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A TRISOMIC PAPAVER RHOEAS

by

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In a culture of *Papaver rhoeas* plants obtained from the botanical garden of Copenhague, I found one plant striking by the abnormal appearance of its flowers. The calyx of these flowers did not open, consequently the flowerbuds began to rot away and perish. By opening artificially the calyx, the corollas opened to some extent and it was possible to pollinate the pistils (fig. 1).

A part of the flowers was opened artificially and was pollinated with pollen of normal *P. rhoeas*, originating from the Botanic Garden at Lyon. Another part of the artificially opened flowers was selfed and still another part was only opened and left for free pollination.

The pollinations with *P. rhoeas* (from Lyon) all failed, but the selfed flowers and the open-pollinated flowers produced seed.

KARYOLOGICAL INVESTIGATION

1. *Meiosis*

Meiosis was studied in anthers. After a preliminary examination with aceto-orcein, the anthers with dividing P. M. C.'s were fixed in acetic-alcohol (1 : 4), after which Feulgen squashes were made. In diakinesis and first metaphase 7 bivalents and 1 univalent were found (pl. I, figs 2, 3, 4), so this plant proved to be a trisomic with $2n + 1 = 15$ chromosomes, which was also affirmed by a somatic metaphase plate of a tapetum cell (pl. II, fig. 9).

Fig. 1. *Papaver rhoeas*, 2n = 15, with artificially opened flowers.

This extra chromosome was always lagging in anaphase, but after that it behaved in different ways. At times it did not divide and moved to one pole of the spindle, in other cases the univalent divided, so that both poles of the spindle got a split half of this extra chromosome.

In the first case the first anaphase resulted in two telophase plates with 7 + 8 chromosomes (pl. I, fig. 5; pl. II, fig. 6); in the second case 8 + 8 chromosomes could be counted (pl. II, fig. 7). A third case occurred, viz. that the dividing extra chromosome was disjoining when the other chromosomes were already in the telophase stage (pl. II, fig. 8). Very probably this case results in the formation of micronuclei.

2. *Mitosis.*

Seeds from an open pollinated flower and seeds from a selfed flower were placed in Petri-dishes upon moist filter paper. After germination, the roots of about $\frac{1}{2}$–1 cm were pretreated with α—bromo-naphthalene during $2\frac{3}{4}$ hours, fixed with acetic-alcohol (1 : 3) and kept overnight in abs. alc. After hydrolysis (15 min.) the roottips were stained with leuco-basic-fuchsin (Feulgen reaction) during about three hours and squashed.

The progeny of a selfed flower (44 seedlings) consisted for the majority (30) of individuals with 14 and others (11) with 15 chromosomes. Moreover one individual had 14 chromosomes with a fragment and two individuals had a diploid number of 21 (see table I). The progeny of an open pollinated flower (33 seedlings) consisted almost only (29) of individuals with 14 chromosomes, with a few exceptions: three with 15 and one with 21 chromosomes were found (see table II). (See pl. III, figs 10, 11, 12, 13 and text fig. 12a).

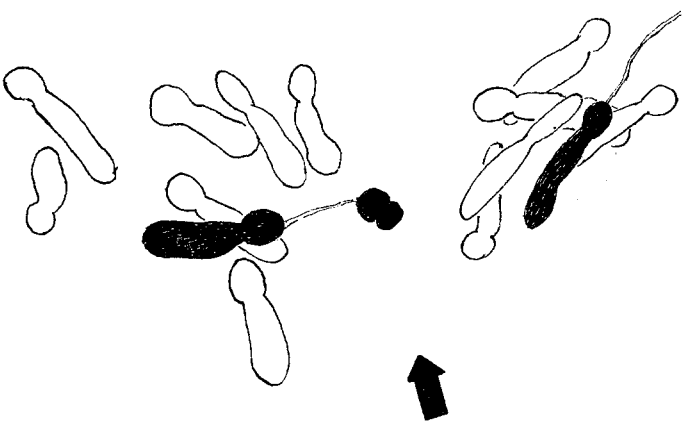


Fig. 12a. Drawing to explain fig. 12, plate III.

