Chapter I

MITOSIS AND KARYOTYPE ANALYSIS IN *Passiflora incarnata* L.

MITOSIS AND KARYOTYPE ANALYSIS IN PASSIFLORA INCARNATA

INTRODUCTION

In the study of evolution, phylogeny and classification of plants, the importance of cytological study has been widely accepted. The karyotype was first defined in 1926 by Delaunay as a group of species resembling each other in the morphology and number of their chromosomes. However, Lewitsky (1931) defined it as a phenotypic appearance of the somatic chromosomes in contrast to their genotype. The term karyotype by definition, implies morphological expression of somatic chromosomes and an 'idiogram' on its diagramatic representation.

It reveals from the previous studies that the genus Passiflora is poorly understood from the standpoint of cytology. The literature in this connection has already been reviewed under 'Review of Literature'.

MATERIALS AND METHODS

The P. incarnata were grown in the Botanical gardens of Botany Department, Shivaji University, Kolhapur.

The somatic chromosome number was determined from the root tips. Excised root tips were washed and treated with 0.2 % colchcine at 7°C for 2-1/2 hours, washed thoroughly and

fixed in Carnoys' fluid for 4-12 hours and stored in 70 % alcohol.

Such pretreated and fixed root tips were washed again and gently heated in a mixture of 2 % aceto-orcein and 1 N HCl over a spirit flame for a few seconds and squashed in 2 % aceto-orcein. Slides were made permanent following the butyl alcohol and acetic acid series method and using DPX as the mountant. For determining the length of the chromosomes, 5 plates were studied and the average length of each individual chromosome was calculated from the data obtained. For the karyotype analysis the method of Levan et al.(1964) has been followed.

The drawings were made with Camera Lucida at X1500 by using oil immersion. Photomicrographs of metaphases were taken under light microscope (X1000).

Mitosis : At the end of the prophase the chromosomes attained their greatest contraction. At early metaphase the nucleolus was prominent. At metaphase, the chromosomes were lying at the equator of the spindle which was of normal shape. It was found that the mitosis in P.incarnata was normal.

Karyotype : For the karyotype analysis of long and short arm was denoted as 'l' and 's' respectively. 'c' was the total length of the chromosome. The location of the centromere was expressed as a difference d = l-s. The ratio of

long and short arm of the chromosome was denoted by 'r'. The centromeric index (i) was calculated as,

$$i = \frac{100.s}{c}$$

The somatic chromosome number of P.incarnata is determined as 2n = 18 (fig.1.1). The length of chromosomes varied from 4.80 μ to 1.56 μ. Chromosomes are idiogrammed in Fig.1.2. The details regarding length of chromosomes, position of centromeres etc. are given in Table 1.1 which indicates that the chromosomes can be classified into the following 5 types :

Type A : (Chromosome I) :

A pair of long chromosomes (4.80 μ) with submedian centromere and satellite on the long arm. This is the longest pair.

Type B : (Chromosomes II to IV) :

Three pairs of long chromosomes (3.76 μ - 3.27 μ) with submedian centromere.

Type C : (Chromosomes V and VII) :

Two pairs of median chromosomes (2.96 μ and 2.50 μ) with submedian centromere.

Type D : (Chromosomes VI and VIII) :

Two pairs of short chromosomes (2.84 μ and 2.41 μ) with median centromere.

Type E : (Chromosome IX) : A pair

A pair of very short chromosomes (1.56 μ) with median centromere. This is the shortest pair.

Fig. 1.1 Somatic chromosomes from a root tip cell showing 2n = 18 (X1500).

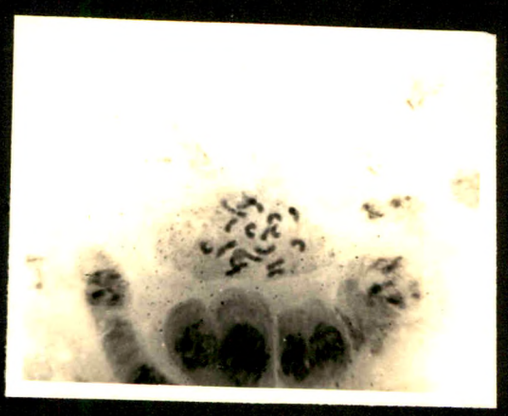

FIG. 1.1

Fig. 1.2 Idiogram of the somatic complement of *Passiflora incarnata* L.

