Karyotype Analysis in 15 Varieties of *Cajanus cajan* (L.) Millsp and *Autylosia lineata* (W. and A.)

M. P. Shrivastava, D. Sharma and Laxman Singh¹

Department of Plant Breeding and Genetics
Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur
Madhya Pradesh, India

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Cajanus cajan (L.) Millsp commonly referred to as pigeon pea, red gram, arhar or tur, is an important pulse crop in India. In spite of having only one cultivated species in the genus, the crop exhibits high amount of diversity in various morphological characteristics and different cultivars have come in existence in different agroclimatic regions of the country. There is very little information on the cytogenetic aspects of variability in different qualitative and quantitative characters, which is highly essential for planned plant breeding efforts for the development of improved varieties. Therefore work has been initiated to determine linkage relationship of some important quantitative and qualitative characters through the use of trisomic lines which is in progress. For the identification of appropriate trisomic stock, information on the karyomorphological characteristics in the species is highly essential. The present work therefore was undertaken to get the information on these aspects in morphologically diverse cultivars. Also study of the karyomorphology of *Autylosia lineata* was undertaken for comparison as it seems to be the only related genera and the species which easily crosses with arhar and attempts in past were made to exploit its resistance to wilt disease, Kumar *et al.* (1958).

Materials and methods

Fifteen types of pigeon pea, representing a wide range of variability in several agronomic characters were selected for the study. The characteristic features of the varieties used in the study are given in Table 1. Also seeds of *Autylosia lineata* were used for this study. For cytological studies seeds of these strains were germinated on moist filter papers in petri dishes at room temperature (32°C). Healthy root tips of 1 cm length were cut and pre-treated with α -bromonephthalene for 1 hour at 15°C. Number of chemicals were tried for pre-treatment such as hydroxyquinoline, paradichlorobenzene and α -bromonephthalene, but latter gave satisfactory mitotic preparations. After treatment, the root tips were thoroughly washed with distilled water and fixed in Carnoy's solution (9:1:3) for 22 hours. For preparation of slides root tips were hydrolized in 1 N HCl at 60°C for 7 minutes and kept in Feulgen solution. After half an hour, smear preparations in

¹ Present address: Professor and Head, Department of Plant Breeding and Genetics, College of Agriculture, Palampur (H.P.), India.

Table 1. Characteristics of the varieties of pigeon pea used in karyotype analysis

S. no.	Cultivars	Maturity in days	Plant height in cm	Plant width in cm	100 seed weight in gm	Flower colour	Pod colour	Seed colour
1.	T. 21	142	240	114	7.3	Yellow	Streaked	Light brown
2.	No. 148	176	233	118	9.2	Yellow	Streaked	Dark brown
3.	R. 3	249	291	86	13.6	Yellow	Green	Light brown
4.	NP (WR) 15	242	261	94	7.6	Yellow	Streaked	White
5.	S. 5	153	170	99	8.7	Yellow	Streaked	Brown
6.	NP. 69	248	226	78	16.2	Reddish Yellow	Streaked	Brown
7.	Baigani	131	239	97	11.8	Yellow	Streaked	Brown
8.	Perennial type		250	90	15.0	Yellow	Dark	Brown
9.	P. 4839	179	—	—	10.0	—	—	—
10.	Gwl. 3	246	277	129	6.7	Yellow	Streaked	Brown
11.	R. 7	247	265	77	—	Yellow	Dark	Brown
12.	P. 958	180	190	60	—	Yellow	Streaked	Brown
13.	P. 1136	239	232	51	—	Reddish Yellow	Streaked	Black
14.	P. 2327	155	241	61	—	Yellow	Green	Brown
15.	P. 1714	165	236	53	—	Yellow	Streaked	Brown

aceto-orcin 1% were made and a gentle pressure applied on the cover slip with pencil to get good spread and complete chromosome set. Intact cells, having complete set of chromosomes were photographed and analysed. Measurements of chromosomes were carried out from the microphotographs of 5 cells from each variety with the help of millimeter scale to the nearest 0.1 millimeter, using a fine pair of dividers and converted to microns. Microphotographs were taken by Olympus research microscope camera at $1500 \times$ (15 $\times$ 100 oil). The gap in the primary constriction was not taken into account in different measurements. The mean of five measurements of each chromosome was taken to express the results. Arm ratio was calculated from the ratio of the length of the long arm to that of short arm. Chromosome size was expressed by the total length of the long and short arms. The ratio between the longest and the shortest chromosome was also obtained to depict the variation in chromosome morphology.

Results and Discussion

Twenty-two chromosomes were found in all the somatic cells examined in 15 varieties of *C. cajan* and *Autylosia lineata* and no accessory chromosomes were observed in any of the preparations (Fig. 1A and 1B) as reported by Krishnaswami and Ayyangar Rangaswamy (1935).

