Cytogenetics of *Crotalaria* VIII. Male meiosis in 26 species

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In an earlier communication, male meiosis in 8 species of *Crotalaria* was described (Verma and Raina 1980). The same study was extended to few more species. The present study deals with male meiosis in 26 species and of these, 13 have not been investigated so far. If only detailed meiosis reported earlier is taken into consideration, in that case 21 species are new in the present investigation.

Material and methods

Seeds of various species of *Crotalaria* were very kindly supplied by U. S. Department of Agriculture and Dr. R. M. Polhill of Kew Botanical Gardens, England.

Young flower buds were fixed in freshly prepared acetic-alcohol (1:3). Following the usual technique, anthers were stained in leucobasic-fuchsin and squashed in 1% iron-acetocarmine. Microphotographs were taken from temporary preparations.

Observations

The meiotic data have been summarized in Tables 1, 2.

Associations

Diploids

C. incana ($n=7$): In the present study *C. incana* was the only species where seven as the gametic number was observed. All the cells analysed had seven bivalents (Figs. 1, 2). The mean values per cell of ring (3.08) and rod (3.92) bivalents were almost equal. Such ratio, as we shall see, was absent in other species. Previous authors have also reported the same gametic number (Atchison 1950, Shibata 1962, Karapovickas and Fuches de Karapovickas 1957, Magoon *et al.* 1963, Patil and Chennaveeraiah 1975, Gupta and Gupta 1978b). In addition to bivalents, Magoon *et al.* (1963) and Patil and Chennaveeraiah (1975) noted cells with univalents, tri-valents and quadrivalents. Patil and Chennaveeraiah (1975) also noted in a few cells a fragment and an extra chromosome. The present authors did not find such abnormalities.

C. agatiflora ($n=8$): All the cells had eight bivalents (Fig. 4). The average values per cell of ring and rod bivalents were 6.28 and 1.72 respectively.

C. alata ($n=8$): This species also had eight bivalents in all the cells analysed (Fig. 6) and the bivalents on the average resolved into 6.96 ring and 1.04 rod bivalents. The same gametic number has been reported by Kawakami (1930) and Chennaveeraiah and Patil (1972).

C. australis ($n=8$): Eight bivalents were encountered in all the cells (Figs. 7-9). On the average 5.64 were ring and 2.36 were rod bivalents.

C. brachystachya ($n=8$): All the cells has eight bivalents (Figs. 10, 11), of which on the average 5.64 were ring and 2.36 were rod bivalents.

Table 1. Average, number and range of associations

Species	2n	No. of cells analysed	Cells with 7/8/16 II		Ring bivalents		
			No.	Percentage	No.	Mean	Range
<i>C. incana</i>	14	25	25	100	77	3.08 $\pm$ 1.44	1-6
<i>C. agatiflora</i>	16	25	25	100	157	6.28 $\pm$ 1.13	3-8
<i>C. alata</i>	16	25	25	100	174	6.96 $\pm$ 1.61	2-8
<i>C. australis</i>	16	25	25	100	141	5.64 $\pm$ 1.07	4-8
<i>C. brachystachya</i>	16	25	25	100	141	5.64 $\pm$ 1.25	3-8
<i>C. falcata</i>	16	25	25	100	150	6.0 $\pm$ 1.29	3-8
<i>C. grahamiana</i>	16	25	25	100	165	6.6 $\pm$ 1.60	3-8
<i>C. grantiana</i>	16	25	25	100	163	6.52 $\pm$ 1.38	5-8
<i>C. grandibracteata</i>	16	25	25	100	160	6.40 $\pm$ 1.22	4-8
<i>C. laburnifolia</i>	16	25	25	100	186	7.44 $\pm$ 0.82	6-8
<i>C. leioloba</i>	16	25	25	100	177	7.08 $\pm$ 0.95	5-8
<i>C. leubnitziana</i>	16	25	25	100	124	4.96 $\pm$ 1.61	0-8
<i>C. maxillaris</i>	16	25	24	96	139	5.56 $\pm$ 1.29	4-8
<i>C. medicaginea</i>	16	25	25	100	177	7.08 $\pm$ 0.81	5-8
<i>C. mucronata</i>	16	25	19	76	125	5.0 $\pm$ 1.89	1-8
<i>C. mysorensis</i>	16	25	25	100	180	7.2 $\pm$ 0.91	5-8
<i>C. ochroleuca</i>	16	25	25	100	128	5.12 $\pm$ 1.42	3-8
<i>C. pallida</i>	16	25	1	4	25	1.0 $\pm$ 0.86	0-3
<i>C. rhodesiae</i>	16	25	25	100	160	6.40 $\pm$ 1.08	5-8
<i>C. saltiana</i>	16	25	25	100	173	6.92 $\pm$ 0.90	5-8
<i>C. sericea</i>	16	25	25	100	192	7.68 $\pm$ 0.85	4-8
<i>C. spectabilis</i>	16	25	25	100	199	7.96 $\pm$ 0.20	7-8
<i>C. striata</i>	16	25	25	100	177	7.08 $\pm$ 0.90	5-8
<i>C. longirostrata</i>	32	25	25	100	308	12.32 $\pm$ 2.09	8-16
<i>C. sagittalis</i>	32	25	25	100	362	14.48 $\pm$ 1.22	11-16
<i>C. stipularia</i>	32	25	25	100	340	13.60 $\pm$ 1.55	11-16

