Pachytene Analysis in *Solanum viarum*

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Received February 22, 1975

McClintock in 1929, was the first to make a detailed study of pachytene in maize. Since then a number of workers have carried out pachytene analysis in several plants and the data have been used for the morphology of chromosomes and phylogeny of plants. The available literature indicates that no study on the pachytene analysis of *Solanum viarum* Dunal has been made so far. The present paper deals in detail with pachytene chromosomes of this species.

Material and methods

The seeds of *Solanum viarum* Dunal were obtained from the Botanical Survey of India, Calcutta and the plants were grown in the Botanical Garden of the Department of Botany, Nagpur University, Nagpur. Anthers of these plants were fixed in acetic alcohol (3:1). Smears were made in 2% propionic-orcein. Camera lucida sketches of the pachytene chromosomes were drawn from temporary slides. The slides were afterwards made permanent. The drawings of chromosomes were ten times enlarged with the pantograph and then measured with the rotameter.

Eleven well spread pachytene stages were scored for the detailed study. The criteria applied for the morphological differentiation of the chromosomes were total length (actual length in microns), relative length (absolute length expressed in percentage of the total complement length) and centromere index.

The centromere index = $100S/C$, where S is the length of the short arm and C is the total length of chromosome.

Observations

The somatic chromosome number of this plant is 24. Chromosomes are quite long and studded with micro-chromomeres which are very distinct. Closely placed and heavily stained micro-chromomeres are identified with the heterochromatic region. The centromeres are not easily discernable. However, they can be spotted out as small gaps flanked on either side by the heterochromatic regions. The standard karyotype is represented in Fig. 2 and data are given in Table 1. The distinguishing morphological features of each of the twelve chromosomes in the haploid complement are described below.

Chromosome I is the longest chromosome of the complement. It is distinguished by the presence of darkly stained heterochromatic region at the distal end of the long arm. It measures nearly one third part of the latter. The part of the heterochromatic region of the short arm flanking the centromere is thrice in length of its

Fig. 1. Pachytene chromosomes. $\times 1500$.

Fig. 2. Idiogram of the pachytene chromosomes.

counterpart in the long arm.

Chromosome II has the longest long arm in the complement. It is almost euchromatic excepting a small patch at the centromere. But its short arm is highly heterochromatic, extending from the centromere to more than half of its length.

Chromosome III is one of the three, bearing the median centromere. Both its arms are highly euchromatic except at the centromere. The heterochromatic regions on both the sides of the centromere are of equal length.

Chromosome IV has completely euchromatic long arm as in the chromosome II. More than one third length of the short arm consists of heterochromatin. But unlike the chromosome II, it is placed distally.

Chromosome V is almost euchromatic as chromosome III. But it, however, differs in having the submedian centromere. The most distinct and unique character of this chromosome is the presence of a small patch of het-

Table 1. Data on the pachytene chromosomes of *Solanum viarum*

Chromosome number	Total length (μ) $\pm$ S.E.	Relative length (Percent)	Short arm length (μ) $\pm$ S.E.	Long arm length (μ) $\pm$ S.E.	Centromere index
I	34.23 $\pm$ 2.28	11.66	10.35 $\pm$ 1.13	24.88 $\pm$ 1.34	30.23 sm
II	33.21 $\pm$ 1.17	11.32	7.67 $\pm$ 0.35	25.54 $\pm$ 0.81	23.09 sm
III	31.93 $\pm$ 2.42	10.88	14.90 $\pm$ 1.14	17.03 $\pm$ 1.18	46.66 ^m
IV	30.66 $\pm$ 2.61	10.45	9.37 $\pm$ 0.01	21.29 $\pm$ 1.59	30.56 sm
V	29.37 $\pm$ 2.23	10.01	10.21 $\pm$ 0.98	19.16 $\pm$ 1.25	34.76 sm
VI	28.29 $\pm$ 2.16	9.64	8.57 $\pm$ 0.73	19.72 $\pm$ 1.40	29.93 sm
VII	27.89 $\pm$ 2.18	9.50	10.56 $\pm$ 1.09	17.31 $\pm$ 1.09	37.87 ^m
VIII	19.16 $\pm$ 2.94	6.53	5.96 $\pm$ 1.32	13.20 $\pm$ 1.61	31.10 sm
IX	17.37 $\pm$ 2.37	5.92	6.31 $\pm$ 1.44	11.06 $\pm$ 0.93	36.32 sm
X	17.37 $\pm$ 2.42	5.92	6.31 $\pm$ 1.16	11.06 $\pm$ 1.26	36.32 sm
XI	14.47 $\pm$ 2.75	4.93	4.26 $\pm$ 1.28	10.21 $\pm$ 1.47	29.44 sm
XII	9.37 $\pm$ 2.84	3.19	3.83 $\pm$ 1.39	5.54 $\pm$ 1.44	40.87 ^m