Morphologically the chromosomes differ somewhat in their length. The centromere is submedian or subterminal, whereas two satellite chromosomes occur. This satellite is very typical, it has the same appearance as has been described for *Papaver argemone* (KOOPMANS, 1954). In most cases the satellite is threadlike, the length depending on the contracting of the chromosome. In contracted chromosomes the satellite can be rather short (fig. 13), in others it is longer (fig. 11) and

Fig. 2. Meiosis. Diakinesis. Seven bivalents and one univalent at 1 o'clock. Enlargement 1300 ×.

Fig. 3. Meiosis. First metaphase. Seven bivalents and at the top one univalent. Enlargement 1600 ×.

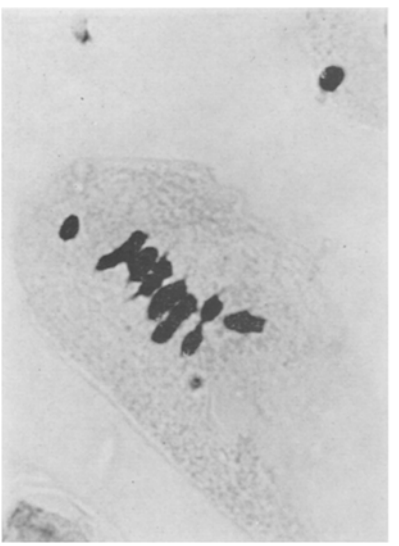


Fig. 4. Meiosis. First metaphase, side view. Seven bivalents and one univalent at the left. Enlargement 1300 ×.

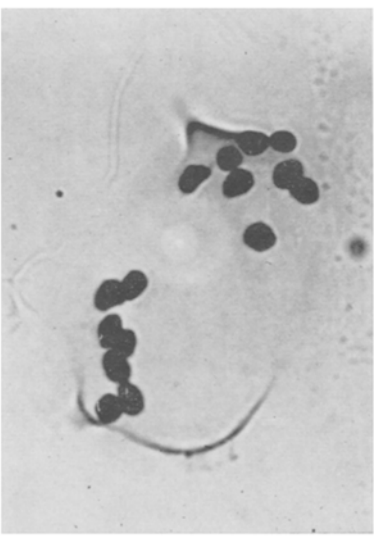


Fig. 5. Meiosis. First anaphase, consisting of seven + eight chromosomes. Enlargement 1800 ×.

Plate II

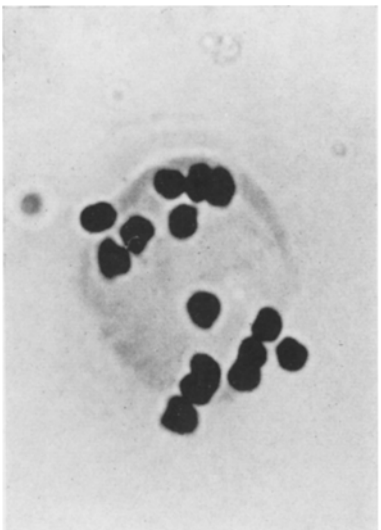


Fig. 6. Meiosis. First anaphase. The univalent is lagging and goes undivided to one pole; result 7 + 8 chromosomes. Enlargement 2300 ×.

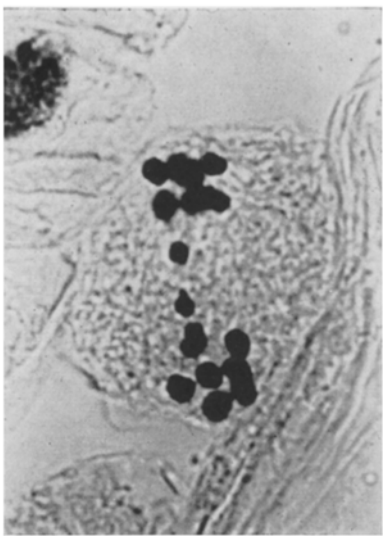


Fig. 7. Meiosis. First anaphase. The univalent has divided and so 8 + 8 chromosomes are formed. Enlargement 1750 ×.