C. falcata ($n=8$): All the cells had eight bivalents (Fig. 13). On the average 6.0 were ring and 2.0 were rod bivalents.

C. grahamiana ($n=8$): Eight bivalents were observed in all the cells (Figs. 16, 17). The bivalents on the average resolved into 6.6 ring and 1.4 rod bivalents. Chennaveeraiah and Patil (1972) have also reported the same gametic number.

C. grantiana ($n=8$): All the cells had eight bivalents (Figs. 19-21). On the average 6.52 were ring and 1.48 were rod bivalents. Same gametic number has been

reported by Chennaveeraiah and Patil (1972).

C. grandibracteata (n=8): Eight bivalents, found in all the cells (Fig. 14), on the average resolved into 6.40 ring and 1.60 rod bivalents.

C. laburnifolia (n=8): All the cells had eight bivalents (Figs. 22, 23) and the mean values of ring and rod bivalents were 7.44 and 0.56 respectively. The same gametic number has been reported by Chennaveeraiah and Patil (1972).

C. leioloba (n=8): Eight bivalents were encountered in all the cells (Figs. 24, 25). On the average 7.08 were ring and 0.92 were rod bivalents.

at diakinesis/metaphase I in *Crotalaria* species

Rod bivalents			Total bivalents			Univalents	
No.	Mean	Range	No.	Mean	Range	Mean	Range
98	3.92±1.44	1-6	175	7			
43	1.72±1.13	0-5	200	8			
26	1.04±1.61	0-6	200	8			
59	2.36±1.07	0-4	200	8			
59	2.36±1.25	0-5	200	8			
50	2.0 ±1.29	0-5	200	8			
35	1.4 ±1.60	0-5	200	8			
37	1.48±1.38	0-3	200	8			
40	1.60±1.22	0-4	200	8			
14	0.56±0.82	0-2	200	8			
23	0.92±0.95	0-3	200	8			
76	3.04±1.61	0-6	200	8			
59	2.36±1.25	0-4	198	7.92±0.40	6-8	0.16±0.80	0-4
23	0.92±0.81	0-3	200	8			
66	2.64±1.46	0-5	191	7.64±0.70	6-8	0.72±1.4	0-4
20	0.84±0.91	0-3	200	8			
72	2.88±1.42	0-5	200	8			
97	3.88±1.56	1-7	122	4.88±1.64	2-8	6.24±3.28	0-12
40	1.60±1.08	0-3	200	8			
27	1.08±0.90	0-3	200	8			
8	0.32±0.85	0-4	200	8			
1	0.04±0.20	0-1	200	8			
23	0.92±0.90	0-3	200	8			
92	3.68±2.09	0-8	400	16			
38	1.52±1.22	0-5	400	16			
60	2.40±1.55	0-5	400	16			

C. leubnitziana (n=8): All the cells had eight bivalents (Figs. 27, 28). The mean values of ring and rod bivalents were 4.96 and 3.04 respectively.