erchromatin situated in the distal half of the long arm, flanked on either side by euchromatic regions.

Chromosome VI is identical to the chromosome V in all its characters, but it has, however, slightly longer heterochromatic region in the longer arm opposite to the centromere than its counter part in the chromosome V.

Chromosome VII can be easily distinguished from the others by its maximum heterochromatic region in the complement. The entire short arm and nearly half of the long arm are made up of heterochromatin. Only distal end of the long arm has faintly stained euchromatic region.

Chromosome VIII like most of the first six chromosomes has both the ends terminating in euchromatic regions. While the heterochromatic region in the middle is equally divided by the centromere.

Chromosome IX is characterised by the presence of medianly placed euchromatin and heterochromatin occupies rest of the region of each arm which amounts to nearly half of the chromosome.

Chromosome X has got the same total length, centromere index and percentage of eu- and heterochromatic regions as in chromosome IX. But unlike that chromosome, the entire short arm and more than quarter of the long arm are heterochromatic. Its long arm terminates in the euchromatic region.

Chromosome XI has both the ends terminated in heterochromatic regions as observed only in chromosome IX. Its short arm is completely heterochromatic, while the long arm has euchromatic region followed by heterochromatic as in case of chromosome IX.

Chromosome XII is the smallest chromosome having the median centromere. Like chromosomes VII, X and XI, its short arm is heterochromatic throughout its length. Its long arm is highly euchromatic except at the centromere.

Discussion

Solanum viarum possesses quite long chromosomes ranging from 9.37 microns to 34.23 microns. Each of the twelve chromosomes can clearly be distinguished from one another as the content of heterochromatin and their distribution along the chromosome differs from each other. Centromeres which are not easily discernable are, however, distinguished as small gaps in the heavily stained portion of the chromosomes. Chromosomes III, VII and XII have median centromeres while the rest of them are classified as submedian (Levan *et al.* 1964). The chromosome complement is arbitrarily divided into two groups. The six longer ones are generally euchromatic while the others have more than one third of their length consisting of heterochromatin.

Prevalence of differentiated pachytene chromosomes in nature was observed by Brown in 1949. Barton (1950) and Brown (1949) noticed highest state of differentiation in tomato which belongs to the same family as *Solanum viarum*. Li *et al.* (1963) found the presence of heterochromatic regions in *Oryza australiensis* but were unable to describe their distribution. Che (1967) also observed the rice chromosomes to be too small to describe the distribution of eu- and heterochromatic

regions. Undifferentiated chromosomes in *Sorghum* were described by Garber in 1947. Busey (1971) noticed asymmetric pairing in *Sorghum*, but *Solanum viarum* shows complete synapsis of the homologous chromosomes indicating monophylatic origin of the species.

The chromomeres of the heterochromatin are as small as microchromomeres of euchromatic region in *Solanum viarum*. But unlike the latter they are deeply stained. The chromosomes of the plant do not terminate in telochromomeres. McClintock (1929) had noted highly conspicuous heterochromatic knobs in corn. Dnyansagar and Torne (1969) have also observed large heterochromomeres at median and submedian positions in some of the chromosomes of *Rauvolfia serpentina*. *Antirrhinum* (Ernst 1938, 1939) has large and densely stained bodies in two of its chromosomes which are closer in resemblance to the knobs in corn than to the heterochromomeres in tomato. Telochromomeres having uniform but much smaller size than the large knobs of corn, have been seen in tomato, *Pennisetum* and *Prunus persica* by Barton (1950), Lobana and Gill (1973) and Jelenkovic and Harrington (1972), respectively.