Detailed measurements of the chromosomes (I to XI) with their classification in groups (median, submedian and subterminal) for 15 varieties of pigeon pea and *Autylosia lineata* are given in Table 2. Figure 1B gives the microphotographs of the designated I to XI chromosomes for 15 varieties of pigeon pea. One pair of satellited chromosome was observed in 13 varieties out of 15. One variety P. 958 did not have the satellited chromosome. This variety is morphologically different from all other varieties by having obovate shape of trifoliates (it is a simple leaf mutation caused by a single recessive gene as reported by Deshpande and Jeswani (1952). Another variety NP. 69 had a heteromorphic pair, with one member being satellited only. The variation was also observed in the total chromatin length among the varieties, ranging from 27.6 ± 0.142 microns in variety T.21 to 44.92 ± 0.795 in variety R.3. The satellite length was 0.33 microns in 10 varieties, 0.50 in two varieties and only 0.16 in two varieties, while it was totally absent in one variety. The arm ratio (Table 3) for each chromosome in different varieties varied to varying degree. On the basis of arm ratio the chromosomes were catagorised as median (0.76–1.00), submedian (0.51–0.75) and subterminal (0.25–0.50). In all the varieties (Table 3), chromosome I was grouped as median. Also chromosome III was median in all except in S.5 and NP.69, while other chromosomes did not show uniform pattern and were grouped differently in different varieties. The ratio of the longest and the shortest chromosome in the cultivars of pigeon pea ranged from 1.52 to 3.03 and it was 1.66 in *Autylosia*. Karyomorphology of *Autylosia* chromosomes (Fig. 2) resembled to that of variety T.21 of pigeon pea. It had total chromatin length of 26.36 against 27.60 for T.21. Similarly its grouping of chromosome in median, submedian a subterminal resembled T.21 except for variation in position of VI, II and IX chromosomes. The satellite length is

Table 2. Classification and measurements of chromosome complements in fifteen pigeon pea varieties and *Autylosia lineata*

S. no.	Cultivars	Arm ratio			Satellite length in micron	Average total length in microns	S.E.	Longest/shortest chromosome ratio
		Median 0.76—1.00	Sub-median 0.51—0.75	Sub-terminal 0.25—0.50				
1.	T. 21	I III IV V VI VII VIII X	IX	II XI	0.33	27.60	± 0.142	1.67
2.	No. 148	I III IV V VI VII VIII IX	X XI		0.33	35.16	± 0.257	1.99
3.	R. 3	I II III IV VII VIII IX	V	VI X XI	0.33	44.92	± 0.795	1.88
4.	NP (WR) 15	I II III IV VIII	VII X	V VI IX X (terminal)	0.33	43.96	± 1.574	2.33
5.	S. 5	I II IV V VI VII	III VIII IX XI		0.33	35.58	± 0.523	1.51
6.	NP. 69	I IV V	II VI VII VIII X	III IX XI	0.33	36.90	± 0.271	1.71
7.	Baigani	I II III IV VI	V VII IX X XI	VIII	0.50	35.92	± 0.374	1.71
8.	Perennial type	I II III IV V VII X	VI VIII IX X		0.33	35.78	± 0.939	1.52
9.	P. 4839	I II III VII IX	X XI	IV V VI	0.33	40.60	± 0.331	2.00
10.	G. 3	I II III V VI VII IX	VIII X	IV XI	0.33	35.12	± 2.178	3.01
11.	R. 7	I III IV V VIII X	II VII IX XI	VI	0.33	41.06	± 0.309	1.60
12.	P. 958	I III IV V VI VII VIII X XI	II	IX	—	30.22	± 1.737	2.51
13.	P. 1136	I II III IV V VI VII	VIII IX X	XI	0.50	39.04	± 0.647	2.58
14.	P. 2327	I II III IV V VI VIII IX	V XI	VII	0.16	32.08	± 0.557	3.03
15.	P. 1714	I II III VI VIII IX X	IV V VII	XI	0.16	37.34	± 0.392	2.32
16.	<i>Autylosia lineata</i>	I III IV V VII VIII X	II VI	IX XI	0.33	26.36	± 0.848	1.66

Table 3. Chromosome arm ratios of different varieties of pigeon pea and *A. lineata*

