Karyological Studies of Sixteen Cultivars of Tepary Bean (*Phaseolus acutifolius* A. Gray)

M. Munian and D. Subramanian

Department of Botany, Annamalai University,
Annamalainagar, 608002, India

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The cultivated species of *Phaseolus acutifolius* A. Gray has a wide range of cultivars with distinct and diffused geographical adaptability. Although the cultivars of this species are known to be diploids ($2n=22$) the detail karyotypic studies on a large number of cultivars have not been made so far to establish the standardised karyotype for this species. The cultivars taken up in the investigation are known to have wide differences in their morphology and behaviour. In view of this, a detailed analysis of the somatic chromosomes is considered necessary.

Material and method

Karyotype of sixteen cultivars of Tepary bean was studied from the root tips obtained by germinating the seeds in 1'clay pots filled up with garden soil, leaf mold and fine sand in a suitable proportion. Fresh root tips were collected at 9.30 a.m. The root tips were pretreated in 0.002% hydroxyquinoline kept at 4°C for 3 hours. After a thorough wash the root tips were then fixed in 1 : 3 acetic alcohol for 3 hours. Following iron alum haematoxylin squash technique (Mariumthu and Subramaniam 1960) root tip squashes were made and the slides sealed.

Six camera lucida drawings were taken for each cultivar, using 12.5x ocular and oil immersion objective 100x from the temporary preparations. Somatic counts and karyotypic formulae were worked out for all the 16 cultivars. The position of the centromeres was determined from a comparative study of several metaphase plates. The chromosomes are assigned to different types basing on their relative length, position of centromere and presence or absence of secondary constriction.

The abbreviations used for the purpose are M=median, Sm=submedian, St=subterminal, Smst=secondarily constricted chromosome with submedian primary constriction. Chromosome types include also the following: A=above 3.0 μm to 4.0 μm , B=above 2.0 μm to 3.0 μm , C=above 1.0 μm to 2.0 μm , D=upto 1.0 μm . The figures in the karyotype formula pertain to the number of pairs of chromosomes. TF% was calculated by the formula given by Huziwara (1962).

An average karyotype was worked out basing in the results of chromosome morphology obtained for all the 16 cultivars.

Results

The cytological study in Tepary bean showed better division from 9 a.m. to 10 a.m. at any season.

The chromosome number for all the 16 cultivars was found to be $2n=22$ (Plate

Table 1. Details of karyotype of sixteen cultivars of *Phaseolus acutifolius* A. Gray

S. no.	Name of cultivars	2n	Type of chromosome				Total chromosome length (μm)	Range in chromosome length (μm)	TF%
			Median	Submedian	Sub-terminal	Submedian with secondary constriction			
1	P.I. No. 197041 EC. No. 131740	22	6	4	1	—	40.0	1.4–2.2	34.50
2	P.I. No. 200902 EC. No. 131741	22	7	4	—	—	42.8	1.6–2.2	38.79
3	P.I. No. 201268 EC. No. 131742	22	7	4	—	—	52.4	2.0–3.0	38.55
4	P.I. No. 263590 EC. No. 131743	22	6	4	1	—	47.2	1.4–2.6	36.02
5	P.I. No. 286303 EC. No. 131745	22	7	4	—	—	27.2	0.8–1.5	37.50
6	P.I. No. 310606 EC. No. 131746	22	6	5	—	—	53.6	1.8–3.4	37.69
7	P.I. No. 318001 EC. No. 131748	22	6	5	—	—	38.0	1.4–2.2	35.79
8	P.I. No. 310803 EC. No. 131750	22	4	3	4	—	44.0	1.6–2.0	30.45
9	P.I. No. 312133 EC. No. 131751	22	6	3	1	1	41.6	1.4–3.2	35.58
10	P.I. No. 319438 EC. No. 131753	22	5	3	3	—	40.4	1.4–2.2	32.67
11	P.I. No. 319442 EC. No. 131755	22	5	4	2	—	33.2	1.0–2.0	33.74
12	P.I. No. 319444 EC. No. 131757	22	6	4	1	—	45.2	1.6–2.4	35.84
13	P.I. No. 319447 EC. No. 131759	22	7	4	—	—	41.6	1.6–2.2	38.46
14	P.I. No. 331181 EC. No. 131760	22	5	4	2	—	32.2	1.2–2.0	32.31
15	Nebraska cultivar	22	3	3	1	4	56.8	2.0–3.2	32.39
16	var. <i>latifolius</i>	22	5	2	3	1	39.2	1.2–3.0	33.16