C. maxillaris (n=8): Eight bivalents were observed in 24 (96%) out of 25 cells analysed (Fig. 30). On the average 7.92 were bivalents and 0.16 were univalents and the former resolved into 5.56 ring and 2.36 rod bivalents. Magoon *et al.* (1963) have also reported the same gametic number. They observed cells with univalents ranging from 2-6.

Table 2. Average number and range of chiasmata and terminalization coefficient

Species	2n	No. of cells analysed	Chiasmata			Terminalization coefficient	
			Mean	Range	Terminalized	Unterminalized	Terminalization coefficient
<i>C. incana</i>	14	25	10.08 ± 1.44	8-13	8.96	1.12	0.88
<i>C. agatiflora</i>	16	25	15.72 ± 2.17	11-20	13.48	2.24	0.86
<i>C. alata</i>	16	25	19.08 ± 4.06	10-24	14.92	4.16	0.78
<i>C. australis</i>	16	25	13.68 ± 1.18	12-17	12.88	0.80	0.94
<i>C. brachystachya</i>	16	25	14.20 ± 1.63	11-17	13.32	0.88	0.93
<i>C. falcata</i>	16	25	15.40 ± 1.82	13-20	13.64	1.76	0.93
<i>C. grahamiana</i>	16	25	15.20 ± 2.67	11-22	14.60	0.60	0.96
<i>C. grantiana</i>	16	25	17.08 ± 2.79	10-23	14.16	2.92	0.82
<i>C. grandibracteata</i>	16	25	15.04 ± 1.71	12-18	14.00	1.04	0.93
<i>C. laburnifolia</i>	16	25	16.60 ± 1.60	14-20	15.44	1.16	0.93
<i>C. letoloba</i>	16	25	15.32 ± 1.24	13-18	15.04	0.28	0.98
<i>C. leubnitziana</i>	16	25	12.96 ± 1.61	10-16	12.56	0.40	0.96
<i>C. maxillaris</i>	16	25	14.20 ± 1.50	12-17	13.00	1.20	0.91
<i>C. medicaginea</i>	16	25	15.88 ± 1.33	13-19	15.12	0.76	0.95
<i>C. mucronata</i>	16	25	13.08 ± 2.51	7-17	12.88	0.20	0.98
<i>C. mysorensis</i>	16	25	15.36 ± 1.25	13-19	15.08	0.28	0.98
<i>C. ochroleuca</i>	16	25	13.88 ± 2.27	11-21	13.00	0.88	0.94
<i>C. pallida</i>	16	25	5.84 ± 2.07	2-11	5.76	0.08	0.99
<i>C. rhodesiae</i>	16	25	15.00 ± 1.35	13-18	14.40	0.60	0.96
<i>C. saltiana</i>	16	25	16.28 ± 1.56	14-20	14.92	1.36	0.88
<i>C. sericea</i>	16	25	17.88 ± 2.45	14-24	15.68	2.20	0.87
<i>C. spectabilis</i>	16	25	18.00 ± 1.58	16-21	15.96	2.04	0.88
<i>C. striata</i>	16	25	16.68 ± 2.23	14-21	14.84	1.84	0.88
<i>C. longirostrata</i>	32	25	29.44 ± 2.23	24-33	28.32	1.12	0.96
<i>C. sagittalis</i>	32	25	32.56 ± 2.21	28-37	30.28	2.28	0.92
<i>C. stipularia</i>	32	25	31.60 ± 2.30	27-37	29.48	2.12	0.95

Figs. 1-15. 1-3, *C. incana*. 1-2, metaphase I, 7 II. 3, anaphase I 7:7. 4-5, *C. agatiflora*. 4, diakinesis, 8 II. 5, anaphase I 8:8. 6, metaphase I in *C. alata*, 8 II. 7-9, metaphase I in *C. australis*, 8 II. 10-12, *C. brachystachya*. 10-11, diakinesis, 8 II. 12, anaphase I 8:8. 13, diakinesis in *C. falcata*, 8 II. 14-15, *C. grandibracteata*. 14, metaphase I, 8 II. 15, anaphase I 8:8.

Figs. 16-30. 16-18, *C. grahamiana*. 16-17, metaphase I, 8 II. 18, anaphase I 8:8. 19-21, diakinesis in *C. grantiana*, 8 II. 22-23, metaphase I in *C. laburnifolia*, 8 II. 24-26, *C. leioloba*. 24-25, metaphase I, 8 II. 26, anaphase I 8:8. 27-29, *C. leubnitziana*. 27, diakinesis, 8 II. 28, metaphase I, 8 II. 29, anaphase I 8:8. 30, diakinesis in *C. maxillaris*, 8 II.