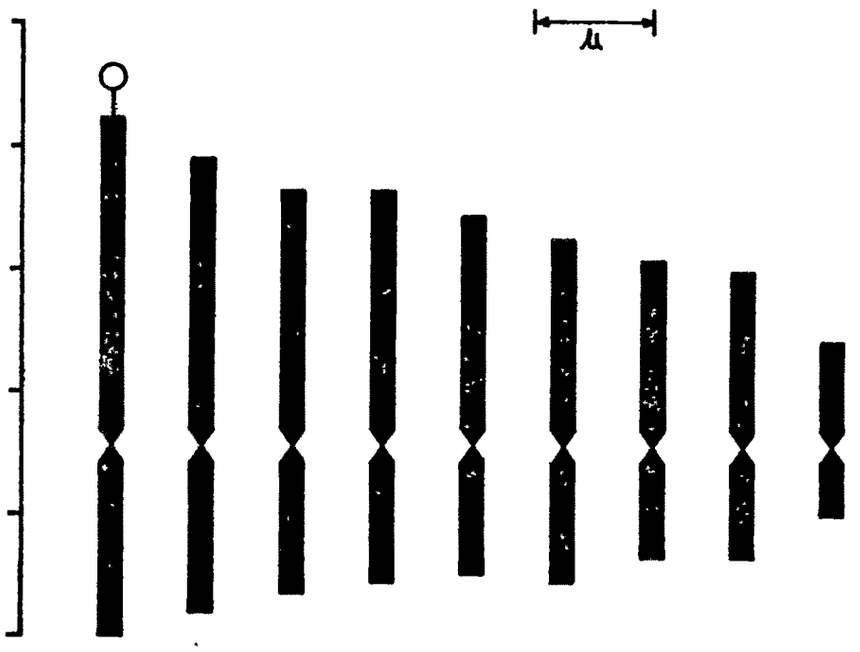

Fig 1-2 - IDIOGRAM OF THE SOMATIC COMPLEMENT OF Passiflora incarnata L.

Table 1.1 : Measurement and position of centromere of somatic chromosomes in P.incarnata

Chromosome pair	Length of the long arm in μ	Length of the short arm in μ	Total length in μ	d value	r value	i value	Centromeric position
I	2.72 + .50 sat.	1.58	4.80	1.64	2.03	32.93	Sm
II	2.39	1.37	3.76	1.02	1.74	36.39	Sm
III	2.13	1.19	3.32	0.94	1.79	35.84	Sm
IV	2.14	1.13	3.27	1.01	1.89	34.56	Sm
V	1.93	1.03	2.96	0.90	1.87	34.79	Sm
VI	1.73	1.11	2.84	0.62	1.52	39.98	m
VII	1.58	0.92	2.50	0.66	1.71	36.88	Sm
VIII	1.48	0.93	2.41	0.55	1.59	38.60	m
IX	0.98	0.58	1.56	0.40	1.69	37.18	m

The karyotype formula for P.incarnata can, therefore, be represented as :

$$K\ (2n)\ :\ 18\ :\ 2\ A^{sm} + 6\ B^{sm} + 4\ C^{sm} + 4\ D^{m} + 2\ E^{m}$$

DISCUSSION

The diploid chromosome number 2n = 18 for P.incarnata determined in the present investigation is in conformity with that reported by Heitz (1927), Bowden (1945) and Storey(1950).

Present study shows that P.incarnata possesses symmetrical Karyotype. The position of centromere varies from median to submedian. Beal (1972) when studying cytology of the native Australian and several exotic Passiflora species has also found that chromosomes are with median and submedian centromeres. Dixit (1979) has also found the symmetrical karyotype with median and submedian centromeres but there was a slight difference in the karypotype formula of P.incarnata. He has found submedian centromeres in VIth and VIIIth chromosome pairs but in present investigation they are median. According to Stebbins and Levitsky's concept of the karyotype evolution, the higher percentage of metacentric chromosomes indicates primitiveness of a species, which is not true in P.incarnata.

The author has come across with only one type of a satellited chromosomes with satellite on the long arm in the present investigation. Dixit (1979) has also found satellite on the long arm in P.incarnata. Beal (1972) has

reported 2 types of satellited chromosomes in P.maliformis and 5 types in P.quadrangularis and P.seemanni. Steobins (1950) stated that in most of the diploid plants only one pair of satellite is found. The occurrence of 2n = 18 species in the euploid interspecific series 2n = 12, 18, 24 is the main evidence of Storey's (1950) suggestion that the ancestral basic chromosome number in the genus was x = 3 rather than x = 6. However, the 2n = 18 species may be aneuploid derivatives of a 2n = 24 type (rather than of hexaploid origin) and this hypothesis still conforms with a basic number of x = 6 for the genus. If x = 3 is the basic number, the 2n = 84 race of P.lutea is an extremely high ploid. Further, the number 2n = 6 has not been recorded and if such species (necessary to a postulated basic number of x = 3), ever existed, they have disappeared from the genus or are very uncommon. Evidence from chromosome numbers of Passiflora species, of species in related genera and from related families (Darlington and Wylie,1955; Hutchinson, 1959) indicates that it is probably x = 6.