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B-chromosomes, pollen germination in situ and connected grains in *Trigonella foenum-graecum*

By

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With 24 Figures

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Introduction

While investigating the cytology of *Trigonella foenum-graecum* certain cells in the root tips, PMCs as well as in pollen mitosis revealed the presence of B-chromosomes. In certain mature anthers pollen germination was noted in situ. Taking all these facts into consideration a detail study was undertaken.

Materials and methods

Seedlings of *Trigonella foenum-graecum* were sown for meiotic study as well as germinated in petri-dishes for root tip squashes. Buds were fixed in acetic-alcohol (1 : 3) and squashes made in acetocarmine, while root tip squashes were made in Feulgen. Permanent preparations were made by ethanol-butanol schedule.

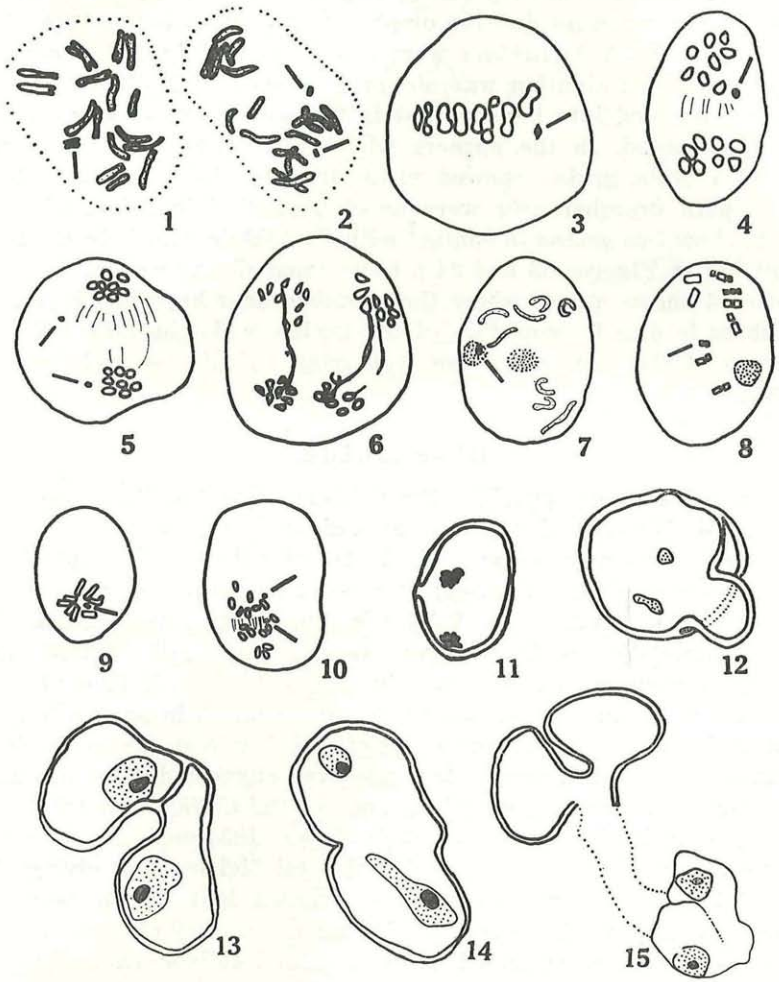
Observations

The karyotypic study revealed similar results as reported by Bhattacharyya 1958 basically, but the presence of B-chromosomes is a new addition to his report. The B-chromosomes in the somatic cells showed a median constriction clearly (Fig. 1). The maximum number of B-chromosomes observed was two (Fig. 2).

