studied.

Materials and methods

Collections for cytological studies were made from Kashmir, Nainital and Simla hills. Voucher specimens have been deposited in the Herbarium of Department of Botany, Panjab University, Chandigarh-14. The technique adopted is the same as described earlier (Mehra and Sachdeva 1976) and the terminology adopted in respect of karyotype is after Battaglia (1955).

Observations

Polygonatum Miller

P. sibiricum Red. The species occurs in Simla and Kashmir hills between 1,800-2,400 m. Somatic count from root-tips showed $2n=30$ (Figs. 1, 1a). The karyotype consisted of fifteen pairs of chromosomes (Fig. 11a). The largest chromosome pair was submetacentric and one chromosome of this pair showed a secondary constriction in the long arm. The next four pairs were acrocentric, one of them showing a secondary constriction in the long arm. The sixth pair, which was also of the same size as the previous four, was submetacentric and possessed a secondary constriction in the long arm. Among the three next pairs of medium-sized chromosomes one pair was metacentric, another submetacentric and the third acrocentric. The remaining six pairs were small, three amongst them metacentric and three submetacentric. Chromosome size ranged from 8.5 to 2.5 μ .

Meiosis was perfectly normal showing 15 chromosomes at each pole at M_{II}

Figs. 1-3. 1 and 2. *Polygonatum sibiricum*. 1, root-tip cell showing $2n=30$. $\times 1510$. 2, M_{II} showing 15 chromosomes at each pole. $\times 2260$. 3, *P. geminiflorum*, $2n=30+2f$, root-tip mitosis. $\times 1510$. 1a and 3a. Explanatory diagrams of Figs. 1 and 3.

(Fig. 2). The difference in size of large and small chromosomes was quite evident, the latter mostly occupying the centre of the plate.

Kurosawa (1966) also reported $2n=30$ in this species from E. Himalayas. However, the karyotype in the two taxa differs in many respects. While in the present population there were four metacentric, six submetacentric and five acrocentric pairs, the taxon investigated by Kurosawa (l. c.) showed four metacentric, four submetacentric and seven acrocentric pairs. Though the largest chromosome pair in both taxa is submetacentric, it was heteromorphic with respect to secondary constriction in the present population, while no secondary constriction was reported in this pair in Kurosawa's material. Furthermore, while in the present taxon out of five acrocentric pairs only one chromosome of one pair possessed secondary constriction in its long arm, in Kurosawa's material out of seven acrocentric pairs one pair had secondary constriction in the long arm and other two pairs were heteromorphic with respect to secondary constriction.

It is also interesting to note that the W. Himalayan material of this species collected from Mussoorie hills showed $2n=36$ (Mehra and Pathania 1960). Kumar (1959) reported $2n=38$ in this species. Other numbers recorded for this species are $2n=20, 21, 26$ (vide Darlington and Wylie 1955).

P. geminiflorum Decne: The species occurs in Kashmir hills at an altitude of 2,700 m. Root-tip mitoses showed $2n=30$ (Figs. 3, 3a). Its karyotype is represented in Fig. 11 b. The largest chromosome pair was submetacentric, while the next five in size gradation were acrocentric. Of the nine pairs of small chromosomes two were metacentric, while the rest were submetacentric. None of the chromosomes revealed any secondary constriction. In addition to the normal complement, two small fragments were also noticed in some of the cells. Chromosome size ranged from 6.5μ to 2.5μ .

PMCs revealed $n=15$ at M_{II} (Fig. 4). Meiosis was normal.

Therman (1953b) recorded $2n=28$ in this species from Scotland.

P. multiflorum All. The species occurs in Nainital hills at 1,800–2,550 m. Chromosome number was determined from root-tip mitoses only which showed $2n=22$ (Fig. 5). The karyotype consisted of five pairs of large and six pairs of small chromosomes (Fig. 11c). Of the former, three pairs were submetacentric and two acrocentric. One chromosome pair belonging to the former category showed a secondary constriction in the short arm. Of the six pairs of small chromosomes, five were submetacentric and one was acrocentric, all without any secondary constriction. Chromosome size ranged from 9μ to 3μ .

The present report of chromosome number agrees with that of Mehra and Pathania (1960) of the population from Dalhousie hills and Simla hills. However, the karyotype in the two cases differs. While the present material showed eight pairs of submetacentric and three pairs of acrocentric chromosomes, there were nine pairs of submetacentric and two pairs of acrocentric chromosomes in the taxon investigated earlier. Also, the latter had two pairs of large chromosomes with secondary constriction, while in the present material secondary constriction was noticed only in a single large pair.

Therman (1953a) reported $2n=18$ in this species. Various other chromosome

Figs. 4-8. 4, *Polygonatum geminiflorum*, $n=15$ at M_{II} . $\times 1510$. 5, *P. multiflorum*, $2n=22$, root-tip mitosis. $\times 1510$. 6 and 7, *P. verticillatum* (Gulmarg taxon). 6, root-tip cell showing $2n=28$. $\times 1510$. 7, M_I of meiosis showing 14 bivalents. $\times 2260$. 8, *P. verticillatum* (Simla taxon), $2n=30$, root-tip mitosis. $\times 1510$.

numbers recorded by different authors from different regions are $2n=20$, 24, 28 and 30 (vide Darlington and Wylie. L. C.).