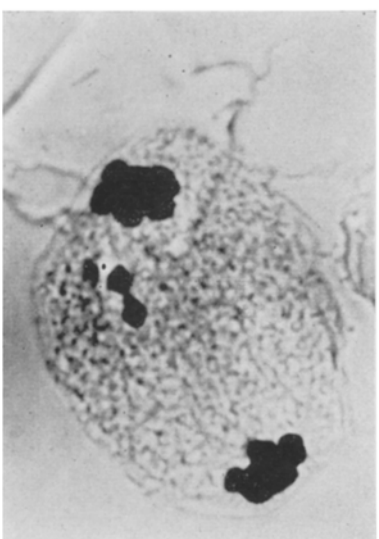
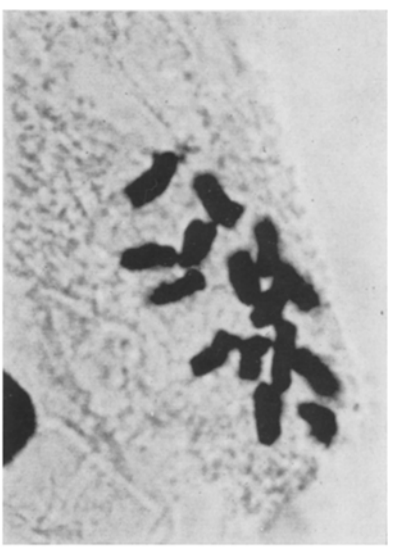


Fig. 8. Meiosis. First telophase. Between the two poles, the dividiug univalent. Enlargement 1800 ×.



Fg. 9. Mitosis. Somatic metaphase in a tapetum cell: 15 chromosomes. Enlargement 1900 ×.

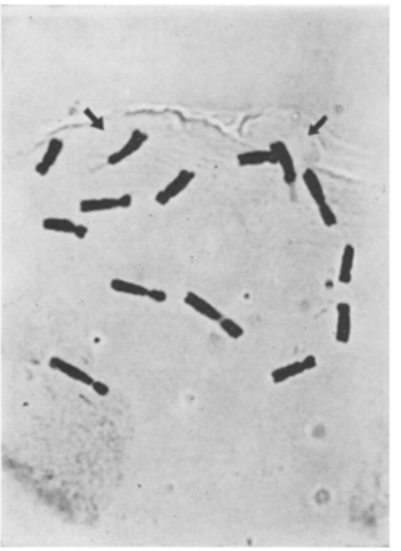


Fig. 10. Mitosis. Metaphase in a roottip with 14 chromosomes. Two chromosomes with a threadlike satellite. Enlargement 1250 ×.

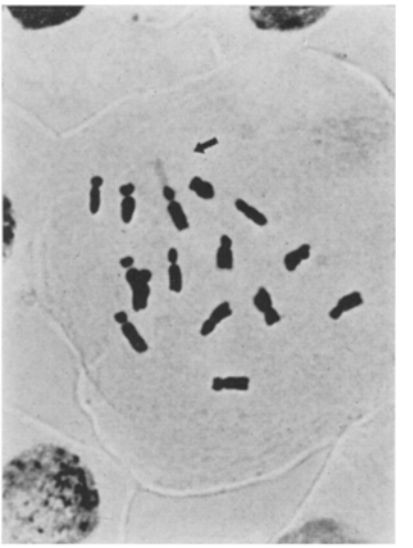


Fig. 11. Mitosis. Metaphase in a roottip with 15 chromosomes. Only one satellite chromosome can be seen. Enlargement 1350 ×.