1, Figs. 1 to 18). All the cultivars possess smaller chromosomes. Out of the total of 11 pairs there are 3 pairs of secondary constricted chromosomes in only one cultivar from Nebraska and only one pair of such chromosomes in two cultivars (var. *latifolius* and P.I. No. 312133). In all other cultivars, secondary constricted chromosomes were absent. Based on the analysis of chromosome morphology, the de-

tails of karyotype of all the cultivars are given in Table 1. The idiogram of somatic karyotypes for all the cultivars and also the idiogram of an average karyotype are here (Plate 2, Figs. 1 to 17)

Plate 1: Figs. 1-18. Plate of cultivars of *Phaseolus acutifolius*. 1 and 9, somatic prometaphase plate of cultivars P.I. No. 197041 and P.I. No. 310606. 2-8, 10-16 and 18, somatic metaphase plates of cultivars: P.I. No. 197041, No. 200902, No. 201268, No. 263590, No. 286303, No. 310606, No. 310606, No. 310801, No. 310803, No. 312133, No. 319438, No. 319442, No. 331181 and No. 331181. 17, P.I. No. 331181 shows precocious movement.

Plate 2: Figs. 1-16. Idiogram of somatic karyotypes of 16 cultivars of *Phaseolus acutifolius* A. Gray.

Fig. 17. Idiogram of an average karyotype of *Phaseolus acutifolius* A. Gray.

Discussion

All the 16 cultivars of *Phaseolus acutifolius* A. Gray are found to have $2n=22$. This agrees with the reports of Karpechenko 1925b and Shibata 1962a (vide Fedorov 1969). So, the primary base number for this species is confirmed as 11.

Table 2. Karyotype formula of sixteen cultivars

Sl. no.	Cultivar name	Karyotype formula
1	P.I. No. 197041 EC. No. 131740	3(B. Sm.)+1(C. Sm.)+6(C. M.)+1(C. St.)
2	P.I. No. 200902 EC. No. 131741	1(B. Sm.)+1(B. M.)+3(C. Sm.)+6(C. M.)
3	P.I. No. 201268 EC. No. 131742	4(B. Sm.)+6(B. M.)+1(C. M.)
4	P.I. No. 263590 EC. No. 131743	3(B. Sm.)+4(B. M.)+1(C. Sm.)+2(C. M.)+1(C. St.)
5	P.I. No. 286303 EC. No. 131745	4(C. Sm.)+3(C. M.)+4(D. M.)
6	P.I. No. 310606 EC. No. 131746	1(A. Sm.)+2(B. Sm.)+4(B. M.)+2(C. Sm.)+2(C. M.)
7	P.I. No. 310801 EC. No. 131748	1(B. Sm.)+4(C. Sm.)+6(C. M.)
8	P.I. No. 310803 EC. No. 131750	4(B. M.)+3(C. Sm.)+4(S. St.)
9	P.I. No. 312133 EC. No. 131751	1(A. Sm. St.)+1(B. Sm.)+2(C. Sm.)+6(C. M.)+1(C. St.)
10	P.I. No. 319438 EC. No. 131753	1(B. Sm.)+2(C. Sm.)+5(C. M.)+3(C. St.)
11	P.I. No. 319442 EC. No. 131755	4(C. Sm.)+5(C. M.)+1(C. St.)+1(D. St.)
12	P.I. No. 319444 EC. No. 131757	3(B. Sm.)+3(B. M.)+1(C. Sm.)+3(C. M.)+1(C. St.)
13	P.I. No. 319447 EC. No. 131759	1(B. Sm.)+2(B. M.)+3(C. Sm.)+5(C. M.)
14	P.I. No. 331181 EC. No. 131760	4(C. Sm.)+5(C. M.)+2(C. St.)
15	Nebraska cultivar	3(A. Sm. St.)+1(B. Sm. St.)+2(B. Sm.)+3(B. M.)+1(C. Sm.) +1(C. St.)
16	var. <i>latifolius</i>	1(B. Sm. St.)+2(C. Sm.)+5(C. M.)+3(C. St.)

