

## COLCHICINE INDUCED CHROMOSOMAL INTERCHANGE AND COLCHITETRAPLOIDY IN *PHLOX DRUMMONDII*

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### ABSTRACT

Young seedlings of *Phlox drummondii* were treated with colchicine. Plants showing characteristic features of interchange (translocation), and colchitetraploidy during meiosis were isolated. The interchange heterozygote exhibited a lot of variation in petal character and therefore these two characters appear to be correlated. The colchiploid exhibited usual characteristic features of autopolyploidy.

**Key words:** Interchange heterozygote, colchicine, colchitetraploidy, *Phlox drummondii*.

*Phlox drummondii* (Polemoniaceae) is a common garden ornamental, cultivated for its beautiful flowers of different shapes and shades. This plant has a low chromosome number ( $2x = 14$ ) and the size of the chromosomes is fairly big. It has therefore been used in many cytogenetical investigations. Though there are a few reports of natural [1] and induced [2, 3] chromosomal interchanges (reciprocal translocation) in *P. drummondii*, none have been associated with any morphological trait. Also, generally the induction of interchange has been achieved by physical and chemical mutagens, and not by colchicine which is a known polyploidizing agent.

During the course of studies on the effect of colchicine on young seedlings of *P. drummondii*, a plant was isolated which exhibited characteristics of chromosomal interchange during meiosis and was also found to have a lot of variation in petal character. Another plant showing characteristics of colchitetraploidy, both at cytological and morphological level, was also isolated. The present report describes both these variants.

### MATERIALS AND METHODS

Young seedlings of *Phlox drummondii* were treated with colchicine. Cotton swabs soaked in 0.2% aqueous colchicine (Sigma) solution were placed in between the two cotyledonary

leaves for 2 or 3 days, 5 h per day. The tip of seedlings was thoroughly washed after the treatment.

For meiotic studies, young flower buds were fixed in acetic acid : absolute alcohol (1:3) mixture for at least 24 h. Anthers were squashed in 1% iron-acetocarmine. Microphotographs were taken from temporary slides. Pollen stainability was determined using iron-acetocarmine.

## RESULTS

*Floral morphology.* In control, the flower number was 5-10 in an inflorescence (Fig. 1: 1) and there were 5 petals in each flower.

The interchange heterozygote produced 8-14 flowers in an inflorescence (Fig. 1: 1). There was a lot of variation in number, shape and size of flowers which were grouped into different types (Fig. 1: 2). All flowers were comparatively smaller than the control. Only 14.3% flowers had 5 small petals (Fig. 1: 2a). Maximum number (42.8%) of flowers had 4 petals (Fig. 1: 2b). This was followed by 20% flowers with 3 big and 1 small petal (Fig. 1: 2c), 2.8% with 2 small and 2 small dissected petals (Fig. 1: 2d), 8.5% with 2 big and 2 dissected petals (Fig. 1: 2e), 4.2% with 4 small petals arranged in only one half (Fig. 1: 2f), 5.7% with 3 small petals (Fig. 1: 2g) and 1.4% with 3 petals, one of them being keel shaped (Fig. 1: 2h). The changes were very prominent so that the inflorescence could easily be distinguished from the control.

In the colchitetraploid, the flowers were slightly bigger than the control and in some cases had 7 petals. The colour of the petals was darker, and they remained in bloom for 2-3 days more than the control.

*Stomata.* The size and number of stomata showed variation in the interchange heterozygote, polyploid plant and the control. There was an increase in the size of stomata in the colchiploid, however, the number of stomata per unit area was reduced (58) as compared to the interchange heterozygote (79) and the control (91).

*Pollen stainability and fertility.* The data on pollen stainability, number of capsules per plant, number of seeds per capsule and total number of seeds per plant are summarized in Table 1. The interchange heterozygote had very reduced fertility as compared to control and colchitetraploid.

## CYTOLOGY

The data on chiasma frequency, association of chromosomes at diakinesis/metaphase I and distribution of chromosomes at anaphase I are presented in Table 2.