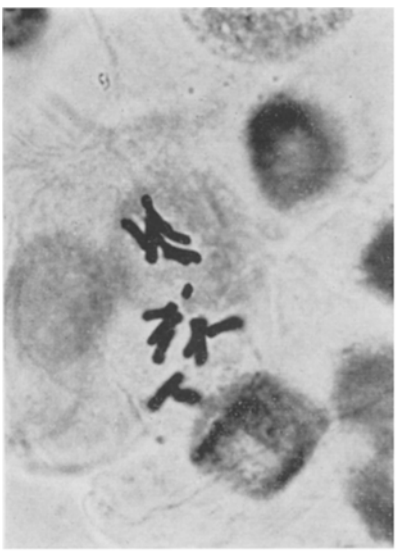


Fig. 12. Mitosis. Metaphase in a roottip with 14 chromosomes + 'fragment' in the centre. Enlargement 1250 ×.

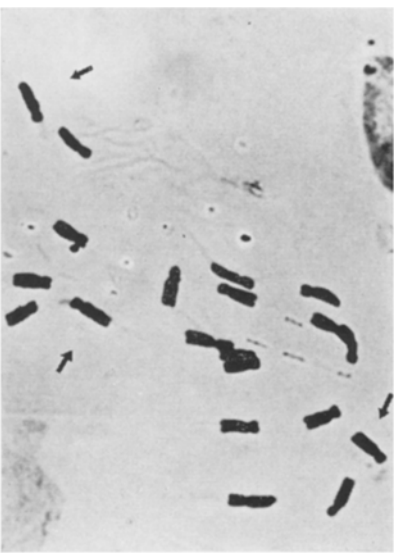


Fig. 13. Mitosis. Metaphase in a roottip with 21 chromosomes. Three satellites. The twenty-first chromosome lies outside the plate. Enlargement 1300 ×.

Plate IV

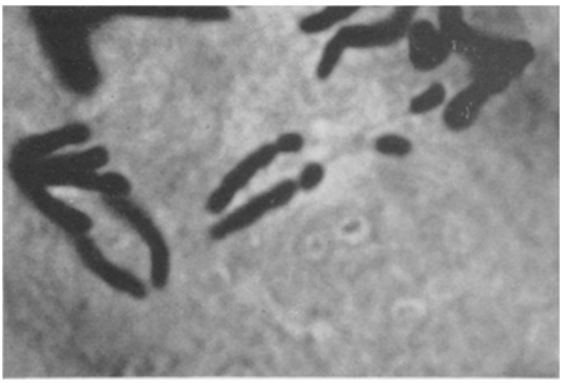


Fig. 14. Mitosis. The divided satellite chromosome with in both halves at the end of the satellite thread a chromosome 'fragment'. Enlargement 2850 ×.