The karyotype analyses of all the cultivars reveal reasonably significant differences with regard to chromosome length, position of primary constriction and TF%. 56.8 μm is the maximum total chromosome length and the minimum is 27.2 μm . A large number of cultivars (10 numbers) have been noted within a range

Plate. 2. Figs. 1-16. Idiogram of somatic Karyotypes of 16 cultivars of *Phaseolus acutifolius*. 1. P.I. No. 197041, 2. No. 200902, 3. No. 201268, 4. No. 263590, 5. No. 286303, 6. No. 310606, 7. No. 310801, 8. No. 310803, 9. No. 312233, 10. No. 319438, 11. No. 319442, 12. No. 319444, 13. No. 319447, 14. No. 331181, 15. Nebraska cultivar and 16. var. *latifolius*.

of total chromosome length of 38.0 μm to 47.2 μm . The study of the range in chromosome length reveals that the cultivar with P.I. No. 286303 has the shortest (0.8 μm) and the cultivar with P.I. No. 310660 has the longest chromosomes (3.4 μm) in their somatic cells as compared with other cultivars.

The striking similarities in the karyomorphology of 13 out of 16 cultivars are in their first chromosomes which are not secondary constricted and the rest of the chromosomes are found either with median or submedian or subterminal primary constrictions. An average karyotype (Plate. 2, Fig. 17) drawn by utilizing arithmetic mean of the individual chromosome of the whole somatic complement for all the varieties, however, reveals a different picture. Datta (1975) on sweet pea and Goswami (1979) on black gram, have utilized the mean values and found similar features.

The TF% for 10 cultivars is found to be in a close range of 34.50 to 38.79 and the cultivars under this range have more or less symmetrical karyotypes. Here, the median constricted chromosomes are more in number than the chromosomes with submedian and subterminal constrictions. The other 6 cultivars have shown comparatively an asymmetrical karyotypes with TF% ranging from 30.45 to 33.74. Here there are more number of submedian and subterminal constricted chromosomes when compared to the median constricted chromosomes. The karyotypes in general for different species of *Phaseolus* have been reported as symmetrical (Sen and Chheda 1958, Krishnan and De 1965, De and Krishnan 1966, Singh and Roy 1970).

The presence of secondary constriction only in one pair of chromosomes of only two cultivars and median primary constriction in a large number of chromosomes of a somatic complement for almost all the cultivars apparently suggested the ancient origin of these cultivars. Anyhow, the presence of 4 pairs of secondary constricted chromosomes coupled with the presence of submedian primary constriction in a large number of chromosomes of the somatic complement in Nebraska cultivar showed the recent origin. Goswami (1979) has stated the recent origin of the varieties of Black gram by studying the karyotypes of thirty two varieties. In absence of any evidence of chromosome aberrations in meiosis which is regular, it is difficult to ascertain the exact mechanism of origin of these karyotypic differences. In any case, these changes may have helped the process of evolution and the emergence of different varieties. On the other hand, the uniformity in the base number and considerable similarity in gross nature of the karyotypes suggest that gene mutation was the principal factor in evolution.

Summary

Karyotypic analysis of sixteen cultivars of *Phaseolus acutifolius* A. Gray showed reasonable differences between the cultivars on chromosome length, position of primary constriction and in TF%. Only in two cultivars, a pair of chromosomes was secondarily constricted and most of the chromosomes in all the cultivars, excepting the Nebraska cultivar, possess mostly medianly constricted chromosomes. In

Nebraska cultivar, there are 4 pairs of secondary constricted chromosomes and most of the chromosomes possess submedian primary constrictions. An average karyotype of *Phaseolus acutifolius* has been suggested basing in the results included herewith.

The observation of TF % ranging from 34.50 to 38.79 revealed a symmetrical karyotypes for 10 cultivars. The others 6 cultivars were observed to be asymmetrical with TF ranging from 30.45 to 33.74. The importance of karyotypic differences in evolution of varieties within a species has been discussed.

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