C. medicaginea (n=8): All the 25 cells had eight bivalents and they on the average resolved into 7.08 ring and 0.92 rod bivalents. Magoon *et al.* (1963), Raghavan and Arora (1958) and Chennaveeraiah and Patil (1972) have reported the same gametic number for *C. medicaginea* and *C. medicaginea* var. *luxurians* while Bir and Kumari (1977) have reported intraspecific polyploidy (n=8, 16) in the species.

C. mucronata (n=8): Eight bivalents were observed in 76% cells. In 24% cells besides bivalents, univalents ranging from 2–4 were also observed (Figs. 31, 32). Out of an average 7.64 bivalents, 5.0 were ring and 2.64 were rod bivalents. The same gametic number has been reported by Atchison (1950), Tandon and Bhat (1970), Patil and Chennaveeraiah (1975) and Gupta and Gupta (1978b).

C. mysorensis (n=8): Eight bivalents, found in all the 25 cells (Fig. 34), resolved on the average into 7.2 ring and 0.8 rod bivalents. Previous authors have also reported the same gametic number (Chennaveeraiah and Patil 1972, Bir and Kumari 1977).

C. ochroleuca (n=8): All the cells had 8 bivalents (Figs. 36, 37). On the average 5.12 were ring and 2.88 were rod bivalents.

C. pallida (n=8): In 24 out of 25 cells, both univalents and bivalents were observed (Figs. 39, 40). The average number of univalents (6.24) outnumbered bivalents (4.88). As many as 12 univalents were observed in a few cells. Rod bivalents (3.88) predominated over ring bivalents (1.00).

C. rhodesiae (n=8): All the cells analysed had eight bivalents (Fig. 42). On the average 6.40 were ring and 1.60 were rod bivalents.

C. saltiana (n=8): Eight bivalents, observed in all the cells (Figs. 44, 45), resolved on the average into 6.92 ring and 1.08 rod bivalents. Previous authors have also reported the same gametic number (Chuang *et al.* 1963, Chennaveeraiah and Patil 1972, Gupta and Gupta 1978b, Bhaumik 1975).

C. sericea (n=8): All the cells had eight bivalents (Fig. 46). On the average 7.68 were ring and 0.32 were rod bivalents. The same gametic number has been reported by Atchison (1950), Roy and Sinha (1958), Magoon *et al.* (1963), Tandon and Bhat (1970), Bhaumik (1975), Bir and Kumari (1977) and Gupta and Gupta (1978b). In some cells, 2–4 univalents and 1 IV + 6 II have been reported by Magoon *et al.* (1963) and Bhaumik (1975), respectively.

C. spectabilis (n=8): Eight bivalents, found in all the cells (Figs. 47, 48), on the average resolved into 7.96 ring and 0.04 rod bivalents.

C. striata (n=8): All the cells had eight bivalents (Fig. 49). On the average 7.08 were ring and 0.92 were rod bivalents. Previous authors have also reported the same gametic number (Frahm-Leliveld 1957, Magoon *et al.* 1963, Chennaveeraiah and Patil 1972, Bhaumik 1975, Gupta and Gupta 1978b, Rao 1950).

Tetraploids

C. longirostrata (n=16): All the 25 cells analysed had sixteen bivalents (Fig. 50). On the average 12.32 were ring and 3.68 were rod bivalents.

C. sagittalis (n=16): It also had sixteen bivalents in all the cells (Figs. 52, 53). On the average 14.48 were ring and 1.52 were rod bivalents.