The behaviour of B-chromosomes in the pollen grains has been studied in detail (Figs. 7—11). At prophase while rest of the chromosomes were long indicating little sign of condensation the B-chromosome was quite condensed and no appreciable change in its size was noticed at later stages. In Fig. 8 the A-chromosomes are also condensed and show clear constrictions. As observed in the root tips the B-chromosomes had a median constriction here also. When the

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A-chromosomes formed the metaphase plate the B-chromosomes also oriented themselves on the equator like the rest of the complement (Fig 9). At anaphase it shows the normal disjunction and two daughter B-chromosomes reached the opposite poles (Fig. 10) which were included in the telophasic nuclei (Fig. 11). No B-chromosome



Figs. 1—2: Roottip cells showing one and two B-chromosomes respectively. — Fig. 3: A PMC at M. I showing a B-chromosome on the plate. — Figs. 4—5: A.I showing a undivided and a divided B-chromosome respectively. — Fig. 6: A.II showing bridges. — Figs. 7—11: Stages of pollen mitosis showing the behaviour of B-chromosomes. — Fig. 12: A pollen grain with an abnormal protuberance. — Fig. 13: Two connected grains with the middle wall separating the passage in the tubes. — Fig. 14: Two grains showing direct connection. — Fig. 15: Connected grains with their contents outside.

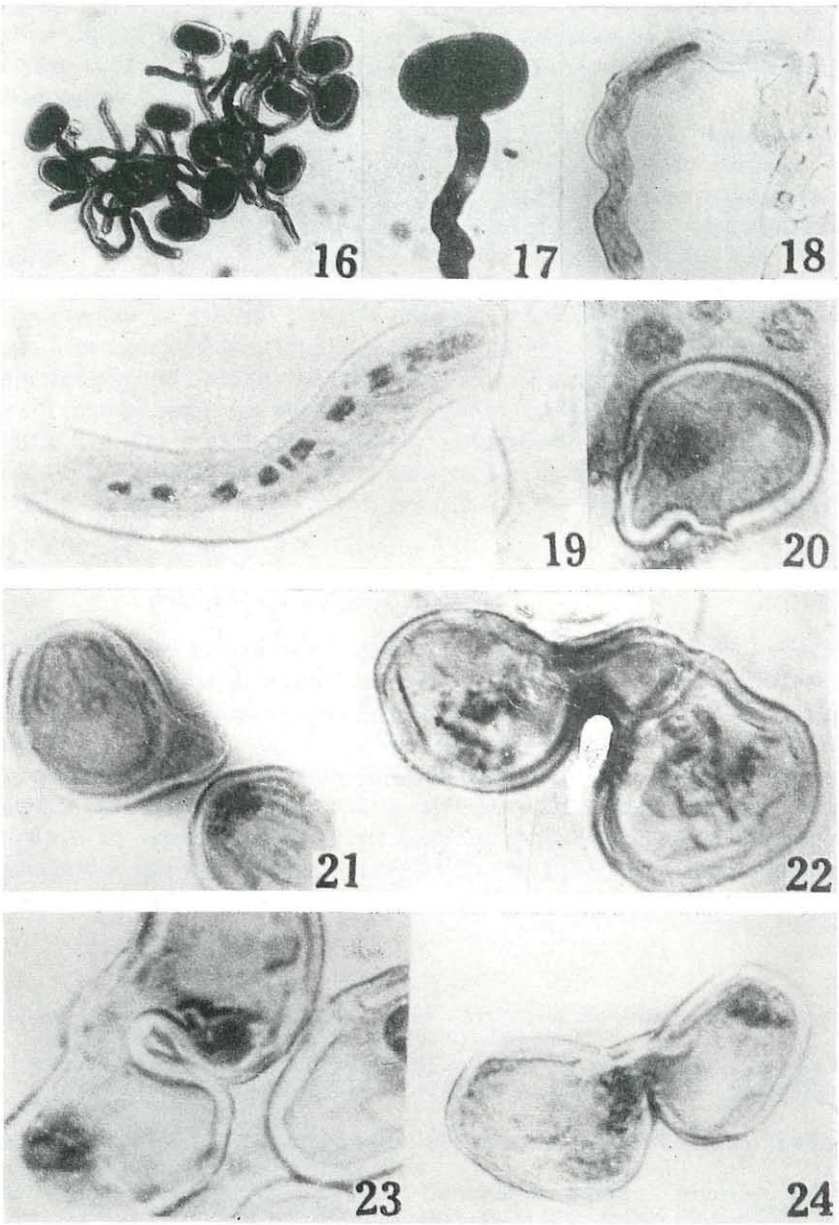
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was observed lagging. It was noted that the second division in the grains took place either as early as at the commencement of the tubes (Fig. 19) or as late as at the tip after a considerable growth. Progeny of this plant consisted of members having a B-chromosome in the PMCs while others had none. In their PMCs a B-chromosome was at M. I which took deeper stain than the rest oft the chromosomes (Fig. 3). It underwent division or passed as such to the poles at A. I (Figs. 4, 5). At A. II Bridges were also present (Fig. 6). In anthers where pollen germination was observed (Figs. 12, 16, 17) the tubes were all jumbled into bunches but in no instance was the fusion of the tubes noted. In the anthers where the pollen tubes were not present certain grains showed protuberances (Figs. 20, 21). Some grains with protuberances were noted connected in pairs. Figs. 13 and 22 show two grains in contact with the middle wall between them intact but in Figures 23 and 24 a clear canal like passage is evident. Figure 14 shows a case where the protuberances have not developed but there is direct connection of the grains, while in Figure 15 the contents of the two grains are seen lying outside the walls of the grains.