P. verticillatum All. The species is distributed in Simla, Kumaon and Kashmir hills at an altitude of 1,800–3,000 m.

The present study revealed the existence of three cytotypes in the W. Himalayas (Fig. 12). The taxon worked out from Gulmarg (Kashmir) showed $n=14$ and $2n=28$, another from Simla hills had $2n=30$, while in the third from Nainital hills the chromosome number was $n=30$ and $2n=60$.

Fig. 12. *Polygonatum verticillatum*. Cytotypes with $2n=28$, $2n=30$ and $2n=60$.

Somatic chromosome number in the Gulmarg taxon was determined from root-tip mitoses which revealed $2n=28$ (Fig. 6). The karyotype was of asymmetrical type (Fig. 11d) with eight pairs of acrocentric, two pairs of submetacentric and four pairs of metacentric chromosomes. One metacentric chromosome pair showed a satellite attached to the distal end of one of its arm. Chromosome size ranged from 5μ to 2μ . Meiosis was perfectly normal, with 14 bivalents at M_I (Fig. 7).

The population from Simla hills showed $2n=30$ in root-tips (Fig. 8). The karyotype consisted of five pairs of long, three pairs of medium-sized and seven pairs of small chromosomes (Fig. 11e). Of the five pairs of long chromosomes, one pair was submetacentric and the remaining four were acrocentric. One pair of chromosomes of this category had a secondary constriction in the long arm. The three pairs of chromosomes next in size were all acrocentric. Of these, one pair had a secondary constriction in the long arm. Out of seven pairs of small chromosomes, three pairs were submetacentric and the remaining four were metacentric.

The population from Nainital hills showed $n=30$, and $2n=60$ as seen in root-tip mitosis (Fig. 9). Of these, twenty chromosomes were metacentric, sixteen submetacentric and twenty four were acrocentric. Secondary constrictions were not visible in any of these sixty chromosomes. PMCs had a highly irregular meiosis

showing multivalents, univalents and laggards both at A_I and A_{II} . Chromosome number could easily be counted only in a few cells at M_{II} (Fig. 10).

Table 1 and Fig. 12 depict the morphological variation present in the three taxa. It is noticed that there is increase in leaf length, stomatal size and pollen size with increase in chromosome number. But while the breadth of the leaf is more in the taxon with $2n=30$ compared to that with $2n=28$, it decreases in the cytotype with $2n=60$. The plant height and diameter of flower, however, remain almost unaffected.

Figs. 9-10. 9, *Polygonatum verticillatum* (Nainital taxon), $2n=60$, root-tip mitosis. $\times 1510$. 10, *Polygonatum verticillatum* (Nainital taxon), $n=30$ at M_{II} . $\times 755$.

Kurosawa (1966) reported $2n=30$ in the population worked out from Sandakpu (E. Himalayas). The various other numbers recorded in this species are $2n=27$, 28, 30, 60, 84, 86-91 (vide Darlington and Wylie, l. c.).

Disporum Salisb.

D. cantoniense (Lour.) Merr. The species is met with in Kumaon hills at eleva-

tions between 1,500–2,400 m. Somatic chromosome number was determined from cells of the sporogenous tissue which revealed $2n=14$ (Fig. 13). The karyotype consisted of five pairs of long chromosomes and two pairs of medium-sized chromosomes (Fig. 15). The longest chromosome pair was submetacentric with one chromosome showing a secondary constriction in the long arm. Of the four pairs of

Figs. 11 a–e. Idiograms of a, *Polygonatum sibiricum*, $2n=30$; b, *P. geminiflorum*, $2n=30+2f$; c, *P. multiflorum*, $2n=22$; d, *P. verticillatum* (Gulmarg taxon), $2n=28$; e, *P. verticillatum* (Simla taxon), $2n=30$.

chromosomes next in size, two pairs were acrocentric and remaining two submetacentric. Among the two pairs of medium-sized chromosomes one was submetacentric while the other was acrocentric. Chromosome size ranged from $14\ \mu$ to $6\ \mu$.

PMCs revealed $n=7$ at M_I (Fig. 14). Meiosis was normal.

Table 1. Analysis of various characters of three races of
Polygonatum verticillatum All.