C. stipularia (n=16): As in *C. longirostrata* and *C. sagittalis*, all the cells had

Figs. 31-45. 31-33, *C. mucronata*. 31, metaphase I, 8 II. 32, metaphase I, 7 II+2 I. 33, anaphase II 8:8:8:8. 34-35, *C. mysorens*. 34, metaphase I, 8 II. 35, anaphase I 8:8. 36-38, *C. ochroleuca*. 36-37, metaphase I, 8 II. 38, anaphase I 8:8. 39-41, *C. pallida*. 39-40, diakinesis, 2 II+12 I. 41, telophase II showing lagging chromosomes. 42-43, *C. rhodesiae*. 42, diakinesis, 8 II. 43, anaphase I 8:8. 44-45, *C. saltiana*. 44, diakinesis, 8 II. 45, metaphase I, 8 II. B chromosomes and localized chiasmata are marked.

sixteen bivalents (Figs. 55, 56), out of which on the average 13.60 were ring and 2.40 were rod bivalents. Previous authors have also reported the same gametic number (Turner and Irwin 1961, Chennaveeraiah and Patil 1972).

Size of the bivalents

On the basis of overall size of the bivalents in a complement, 23 diploid species could be more or less categorized into three groups viz. large, medium and small sized bivalents. *C. saltiana*, *C. rhodesiae* had large; *C. grandibracteata*, *C. mucronata*, *C. alata*, *C. spectabilis*, *C. brachystachya*, *C. grahamiana*, *C. australis*, *C.*

Figs. 46-57. 46, metaphase I in *C. sericea*, 8 II. 47-48, metaphase I in *C. spectabilis*, 8 II. 49, metaphase I in *C. striata*, 8 II. 50-51, *C. longirostrata*. 50, diakinesis, 16 II. 51, anaphase I 16:16. 52-54, *C. sagittalis*. 52, diakinesis, 16 II. 53, metaphase I, 16 II. 54, anaphase I 16:16. 55-57, *C. stipularia*. 55-56, diakinesis 16 II. 57, anaphase I 16:16.

incana, *C. sericea*, *C. agatiflora*, *C. medicaginea*, *C. laburnifolia*, *C. ochroleuca*, *C. leioloba* and *C. maxillaris* had medium and *C. grantiana*, *C. leubnitziana*, *C. mysorensis*, *C. pallida*, *C. striata* and *C. falcata* had small sized bivalents. Among the three tetraploid species, *C. stipularia* had medium while *C. sagittalis* and *C. longirostrata* had small sized bivalents. However, it may be mentioned here that the gradation between large, medium and small size classes was not as distinct in some species as it was in others.

Chiasma frequency

Number of chiasmata per cell ranged from 8–13, 11–20, 10–24, 12–17, 11–17, 13–20, 11–22, 10–23, 12–18, 14–20, 13–18, 10–16, 12–17, 13–19, 7–17, 13–19, 11–21, 2–11, 13–18, 14–20, 14–24, 16–21, 14–21, mean number being 10.08, 15.72, 19.08, 13.68, 14.20, 15.40, 15.20, 17.08, 15.04, 16.60, 15.32, 12.96, 14.20, 15.88, 13.08, 15.36, 13.88, 5.84, 15.0, 16.28, 17.88, 18.0, 16.68, in *C. incana*, *C. agatiflora*, *C. alata*, *C. australis*, *C. brachystachya*, *C. falcata*, *C. grahamiana*, *C. grantiana*, *C. grandibracteata*, *C. laburnifolia*, *C. leioloba*, *C. leubnitziana*, *C. maxillaris*, *C. medicaginea*, *C. mucronata*, *C. mysorensis*, *C. ochroleuca*, *C. pallida*, *C. rhodesiae*, *C. saltiana*, *C. sericea*, *C. spectabilis* and *C. striata*, respectively, out of which on the average 8.96, 13.48, 14.92, 12.88, 13.32, 13.64, 14.60, 14.16, 14.0, 15.44, 15.04, 12.56, 13.0, 15.12, 12.88, 15.08, 13.0, 5.76, 14.40, 14.92, 15.68, 15.96, 14.84, were terminalized, giving terminalization coefficient of 0.88, 0.86, 0.78, 0.94, 0.93, 0.93, 0.96, 0.82, 0.93, 0.93, 0.98, 0.96, 0.91, 0.95, 0.98, 0.98, 0.94, 0.99, 0.96, 0.88, 0.87, 0.88, 0.88, respectively (Table 2). Similar values in the tetraploid taxa, *C. longirostrata*, *C. sagittalis* and *C. stipularia* were 24–33, 28–37, 27–37; 29.44, 32.56, 31.60; 28.32, 30.28, 29.48; 0.96, 0.92, 0.95, respectively.