Discussion

Animal kingdom provides the evidence that the B-chromosomes are the derivatives of the sex chromosomes. Cases where certain chromosomes lie more or less on the boarder line of the typical sex chromosomes and the accessory chromosomes have been reported by MELANDER 1950 and VIRKII 1954. Disturbance of sex determination in *Sphagnum* due to the presence of many accessory chromosomes leading to monoecism is reported by SORSA 1956. It is evident from this that as in animals the accessory chromosomes in mosses may be derived from sex chromosomes. Origin of B-chromosomes by fragmentation of A-chromosomes has also been suggested. CLELAND 1951 and REESE 1954 obtained data in *Oenothera* and *Caltha* which supports this hypothesis. In *Caltha palustris* REESE 1954 actually observed certain small A-chromosomes undergoing misdivision and giving rise to two telocentric small chromosomes which look and behave like B-chromosomes in this species. Unlike such accessory chromosomes in *Xanthisma texanum* described by BERGER and collaborators (BERGER & WITKUS 1954, WITKUS & al. 1955) the B-chromosomes in *Trigonella foenum-graecum* has a median constriction and hence the postulation of origin by fragmentation is not valid in the present material. The connected grains may be the result of misdivision at cytokinesis following T. II. In places where the partition wall between two such grains is present it may indicate partial laying down of the separation wall ultimately permitting the deposition of ornamentation on it along

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Figs. 16—17: Germinated pollen grains in situ. — Figs. 18—19: The generative nucleus approaching the tip of the tube and the same at metaphase near the point of origin of tube. — Figs. 20—21: Grains showing protuberances. — Fig. 22: Two connected grains with middle partition wall. — Figs. 23—24: Connected grains with clear passage.

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with the rest of the grains. The nuclear position indicating migratory action might only represent the abnormal position of the nuclei following T. II. However, occurrence of such grains makes an interesting observation in the present study.

In *Coriandrum sativum* the authors have reported a unique case of nuclear migration from one grain into another (JOSHI & RAGHUVANSHI 1965). The migration of the nuclear material takes place in the form of a protuberance through the pores, hence no canal like passage was needed. In this instance one of the grains type α, the donor is the direct transformation of a PMC into a pollen without the intervention of meiosis while the recipient grain has developed from one of the microspores comming out of a sporad. Hence it is a positive case leaving no grounds to doubt that the nuclear migration from one grain into another can take place in grains developing from different PMCs i. e. not a case of incomplete cytokinesis. Keeping in view the case of *Coriandrum sativum* the authors are of the view that although it may be case of imcomplete cytokinesis the possibility of fusion of grains also remains open.

Summary

The paper presents a report of B-chromosomes in the PMCs, pollen grains as well as root tip cells of *Trigonella foenum- graecum*. Certain anthers revealed germinated pollen grains with long tubes jumbled into a mass i n s i t u, in some cases the second division in the tubes was noted. Certain joined grains showed connection either directly or through a canal like structure. Evidence has been put forward in support of the view that this may be due to incomplete cytokinesis after T. II but the possibility of the fusion of the grains has also been discussed.

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