	2n=28	2n=30	2n=60
Locality and altitude	Gulmarg, Kashmir hills, 2,700 m	Narkanda, Simla hills, 2,700 m	Nainital, Kumaon hills, 2,400 m
Habitat	Shaded forest where soil is rich in humus	Shaded forest where soil is rich in humus	Shaded forest on slopes
Habit	Erect, leafy, rootstock creeping	Erect, leafy, rootstock creeping	Erect, leafy, rootstock creeping
Plant height	48-56 cm	46-58 cm	48-64 cm
Leaf characters:-			
a) leaves	4-nately whorled, sessile, linear lanceolate, acuminate	4-nately whorled, sessile, linear lanceolate, acuminate	4-nately whorled, sessile, linear and acuminate
b) Length of the leaf	5-7 cm	8-12 cm	9-12 cm
c) Breadth of the leaf	1.1-1.3 cm	1.4-1.9 cm	0.5-0.8 cm
d) Stomatal size	22.5 × 21.7 μ	30.4 × 26.6 μ	38.92 × 38.75 μ
Peduncles	Whorled, 2-3 fid	Whorled, 2-3 fid	Whorled, always 2 fid
Flowers	White green with perianth constricted in the middle	White green with perianth constricted	White green with perianth constricted
Diameter of flower	0.5-0.6 cm	0.6-0.7 cm	0.6-0.7 cm
Pollen size	35.72 × 27.88 μ	34.96 × 30.7 μ	40.91 × 36.47 μ

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Figs. 13-15. 13 and 14. *Disporum cantoniense*. 13, mitotic metaphase with 2n=14, arrow indicates the position of secondary constriction. ×1510. 14, M_I showing 7 bivalents. ×1510. 15, idiogram of *Disporum cantoniense*, 2n=14.

The present report is in accord with the number reported for *D. cantoniense* var. *parviflorum* (Wall) Hara, in which Kurosawa (1966) discovered two cytotypes with $2n=14$, 30 from Darjeeling (E. Himalayas). The karyotype of the present taxon agrees almost completely with the description of Kurosawa for the cytotype $2n=14$ (l. c.) except for a slight difference. While no secondary constrictions were observed by the latter author in any of the chromosomes, in the present material one chromosome of the longest pair was found to possess a secondary constriction in its long arm.

Four base numbers $x=6$, 8, 9 and 11 have been mentioned for this genus by Darlington and Wylie (1955). Chao *et al.* (1963) suggested another base number $x=7$ on the basis of $2n=14$ in *D. shimadai*. The finding of $2n=14$ in *D. cantoniense* lends further support to this view.

Another W. Himalayan population of this species from Mussoorie hills worked out by Mehra and Pathania (1960) showed $2n=16$. It was observed that the karyotype of this Himalayan taxon was different from that of Japanese form (Hasegawa 1932), although both have 16 chromosomes.

An interesting point to note is that all East Asian species of *Disporum* possess large chromosomes as compared to the North American species *D. smithii* ($2n=16$), *D. oreganum* ($2n=18$) and *D. hookerii* ($2n=18$) in which the chromosome size ranges from 4.9μ to 2.0μ (Therman 1956).

Discussion

From the above it is clear that there is not only numerical alteration of chromosomes in the different populations of the species of *Polygonatum* and *Disporum* but there is structural alteration in chromosomes as well. This phenomenon is not restricted to these two genera alone but is widespread in monocots and in dicots too. There could be three different explanations. 1) Such alteration could occur in floral primordia so that both the ovules as well as the anthers would have the same initial constitution. Fertilization occurring between the sex cells from these would give rise to seeds with the altered chromosome number and/or karyotype from the start from which would emerge plants which would show normal pairing and fertility. 2) Alterations could occur in the embryos within the seeds. This could be the effect of endosperm in ageing or old seeds. D'Amato (1948, 1951) has shown that in old seeds such alterations are visible in the primary roots produced on their germination. These could as well occur in embryonal shoot tips. 3) Such alterations could occur in the primordia responsible for vegetative reproduction in species that multiply by this means. This mechanism has specially been emphasised upon by Sharma (1956). Any or all of these methods could have been operative in nature. Chromosomal dynamism involving their numbers as well as structural changes seem to have been the source of polymorphic populations adapted to different geographic areas and habitats in the two genera *Polygonatum* and *Disporum*.

With alteration in chromosome numbers, or of karyotype, even if the number remains the same, it is conceivable that there will be change in the phenotypic characters of populations. What should be the status of such variants, say for example

the different forms exhibited in *Polygonatum verticillatum* (cf. Fig. 12), and this holds good in several other cases too. In the case of *Saccharum spontaneum* with wide variations in chromosome numbers in the chromosomally floating populations, there are observed significant morphological variations (Mehra and Sood 1975). These, however, cannot justifiably be used for erecting new species or even new varieties in the taxonomic sense. At best they can be regarded merely as cytotypes in the dynamic variability of the species. Variation could occur because of gene changes, position effect of genes due to changes in karyotype, alteration in chromosome numbers which may be balanced as in polyploidy, or uneven as in dysploidy, but these variations have to be judged in the total context of variation pattern. If it is merely hovering around a particular plan, no formal taxonomic recognition need be given.

Summary

Six taxa belonging to four species of the genus *Polygonatum* and one species of the genus *Disporum* have been studied cytologically. New chromosome number $2n=30$ has been recorded in *P. geminiflorum*. In addition to the earlier reports made by Mehra and Pathania (1960), one more cytotype in *P. sibiricum* and three more cytotypes in *P. verticillatum* have been found to occur in the W. Himalayas. *Disporum cantoniense* with $2n=14$ has been recorded from Nainital hills. The possible mechanisms for alteration in chromosome number and karyotype have been suggested.

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