In the species presently investigated, non-random distribution of chiasmata was one of the important features observed. As is clear from the figures, the chiasmata were generally confined to short regions near the non-centric ends of the chromosomes. Such distal localization seems to be a characteristic feature in many species. Usually not more than two chiasmata were observed in a bivalent. Some typical configurations (V or X) of rod bivalents, indicative of non-random distribution of chiasmata were observed in *C. brachystachya*, *C. ochroleuca*, *C. saltiana* and *C. longirostrata* (Figs. 10, 11, 36, 37, 44). Similarly ring bivalents apparently with distally localized chiasmata were observed in many species.

B chromosomes

During the present investigation Feulgen positive B chromosomes were recorded in six species, namely *C. grantiana*, *C. australis*, *C. grahamiana*, *C. leioloba*, *C. leubnitziana* and *C. mucronata* (Figs. 8, 9, 17, 19, 20, 24, 27, 31, 32). In *C. grantiana*, *C. grahamiana* and *C. mucronata* one or two B chromosomes were encountered in a few cells, while in *C. australis*, *C. leioloba* and *C. leubnitziana* no cell was recorded with more than one B chromosome. Majority of the cells in the six species were, however, without B chromosomes. *C. mucronata* had comparatively larger B chromosomes and they showed pairing affinity (Fig. 31). Quite often, B chromosomes were found lying outside the metaphase plate (Fig. 9). At anaphase I, the B chro-

mosome/chromosomes moved as such, to one of the two poles (Figs. 18, 26, 29) or to two respective poles. In *C. mucronata*, in paired condition, they were found to lag behind at anaphase II (Fig. 33). At this stage, some cells showed a bridge like structure which might have possibly resulted from the attenuation of paired B chromosomes. B chromosomes did not have any perceptible effect on the morphology, meiotic behaviour and pollen fertility.

Gupta and Gupta (1977) have also reported B chromosome (0-3) in the root tip cells of *C. brownei*, *C. juncea*, *C. medicaginea*, *C. retusa*, *C. sericea* and in pollen mother cells of *C. brownei* and *C. retusa*.

Anaphase I, II and telophase I, II

In all the present 26 species, anaphase I had equal distribution (7:7, 8:8, 16:16) of chromosomes at the poles (Figs. 3, 5, 12, 15, 18, 26, 29, 35, 38, 43, 51, 54, 57). Late disjunction of bivalent/bivalents at anaphase I/telophase I and/or lagging chromosomes (chromatids) at anaphase II/telophase II were very uncommon and were observed only in a few cells in *C. sericea*, *C. incana*, *C. mucronata*, *C. pallida* and *C. sagittalis* (Figs. 41, 54).

Chromatin bridges and acentric fragments were present in three species namely *C. sericea*, *C. mysorensis* and *C. incana*. While in *C. mysorensis* and *C. incana* not more than one bridge-fragment per cell was observed, in *C. sericea* the range was from 1-3 per cell. In *C. sericea*, 92 out of the 125 clear meiotic anaphases analysed, revealed various types of configurations for paracentric inversions which have been described elsewhere (Verma *et al.* 1981).

Chromatin bridge/bridges without acentric fragment/fragments were encountered in a few cells of *C. grahamiana*, *C. incana* and *C. pallida* at anaphase I/anaphase II.

The perusal of earlier literature on the study of anaphase I, II and telophase I, II in some *Crotalaria* species shows that many abnormalities like precocious division of univalents, late disjunction of bivalents, lagging of chromosomes, chromatin bridges with or without fragments, unequal distribution of chromosomes, non-orientation, stickiness, micronuclei and three or more than four groups of chromosomes at anaphase II have been reported in *C. incana*, *C. striata*, *C. quinquefolia*, *C. usaramoensis*, *C. maxillaris*, *C. lanceolata*, *C. burhia*, *C. medicaginea*, *C. verrucosa*, *C. sericea*, *C. retusa*, *C. astragalina*, *C. juncea*, *C. saltiana*, *C. species* and *C. species* (Magoon *et al.* 1963, Datta and Biswas 1962, Bhaumik 1975, Patil and Chennaveeraiah 1975, Roy and Sinha 1958). Magoon *et al.* (1963) have also reported cells (18.97-53.21%) with unbalanced tetrads in *C. incana*, *C. striata*, *C. quinquefolia*, *C. usaramoensis*, *C. maxillaris*, *C. lanceolata*, *C. burhia*, *C. medicaginea*, *C. verrucosa*, *C. sericea*, *C. retusa*, *C. astragalina*, *C. species* and *C. species*.