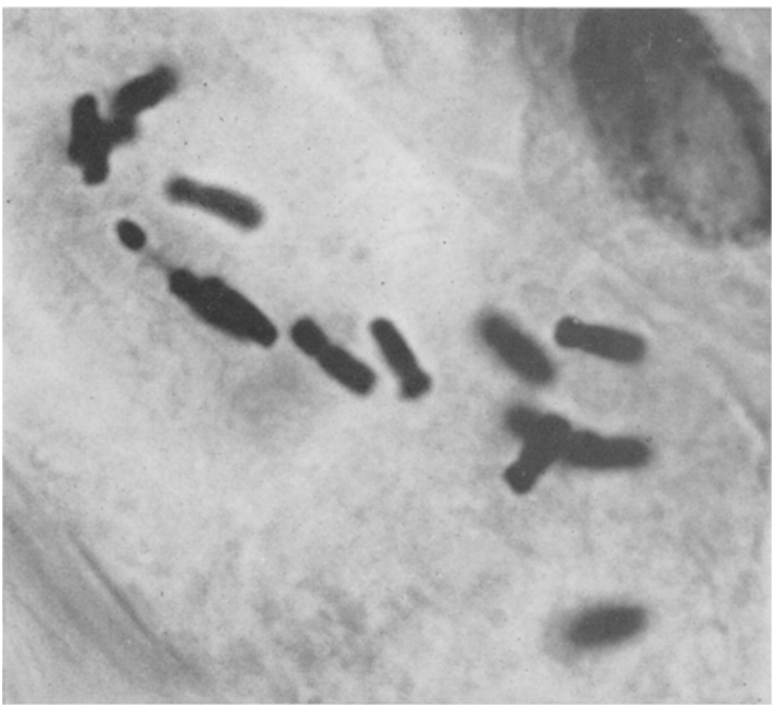


Fig. 15. The two satellite chromosomes, one with a 'fragment' at the end of the satellite thread, the other one with only a satellite thread. The satellite chromosomes are partly overlapping. Enlargement 2900 ×.

sometimes satellite chromosomes occur where the satellite is twice as long as the chromosome itself.

As to the chromosome 'fragment' at first I thought indeed that I had to do with a real fragment, perhaps arisen by misdivision of the extra chromosome. By closer examination however, it turned out that this 'fragment' has a constriction and moreover that it is attached to the satellite thread of one of the satellite chromosomes. The other satellite chromosome has the normal appearance of a threadlike satellite. Fig. 14 (plate IV) shows a metaphase plate where thanks to the pretreatment with α-bromo-naphtalene all chromosomes have divided and so we see here the two halves of the satellite chromosome + fragment. There is no doubt that at the end of the satellite thread there is an euchromatic chromosome part. In fig. 15/15a (plate IV) the two

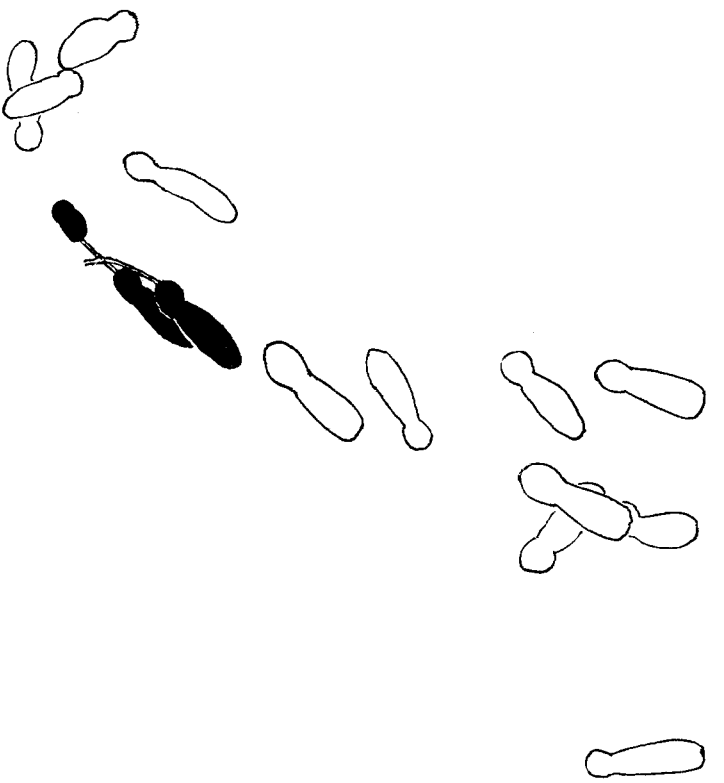


Fig. 15a. Drawing to explain fig. 15, plate IV.

satellite chromosomes are lying close together: one with a threadlike satellite, the other one has this chromosome fragment at the end of the thread. It seems rather enigmatical how this has been brought about. May we suppose that by some unknown cause the end has changed into an euchromatic part? As I have found this phenomenon only once in 77 inviduals, I am unable to propose an acceptable explanation.