The conspicuous absence of nucleolus at pachytene (except in one cell) is also a specific character of *Solanum viarum*. Chromosomes II and III are found in the vicinity of nucleolus in one of the cells. On the contrary, more than one darkly stained nucleoli-like bodies in the cytoplasm have been observed in *Prunus* by Jelenkovic and Harrington (1972) and in all the cells of *Medicago rhodopea* and two cells of *M. rhodopea* × *M. rupestris* by Gillies (1972). The early disappearance of nucleolus in *Solanum viarum* may possibly be due to extraction of RNA from the nucleolus by the cell for its rapid metabolic activities.

The basic chromosome number in *Solanum* is a controversial point. This controversy still continues as Swaminathan and Howard (1953) consider 12 as the most probable basic number, but they did not rule out the possibility of a lower basic number, since in *Petunia* species the lowest gametic number 7 has been recorded. Although not a single plant with chromosome no. $x=6$ has been so far reported, there are a number of workers who believe that the basic number is only six in *Solanum*.

However, our observations in this paper, have initiated us to postulate the theory of $n=10$, for *Solanum viarum* in particular and the genus in general. Two of the chromosome pairs namely, V and IX strongly resemble the other two chromosome pairs VI and X, suggesting the origin of one from another. Such duplication of a pair or two of chromosomes has been observed by Bask (1959) in *Corchorus sodooides* and Lewis (1962) in *Claytonia virginica*. If the duplication of chromosomes had occurred, it could be assumed that the present structure of the chromosomes might have been achieved after structural modifications had been effected through the process of meiosis during the course of evolution. Further, this assumption of duplication of chromosomes gains strength by the fact that there is a similarity of morphological characters of chromosome V with VI and IX with X.

Chromosome pairs V and VI are highly euchromatic and their centromeres are flanked by small regions of heterochromatin. The most striking character is the presence of small heterochromatic portion, situated in the distal half of the long

arm. It is followed by euchromatic regions at both ends in both chromosome pairs. Variation in the centromere index *i.e.* 34.76 and 28.09 in chromosome V and VI, respectively and difference of 1.28 microns in total length may be due to the shifting of small portion in chromosome VI from the shorter to the longer arm and the simultaneous deletion of a portion of 1.28 microns in the latter. Unlike the above pairs, chromosomes IX and X show a number of identical characters *viz.*, total length, position of centromere and total euchromatic and heterochromatic regions suggesting common origin. The only difference is in the position of eu- and heterochromatic regions. In chromosome IX, euchromatic region is followed by heterochromatic at both its ends in the shorter as well as longer arm. While in chromosome X, the entire short arm and one third of the long arm preceding centromere are completely heterochromatic. Inversion in the long arm and intrachromosomal translocation in two arms of the chromosome X can be depicted to illustrate the present structure of chromosome IX.

This structural similarity in the morphology of chromosomes at pachytene suggests that the two of the chromosomes might have arisen through the duplication of two chromosomes of the complement with the normal basic number $n=10$. Ross and Rowe in 1969 have reported $2n=60$ for *Solanum curtilobum* which seems to be the hexaploid of 10 than the pentaploid of 12 as the pentaploids are usually not stable. Magoon (1958, 1959 and 1962) and Magoon and Ramanujam (1960), on the basis of meiotic study of tuber bearing *Solanums*, are inclined to suggest the basic number of chromosomes to be less than 12 for the genus.

In view of the above discussion, it seems more reasonable to assume that the evolution of 12 pair of chromosomes in *Solanum viarum* might have taken place through a different line of evolution from the plant with the basic number ten.

Summary

Twelve pairs of chromosomes of *Solanum viarum* Dunal are described in detail. Morphological characters of chromosomes at pachytene stage as total length, relative length, centromere index and position of eu- and heterochromatic regions are used as parameters for identification. The nucleus is not persistent at pachytene. There is complete pairing between all homologous chromosomes. There is also a striking similarity between the chromosome V and VI as well as IX and X, suggesting that chromosomes VI and IX might have been evolved from chromosome V and X, respectively through duplication. The present structure of the chromosomes could have been achieved after structural modifications had been effected through the process of meiosis during the course of evolution. Therefore, it is assumed that *Solanum viarum* with $n=12$ might have originated from a plant with $n=10$.

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