Pollen stainability

All, but two species, had 75-100% stainable pollen. The pollen stainability in *C. pallida* and *C. sericea* was 45 and 60 per cent respectively.

Discussion

The present data on the male meiosis in 26 species together with the earlier data revealed that the gametic numbers recorded so far in *Crotalaria* are $n=7$, 8, 16 and 21. While compiling the above information no cognizance has been taken of the aberrant cells like 8 II+4 I, 9 II and 7 II+1 I observed in *C. usaramoensis*, *C. verrucosa* and *C. incana* (Datta and Biswas 1962, Patil and Chennaveeraiah 1975). Similarly the inconsistent number ($n=10$) reported by Datta (1933) in *C. juncea* has been left out of consideration. 57 out of 64 species studied so far have $n=8$. This is followed by $n=16$ in 6 species and $n=7$ in one species. Besides, two gametic numbers in the same species have been reported in *C. ferruginea* ($n=8$, 21) and *C. medicaginea* ($n=8$, 16). Taking mitotic data also into consideration it becomes clear that 125 (90.5%) out of 138 species have $n=8$, $2n=16$, followed by $n=16$, $2n=32$ in 9 (6.5%) and $n=7$, $2n=14$ in 4 (2.8%) species. Based on $n=8$, two somatic numbers ($2n=16$, 32) have been reported in *C. paulina*. The inconsistent somatic number ($2n=20$) as reported in *C. falcata* and *C. dissitiflora* by Datta and Mondal 1969, Datta and Neogi 1970 has been left out of the consideration.

There exist three divergent views regarding the basic number of the genus *Crotalaria*. Wanscher (1934) has suggested $x=4$, and Bhaumik (1975) has suggested $x=7$ as the true basic number. While a number of workers including Senn (1938), Atchison (1950), Magoon *et al.* (1963), Patil and Chennaveeraiah (1975), Chennaveeraiah and Patil (1973) and Gupta and Gupta (1978a, b), however, have proposed 8 as the true basic number in the genus. The details regarding the basic number have been discussed earlier (Raina and Verma 1979, Verma and Raina 1980). Present authors support the view of Senn (1938), Atchison (1950), Magoon *et al.* (1963), Patil and Chennaveeraiah (1975), Chennaveeraiah and Patil (1973) and Gupta and Gupta (1978a, b) regarding 8 as the true basic number of the genus *Crotalaria*. The reasons for its validity are that this number is not only found in majority (90.5%) of the species analysed cytologically so far, but is found in the taxa that are not only morphologically distinct but are widely distributed in tropics and subtropics of the world. Furthermore, all but one, polyploids recorded so far are based on this number.

The plants representing *C. incana*, *C. agatiflora*, *C. alata*, *C. australis*, *C. brachystachya*, *C. falcata*, *C. grahamiana*, *C. grantiana*, *C. grandibracteata*, *C. laburnifolia*, *C. leioloba*, *C. leubnitziana*, *C. medicaginea*, *C. mysorensis*, *C. ochroleuca*, *C. rhodesiae*, *C. saltiana*, *C. sericea*, *C. spectabilis*, and *C. striata* were characterized by the presence of 7 (*C. incana*) or 8 bivalents in all the cells at diakinesis/metaphase I (Table 1). In comparison, a few cells in *C. maxillaris* and *C. mucronata* had 1-4 univalents, which behaved normally at anaphase I leading to organized and/or equal distribution. The occurrence of high frequency of cells having eight bivalents in these very species suggests that such univalents might not be due to complete lack of homology. Their presence might be attributed either to early separation of synapsed homo/homologues with or without formation of chiasmata or precocious separation of rod bivalents. Such behaviour will convert bivalents to pairs of univalents which move to the poles.