Discussion

As already mentioned this abnormal plant of *Papaver rhoeas* formed pollen with 7 and with 8 chromosomes. Supposing that the meiosis of the E. M. C. has been accomplished in the same manner and that the growth rate of the pollen tubes with 7 and with 8 chromosomes is the same, theoretically the progeny must consist of individuals with 14, 15 and 16 chromosomes (resp. 25, 50 and 25%). Since individuals with 16 chromosomes have not been found and individuals with 15 chromosomes are found much less than individuals with 14 chromosomes (resp. 25% and 9.09%, see tables), it is most likely that gametes with 7 chromosomes and gametes with 8 chromosomes are not formed in the same number. Supposing there are formed x gametes with 7 and y gametes with 8 chromosomes, then the number of individuals with 14 chromosomes must be x^2 and the number of individuals with 15 chromosomes must be 2xy.

From $x^2 : 2xy = \pm 3 : 1$ (see table I, 30 : 11) it follows that $x = 6y$, in other words the ratio of gametes with 7 and 8 chromosomes is 6 : 1.

Theoretically we might expect individuals with 14, 15 and 16 chromosomes in the ratio of 36 : 12 : 1. On the total number of 41 individuals (30 with 14 and 11 with 15 chromosomes, if we omit the individuals with 14 + 'fragment' and those with 21 chromosomes), we should find individuals with 14, 15 and 16 chromosomes in the ratio: 30·12 : 10·04 : 0·84.

Actually I found 30 individuals with 14 and 11 with 15 chromosomes. Individuals with 16 chromosomes did not occur, but regarding the small number of investigated individuals this is not surprising. Comparing the two progenies it strikes that the percentage of individuals with 15 chromosomes is rather different. As the trisomic plant stood

among normal *P. rhoeas* plants, it is most likely that the open pollinated flowers were pollinated with normal *P. rhoeas* pollen, that is to say pollen with 7 chromosomes.

On the above mentioned assumption that gametes with 7 and with 8 chromosomes are formed in the ratio 6 : 1 we might expect on a total number of 32 individuals from open pollinations (see table), individuals with 14 and with 15 chromosomes in the ratio of 27·'43 : 4·'57.

Actually I found 29 individuals with 14 and 3 with 15 chromosomes. Admitting this is no sufficient proof for the supposition that gametes with 7 and with 8 chromosomes occur in the ratio 6 : 1, this might be considered an indication that both in pollen and in embryosacs this ratio is the normal one.

As for the individuals with 21 and the one with 14 + 'fragment', it is at the moment difficult to give an explanation. As seed now has been sown in the experiment garden it is perhaps possible to recognize these individuals morphologically. Perhaps the cultures of this year will solve also the question if there is any connection between the occurrence of the extra chromosome and the abnormality of a non-opening calyx.

LITERATURE

KOOPMANS, A., 1954. A. karyological study of *Papaver argemone* (Excerpta medica, 8 : 418).

TABLE I. *Offspring obtained by selfing*

number of plants	number of chromosomes 14	14+f	15	21	number of metaphase plates counted
44	30	1	11	2	for 14 chromosomes: 4+1+7+7+1+12+15+7+5+24+1+19+5+9+3+8+18+12+7+4+8+4+20+5+6+4+2+2+2+13 = 235 plates for 14 chromosomes + 'fragment': 4 = 4 plates for 15 chromosomes: 3+1+11+22+5+17+2+4+7+12+2 = 86 plates for 21 chromosomes: 13+5 = 18 plates Total number of plates counted: 343

TABLE II. *Offspring obtained by open pollination*

number of plants	number of chromosomes 14	14+f	15	21	number of metaphase plates counted
33	29	–	3	1	for 14 chromosomes: 8+9+34+6+3+8+12+11+8+1+4+5 +7+9+13+10+4+9+2+1+4+1+2+ 7+2+19+5+8+6 = 218 plates for 15 chromosomes: 7+4+11 = 22 plates for 21 chromosomes: 4 plates Total number of plates counted: 244