Name of the cultivars	Chromosome number and arm ratio										
	I	I	III	IV	V	VI	VII	VIII	IX	X	XI
1. T. 21	1.00	0.49	1.00	0.75	0.75	0.75	0.75	1.00	1.00	0.50	0.39
2. No. 148	1.00	0.50	0.75	0.79	1.00	0.83	0.83	1.00	0.83	0.49	0.43
3. R. 3	1.00	1.00	0.79	1.00	0.62	0.44	0.87	0.87	0.87	0.50	0.37
4. NP (WR) 15	1.00	1.00	1.00	0.83	0.49	0.44	0.62	1.00	0.39	0.75	0.22
5. S. 5	1.00	1.00	0.60	0.79	0.79	1.00	1.00	0.66	0.56	0.71	0.60
6. NP 69	0.75	0.60	0.50	0.79	0.79	0.56	0.56	0.71	0.49	0.71	0.37
7. Baigani	0.75	0.75	1.00	0.95	0.66	1.00	0.66	0.43	0.83	0.71	0.60
8. Perennial type	1.00	1.00	1.00	1.00	0.79	0.66	1.00	0.71	0.71	1.00	0.60
6. P. 4839	0.75	1.00	0.79	0.43	0.37	0.49	0.78	0.62	1.00	0.55	0.66
10. G. 3	1.00	1.00	1.00	0.39	0.75	1.00	1.00	0.60	0.83	0.71	0.50
11. R. 7	0.75	0.60	1.00	1.00	0.83	0.49	0.71	1.00	0.55	1.00	0.60
12. P. 958	1.00	0.66	0.75	0.75	1.00	1.00	0.79	0.79	0.50	1.00	0.79
13. P. 1136	0.75	1.00	1.00	1.00	0.79	1.00	0.83	1.00	0.87	1.00	0.37
14. P. 2327	1.00	1.00	1.00	1.00	0.79	1.00	0.43	1.00	1.00	0.71	0.60
15. P. 1714	1.00	0.75	1.00	0.60	0.60	1.00	0.71	0.86	0.86	1.00	0.28
16. <i>Autylosia lineata</i>	1.00	0.66	1.00	1.00	1.00	0.75	0.79	1.00	0.43	1.00	0.50

0.33 as generally observed for pigeon pea. *Autylosia* had longest to shortest chromosome ratio of 1.66 which was close to 1.67 of T.21 pigeon pea.

Fig. 1A. Microphotographs of mitotic chromosome complements ($2n=22$) of 15 strains of *Cajanus cajan* (L.) Millsp. (Table 1 gives details of the fifteen strains).

The karyotype analysis of the varieties of pigeon pea revealed considerable intervarietal variations in chromosome complement of the species in regard to arm ratio, total length and ratio of longest and shortest chromosome. This varia-

Fig. 1B. Photographic karyogram showing 11 pairs of chromosomes in 15 strains of *Cajanus cajan* (L.) Millsp.

tion, however, cannot be universally associated with any pattern of variation in the morphological and agronomic characters of the varieties. Though the absence of satellite in the variety P.958, also carrying the distinct character of leaf mutation cannot be overlooked. Similarly the tallest variety with height of 291 cm and

Figs. 2A–2B. 2A (above), microphotograph of mitotic chromosome complement ($2n=22$) *Autylosia lineata* (W. and A.) Approx. $\times 4500$. 2B (below), photographic karyogram showing 11 pairs of chromosomes in *Autylosia lineata* (W. and R.).

characteristic narrow spread, also possessed highest value of total chromosomal length of 44.92 microns. These associations appear interesting and do indicate that variability in morphological characters observed among the cultivars of *Cajanus cajan* is expressed to some extent through variation in the karyotype. The present study, however, reveals that the varieties studied had some distinctive features of their karyotype, just as they have distinguishing features in their morphological characteristics.

The chromosome morphology of *Autylosia lineata* closely resembled the variety T.21 of pigeon pea in the present study. This indicates that the differences observed by Kumar *et al.* (1958) in

karyomorphology of *Autylosia lineata* and the pigeon pea are specific to the particular variety studied by them. Also it appears that there are no major differences in karyomorphology of a large group of pigeon pea varieties and the closely related genera *Autylosia* which can easily be crossed with pigeon pea. The wide variation in the pigeon pea and *Autylosia lineata* might have generally resulted from gene mutations rather than from gross changes in karyomorphology.

The high sterility and appearance of heteromorphic pairs between pigeon pea and *Autylosia* crosses reported by Kumar *et al.* (1958) could be reduced if the variety T.21 is used which closely resembles *Autylosia* in its karyomorphology. On the basis of above it may be concluded that the variation in inter-generic cross between *Cajanus* and *Autylosia* will greatly depend on the variety of *Cajanus* used in crosses.

Therefore, it may be suggested that in breeding programmes where *Cajanus* and *Autylosia* crosses are being exploited the work should not be confined to a single variety rather several varieties of *Cajanus* should be used in crossing with *Autylosia*.

Summary

Fifteen varieties of *Cajanus cajan* (L.) Millsp differing in morphological characteristics and *Autylosia lineata* W. and A. were studied for karyomorphology. Twenty-two chromosomes were found in all the varieties of *Cajanus cajan* and *Autylosia lineata*. Considerable inter varietal variation in chromosome complement was observed, in regard to arm ratio, total length etc. Variability in morphological characters observed among the cultivars of *Cajanus cajan* is expressed to some extent through variation in its karyotype. One pair of satellited chromosome was observed in 13 varieties of *Cajanus cajan* and *Autylosia lineata*. A variety P.958 did not have satellited chromosome, while variety NP.69 had a heteromorphological pair with one member being satellited only. Variety T.21 had shortest total chromatin length of 27.60 ± 0.142 microns against 44.92 ± 0.795 for variety R.3. It was 26.36 ± 0.848 for *Autylosia lineata*. On the basis of arm-ratio the chromosomes were catagorised as median (0.76–1.00), submedian (0.51–0.75) and sub-terminal (0.25–0.50).

Literature cited

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