As is evident from Table 1, *C. pallida* was an exception in having 0–12 univalents per cell, the average being 6.24. The presence of consistently large number of univalents in *C. pallida* could be due to either partial failure of pairing (partial asynapsis) or else, due to the failure of chiasma formation following normal pairing (desynapsis). The exact nature cannot be made out with certainty in the absence of pachytene studies. However, at diakinesis, univalents were seen in close proximity than expected on the basis of random arrangement (asynapsis), implying thereby that the univalents might have been produced from the failure of chiasma formation (desynapsis). Various causes such as drastic temperature fluctuations, ageing, water content, humidity, soil conditions, apomixis, loss of chromosome pair, structural and numerical changes of chromosomes and gene mutation (Prakken 1943, Sybenga 1972) have been suggested for causing desynapsis/asynapsis.

Except in *C. pallida* where chiasma frequency was much lower due to prevalence of on the average 6.24 univalents and *C. incana* where the diploid number was less by two chromosomes, the highest chiasma frequency in diploids was recorded in *C. alata* (19.08) and the lowest in *C. leubnitziana* (12.96). All other species had values between these two extremes (Table 2). Majority of the species had in general 1 or 2 chiasmata in the bivalents and these chiasmata were usually located at the distal ends of the chromosomes. Such non-random distribution, seen even at very early stages of meiosis, attributes to the formation of chiasmata in specific areas of the chromosomes. The occurrence of localization of chiasmata, which prevents some chromosome segments from recombining and thus keeping special gene combinations intact, seems to be a strong feature in the genus *Crotalaria*, as has been observed in *C. intermedia* by Sybenga (1960). The presence of localized chiasma, proximal or distal, found in large number of plants (John and Lewis 1965, Sybenga 1972), has been attributed to the course of chromosome pairing, interference pattern or availability of only short segments for pairing of the chromosomes. Among the three tetraploid species, highest chiasma frequency was noticed in *C. sagittalis* (32.56) followed by *C. stipularia* (31.60) and *C. longirostrata* (29.44). The bivalents generally had one or two distally placed chiasmata.

Abnormalities at anaphase I and subsequent stages, like bridges with or without fragments and lagging bivalents/chromosomes, were present in 5 species only. The bridges without fragments at anaphase I may be because of breakage and subsequent sub-chromatid exchanges between either sister or non-sister chromatids.

The incidence of polyploidy in *Crotalaria* appears to be very low. So far, polyploidy has been reported in only 12 species and all, but one, are tetraploids ($2n=32$), based on the basic number 8. The remaining one species (*C. ferruginea*) is a hexaploid ($2n=42$) based on the basic number 7. In *C. ferruginea*, *C. humifusa*, *C. medicaginea*, *C. nitens*, *C. pumila*, *C. stipularia*, *C. sagittalis* and *C. longirostrata*, the chromosome associations are largely confined to bivalents and multivalents are uncommon. On the basis of chromosome associations all natural polyploids seem to have allopolyploid or segmental allopolyploid origin.

It is clear from the foregoing pages, that non-random distribution of chiasmata, reduced chiasma frequency, cryptic structural hybridity and paracentric inversion have played an important role in the evolutionary process of the genus. Much of

the evolution in the genus, however, appears to have taken place through gene mutations at diploid level. Polyploidy does not seem to have played a significant role.

Summary

Detailed male meiosis was studied in 26 species. Twenty three species were diploid ($n=7, 8$) and three were tetraploid ($n=16$). Various evidences show that basic number of the genus is eight.

Twenty diploid species had either 7 (*C. incana*) or 8 bivalents at diakinesis or metaphase I. The occurrence of 0–4 univalents in a few cells in *C. maxillaris* and *C. mucronata* has been ascribed to early separation of synapsed homo/homoeologues and/or precocious separation of rod bivalents. The presence of 0–12 univalents in *C. pallida* (6.24/cell) has been attributed to asynapsis/desynapsis. Complete bivalent formation in three tetraploid species indicates their allopolyploid origin. The occurrence of non-random distribution of chiasmata in many species seems to be an important genetic factor. B chromosomes were recorded in 6 species.

Only in a few species abnormalities like late disjunction of bivalent/bivalents, lagging chromosomes, chromatin bridges with/without fragments were observed at anaphase I, II/telophase I, II.

It has been concluded that apart from gene mutations, important genetic factors like non-random distribution of chiasmata, reduced chiasma frequency, cryptic structural hybridity and paracentric inversions have played an important role in the evolution within the genus. Polyploidy has played a minor role.

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