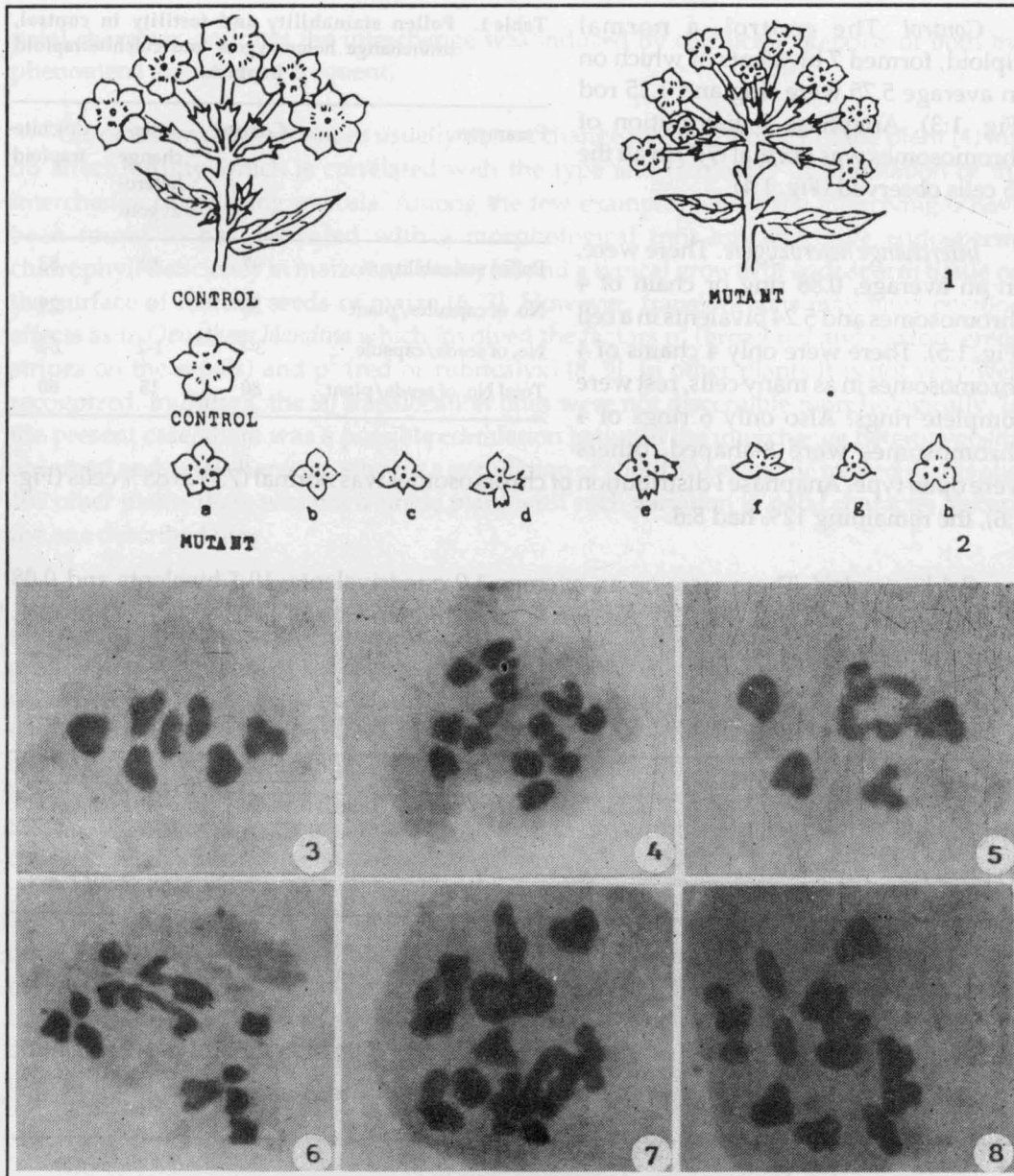


Fig. 1. Plant morphology and cytological patterns in *Phlox drummondii*. 1) A twig with inflorescence in control and translocation heterozygote mutant. 2) Variation in number, size and shape of petals in the mutant. 3,4) Control: 3) metaphase I of meiosis with 7 II; 4) anaphase I with 7:7 distribution of chromosomes. 5,6) Translocation heterozygote: 5) metaphase I showing 1 ring of 4 + 5 II; 6) anaphase I with 7:7 distribution of chromosomes. 7,8) Colchitetraploid: 7) MI with 2 IV + 10 II; 8) MI of meiosis showing 14 II.

*Control.* The control, a normal diploid, formed 7 bivalents of which on an average 5.75 were ring and 1.25 rod (Fig. 1:3). Anaphase I distribution of chromosomes was normal (7:7) in all the 25 cells observed (Fig. 1:4).

