# REDUCED QUADRIVALENT FREQUENCY IN C<sub>1</sub> COLCHIPLOIDS OF *TITHONIA*

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### **ABSTRACT**

Tithonia speciosa (Mexican sunflower) is a diploid with 2n=34. Morphological comparison of the diploid and  $C_1$  generation tetraploid showed that the latter had bigger, long lasting capitula with increased number and size of ray florets. Autotetraploids (2n=4x=68) in  $C_0$  and  $C_1$  generations showed mean chromosome association/pmc as 8.5 IV + 1.3 III + 1.3 II + 1.5 I and 3.6 IV + 1.7 III + 23.4 II + 1.7 I, respectively at MI. Anaphase 1 appeared normal with 34:34 distribution. The  $C_0$  plants showed 60.8% pollen fertility and 30.4% seed set. Reduced quadrivalent frequency, increased bivalent formation and improved pollen (85.0%) and seed (67.0%) fertility as observed in  $C_1$  plants may be due to lower chiasma frequency in the diploid and some preferential pairing in this outcrossing cultivar.

Key words: Tithonia, quadrivalent, colchiploids, chiasma, preferential pairing.

Tithonia (Asteraceae) is a small genus of robust semihardy annuals, distributed in Central America and the West Indies [1, 2]. Two species, namely *T. diversifolia* A. Gray and *T. speciosa* Hook (*T. rotundifolia* Blake), are cultivated in gardens all over India. *T. speciosa* commonly known as Mexican sunflower is a tall plant with large, coarse, cordate leaves and deep orange-red capitula with tufted yellow centre. It is ideal for screen planting and cutflower [3]. Investigations on the genus are confined to chromosome counts which show inter and intraspecific numerical variations in chromosomes, e.g. 2n=22 in *T. diversifolia* A. Gray [4, 5] and 2n=34 in *T. rotundifolia* (Mill) Blake [6]. Colchitetraploids of *Tithonia speciosa* were raised for the first time with the objective of studying the meiotic behaviour and enhancing its ornamental characteristics.

## MATERIALS AND METHODS

Seeds of *Tithonia speciosa* were obtained from the garden section of the National Botanical Research Institute. Young seedlings were treated with different concentrations (0.2%, 0.3%, 0.5%) of aqueous colchicine solution thrice a day for three consecutive days. The C<sub>0</sub> plants were selfed to produce C<sub>1</sub> plants. For meiotic studies young capitula of

suitable size were fixed in Carnoy's solution (1 acetic acid: 3 chloroform: 6 alcohol) for 48 h in which acetic acid was saturated with ferric acetate. Anthers were squashed in 1% acetocarmine. The pollen fertility estimates are based on stainability in 1:1 acetocarmine-glycerol. Photo-micrographs were taken from temporary slides.

# **RESULTS**

Positive results were obtained with 0.3% colchicine. Concentrations of 0.2% and 0.5% were ineffective and lethal, respectively. The autotetraploid seedlings showed "colchicinegigantism" vis-a-vis the diploids. The  $C_1$  tetraploid seedlings initially showed slower growth compared to diploids. The mature  $C_1$  plants were morphologically different from the diploids. Flower buds in the  $C_1$  autotetraploids were bigger (6.9 cm) with longer duration of flowering (75 days) as compared to diploids (35 days). The pollen size increased significantly in tetraploid  $C_1$  (27%) in comparison with the control. The  $C_1$  had 18 ray florets/capitulum as compared to 11 in the diploids. The ray florets were significantly bigger (2.9 x 0.9 cm) in the  $C_1$ . Seed set was poor in  $C_0$  (30.4%) but was reasonably good in  $C_1$  plants (67%). The tetraploid plants were shorter (112.1 cm tall) profusely branched with smaller leaves (9.8 x 5.2 cm) than the diploids (137.5 cm tall; 14.8 x 7.0 cm leaf size).