*Interchange heterozygote.* There were, on an average, 0.88 ring or chain of 4 chromosomes and 5.24 bivalents in a cell (Fig. 1:5). There were only 4 chains of 4 chromosomes in as many cells, rest were complete rings. Also only 6 rings of 4 chromosomes were 8-shaped, others were open type. Anaphase I distribution of chromosomes was normal (7:7) in 88% cells (Fig. 1:6), the remaining 12% had 8:6.

*Colchitetraploid.* There were on an average 1.9 quadrivalents, 10.1 bivalents and 0.08 univalents per cell (Figs. 1:7, 1:8). Anaphase I distribution was 14:14 in 80%, 15:13 in 12%, and 16:12 in 8% cells (Table 2).

**Table 1. Pollen stainability and fertility in control, interchange heterozygote and colchitetraploid in phlox**

| Parameter                | Control | Interchange heterozygote | Colchitetraploid |
|--------------------------|---------|--------------------------|------------------|
| Pollen stainability, %   | 97      | 80                       | 83               |
| No. of capsules/plant    | 38      | 9                        | 29               |
| No. of seeds/capsule     | 3-4     | 1-2                      | 2-3              |
| Total No. of seeds/plant | 80      | 15                       | 60               |

**Table 2. Chiasma frequency, types of associations and anaphase distribution in phlox**

| Genotype                 | No. of cells analysed | Chiasma frequency | Mean No. of   |         |        |          |            | Anaphase distribution |             |
|--------------------------|-----------------------|-------------------|---------------|---------|--------|----------|------------|-----------------------|-------------|
|                          |                       |                   | quadrivalents | ring II | rod II | Total II | univalents | chromosome separation | frequency % |
| Control                  | 25                    | 17.4              | —             | 5.75    | 1.25   | 7.0      | —          | 7:7                   | 100         |
| Interchange heterozygote | 25                    | 14.0              | 0.88          | 4.08    | 1.16   | 5.24     | —          | 7:7                   | 88          |
|                          |                       |                   |               |         |        |          |            | 8:6                   | 12          |
| Tetraploid               | 25                    | 25.8              | 1.92          | 6.24    | 3.88   | 10.12    | 0.08       | 14:14                 | 80          |
|                          |                       |                   |               |         |        |          |            | 15:13                 | 12          |
|                          |                       |                   |               |         |        |          |            | 16:12                 | 8           |

## DISCUSSION

The present cytogenetic investigation on *P. drummondii* mainly deals with two aspects. One, that the chromosomal interchange was associated with a morphological trait namely

petal character, and that the interchange was induced by colchicine. Reports of both the phenomena are not very frequent.

Chromosomal interchanges usually do not change the morphology of the plant [4] but do affect fertility which is correlated with the type and frequency of orientation of the interchange ring during meiosis. Among the few examples where the interchanges have been found to be associated with a morphological trait are: defective endosperm, chlorophyll deficiency in maize and barley [5] and a typical growth of endosperm tissue on the surface of mature seeds of maize [6, 7]. However, translocations may have position effects as in *Oenothera blandina* which involved the factors  $p^s$  (broad red and narrow green stripes on the sepals) and  $p^r$  (red or rubricalyx) [8, 9]. In other plants it is not very well recognized. In *Datura*, the 90 translocation lines were not discernible morphologically. In the present case, there was a possible correlation between the interchange heterozygosity observed and petal character, since in a population of about 50 colchicine treated plants and 100 other plants there was not a single plant with such variation in petal character, except the one described here.

The use of colchicine in bringing about chromosome doubling is well known. However, sometimes mutations have been induced in *Sorghum* [10, 11] and flax [12]. In *Sorghum*, despite many morphological changes, no change in the chromosome structure or number was observed [13]. Colchicine induced partial desynapsis associated with floral character was reported in *Lathyrus* but that was explained to be due to gene mutation and not due to chromosome structural changes [14]. However, in the present case colchicine seems to have led to chromosome breakage otherwise interchange could not have occurred. The possibility that the interchange was a natural one is also ruled out as the seeds used in this study were collected from a plant which was cytologically normal.

The cytological behaviour of the interchange heterozygote was typical as observed in other plants. There was a preponderance of ring of 4 chromosomes and most of them were open type which would result in adjacent I/II orientation. This type of orientation had its effect on fertility which was significantly reduced (Table 1) and was similar to the results obtained earlier [3]. The cytology of the colchitetraploid was also typical as reported earlier in *P. drummondii* [15].

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