The diploids formed 17 bivalents at diakinesis and metaphase I (Fig. 1:1,2). Five to eight of these were ring types. Mean chiasma frequency was  $27.2 \pm 1.9$  per cell. Subsequent course of meiosis was regular (Fig. 1:3) with normal pollen and seed fertility. In contrast, meiosis in  $C_0$  and  $C_1$  colchiploids was characterized by quadrivalents at diakinesis and metaphase I (Fig. 1:4–8). The number of quadrivalents ranged from 5 to 14 in  $C_0$  and 1 to 6 in  $C_1$ . None of the PMCs showed 34 bivalents at metaphase I. The percentage of chromosomes associated as quadrivalents, trivalents, bivalents and univalents in the  $C_0$  and  $C_1$  was 50.3, 5.4, 42.1, 2.2 and 21.0, 7.5, 69.0 and 2.5, respectively. On an average, 8.5 IV + 1.3 III + 14.3 II + 1.5 I were observed in  $C_0$  and 3.6 IV + 1.7 III + 23.4 II + 1.7 I in  $C_1$ . A maximum of 14 IV in  $C_0$  (Fig. 6) and 6 IV in  $C_1$  (Fig. 8) was found. Chiasma frequency per chromosome in colchiploids ( $C_0$  and  $C_1$ ) never equalled that of the diploid progenitor.

Despite the highly complex nature of chromosomal associations at metaphase I, the meiosis was generally regular. With the exception of 14.8% in  $C_1$  with unequal AI distribution (36:32) almost all had normal 34:34 chromosomal distribution. Pollen fertility was higher in  $C_1$  (84.9%) which resulted in good seed set.

## DISCUSSION

Observations indicate "colchicine-gigantism" in determinate parts of the autotetraploids. Similar observations were reported in *Brassica* [7], forage crops [8] *Amaranthus* [9], *Crossandra* [10] and *Petunia* [11].

The range of quadrivalents per cell (5-14 in C<sub>0</sub> and 1-6 in C<sub>1</sub>) showed a significant reduction in quadrivalent frequency. The low multivalent frequency was probably due to the lower chiasma frequency in the diploid and autotetraploids [12]. Preferential pairing in autotetraploids may also reduce the quadrivalent frequency [9, 13, 14]. Further the significant reduction in quadrivalent frequency from C<sub>0</sub> to C<sub>1</sub> is an important feature and subsequent studies will reveal if the trend is maintained in further generations.

On the basis of pollen fertility in Co of T. speciosa, one should expect good seed set. However, it was low as compared to the diploid. The factors responsible for such fertility reduction in induced polyploids have not been clearly understood. So far two explanations, cytological and genetic, have been offered [12]. The differences in fertility of autotetraploids are described to variations in chromosome behaviour at meiosis [15] and where no cytological irregularities discernible [16, 17].

The improved seed set in C<sub>1</sub> is attributed primarily to a significant reduction in quadrivalents and increased bivalents, resulting in good pollen fertility, as observed in autotetraploid pearl millet [18, 19]. This may not be true for all the synthetic polyploids because in

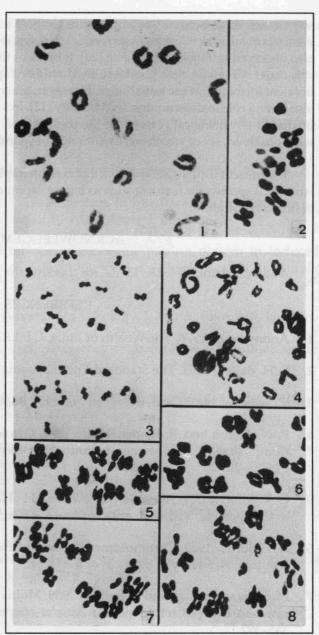


Fig. 1. Meiotic patterns in colchiploids of *Tithonia*:
1, 2) diakinesis and MI, 17II; 3) AI, 17:17; 4) diakinesis
C<sub>0</sub>, 6IV + 22II; 5, 6) MI C<sub>0</sub>, 9IV + 15II + 2I, and 14IV +
5II + 2I; and 7, 8), MI, C<sub>1</sub>, 6IV + 22II and 4IV + 26II.

some cases it has been observed that though there was no appreciable change in bivalent vis-a-vis quadrivalent frequencies for many generations, yet seed fertility increased, as was also observed in *Trifolium hybridum* [20]. If bivalent formation under normal meiosis is the sole cause of high fertility, good fertility should be expected in the cases with predominant bivalent formation at the initial stage. However, other factors e.g. genetic constitution, may also have a role in determining seed fertility [12]. In synthetic polyploids sterility may also be due to physiological reasons. In the present case, average number of 8.5 quadrivalent were observed, six of which were either ring or chain.

The present investigation shows that colchitetraploidy enhances ornamental characteristics of *T. speciosa* for features such as bigger capitula, long lasting and increased number of florets/capitulum.

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