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Induced Polyploidy via Colchicine Treatment Increases Flower Size and Fruit Weight in Cape Gooseberry (*Physalis peruviana* L.)

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Abstract: An induced polyploid plant through colchicine treatment offers probably the best scope for improvement in flower size and fruit weight. Thus, in the present study, an attempt was made to induce polyploidy in Cape gooseberry using colchicine with the objective of creating more genetic variability. The colchicine concentrations were used as 0.10% (C_1), 0.20% (C_2) and 0.40% (C_3) for the duration 12 (H_1), 24 (H_2) and 36 (H_3) hours for each concentration with seedling apex dip method (M_1), cotton plug method (M_2) and lanolin paste method (M_3). The plants treated with 0.10% of colchicine by cotton plug method for 12 h showed the better performance during the years 2017-2018 and 2018-2019 in respect of more delay in the flower bud emergence (54 d and 53 d from the date of transplanting), anthesis (19 d and 20 d from the first appearance of bud to full anthesis of flower) and fruit setting (8.00 d and 9.00 d from the date of anthesis), bigger flower size (2.93 cm^2 and 3.00 cm^2) than the untreated plants. The lower percentage of pollen viability (40.33% and 40.67%) was noticed in the same treatment in comparison to control (70.33% and 72.33%). The fruit maturity was also extended (59 d and 60 d from the date of fruit set) with bigger sized fruits (length: 2.53 cm and 2.57 cm, breadth: 2.27 cm and 2.33 cm) as well as more fruit weight (8.70 g and 8.33 g) by the application of colchicine at 0.10% with cotton plug method for 12 h. On the basis of results obtained, the application of colchicine at 0.10% for 12 h duration with cotton plug method was found to be the best and effective treatment for induction of polyploidy as well as more flower size and fruit weight in Cape gooseberry.

Key words: Cape gooseberry, colchicine, polyploid, flower size, fruit weight.

1. Introduction

Cape gooseberry (*Physalis peruviana* L.) is a minor and short duration growing fruit crop, belongs to the family Solanaceae and is generating appreciable income to the farmers during the recent time. It is native to Peru of South America but cultivated in South Africa in the region of Cape of Good Hope during the 19th century. It was introduced in India in the 19th century by the early European settlers. It is commonly called as Poha in Hawaii, Golden Berry in South Africa and Rasbhari, Makoi or Tepari in India. The first description of *Physalis* genus was made by Linnaeus in 1753 [1]. The genus *Physalis* has more than 100 species but only few are of economic value. One is the strawberry tomato, husk tomato or ground cherry, *P. pruinosa* L., grown for its small yellow fruits used for sauce, pies and preserves in mild-temperate climates. Muntzing (1951) [2] suggested that, Cape gooseberry is tetraploid in nature and has chromosome number 2*n* = 48. The main stem is green, herbaceous and composed of 8-12 nodes, giving origin to productive ramifications by dichotomy. Flowers are unique, pedunculate and hermaphrodite, derived from the auxiliary bud with five yellow petals. Calyx is green, formed by five sepals around 5 cm long, covering completely the fruit throughout its development. When the fruit is ripened, calyx shows a brown colour which

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is an indicator for determining the point of harvest [3]. *Physalis* seeds have high germination percentage and take around 10-15 d to germinate [4]. *Physalis* seeds germinate more easily when the temperature becomes 7-13 °C at night and 22-28 °C during the day. It can grow around 1.0-1.5 m height. However, with training it can exceed up to 2.0 m height [5].

Cape gooseberry is famous for its flavour and having good blend of acid-sugar. The fruits are very attractive in colour at maturity time and if properly packed, it can easily be sent to distant markets. The fruit type is berry, like a small globe having colour from green to yellowish, with the diameter around 12.5-25.0 mm and a weight ranges from 4 g to 10 g, containing around 100-300 seeds. *Physalis* fruits show high level of ascorbic acid (36 mg/100 g pulp), and are rich in vitamin A (1,730 IU/100 g pulp), iron (38 mg/100 g pulp) and phosphorus (1.2 mg/100 g pulp) [5]. The fruit is small round, bright orange and sweet when ripe, making it ideal for preparing delicious squash, nutritive jam and sweet pickles. A number of species in the genus are of horticultural and economic importance due to their high nutritional value in vitamin A, C and B complex, minerals and antioxidants as well as potential of medicinal properties including anti-bacterial, anti-inflammatory and anti-cancer properties.

Very little information is available on the crop as little work has been done. At present, there is neither any recognized variety nor have any serious efforts been made for improvement of this fruit. Improvement is very much desired in view of getting varieties with getting information of flowering and fruiting characters of Cape gooseberry.

Morphological, physiological and genetical change brought about by doubling of chromosome. Present day possibility for the artificial production of polyploids has opened a new vista for breeders. Several methods are employed to induce polyploidy through colchicines, viz, seedling apex dip method, cotton plug method and seed treatment as well as lanolin paste. It is generally noted that with increase in chromosome number the adaptability and variability of species increase progressively. The duplication of chromosome number, in general is associated with increased plant height, thick stem, broader as well as thicker leaves, dark green leaves, larger stomata size, bigger flowers and flower parts, increased pollen size, delayed flowering, late maturity of fruit, bigger fruit size, more vitamin C content and reduced seed number than diploid.

In the present investigation, attempt has also been made to develop tetraploids in this fruit. If this technique is standardized then the induced tetraploids can be crossed with normal diploids to produce desired traits of Cape gooseberry fruits. By keeping the above facts in the mind, the present investigation was carried out with the objective "induced polyploidy via colchicine treatment increases flower size and fruit weight in Cape gooseberry (*P. peruviana* L.)".

2. Materials and Methods

The present investigation was carried out in the experimental area of the Department of Horticulture, Birsa Agricultural University, Kanke, Ranchi, during two successive seasons (2017-2018 and 2018-2019). The experimental site comes under VIIth Agro-Climatic Region, i.e., Eastern Plateau and Hills. It is situated between 23°17′ North latitude and 85°19′ East longitude and the height from the mean sea level is 625 m. The soil of the experimental plot was sandy loam in texture with average fertility and thus considered suitable for cultivation of Cape gooseberry. The randomized block design was adopted for the trial. The number of treatment combinations was 22 with three replications during both the years.

2.1 Field Preparation

The field was prepared thoroughly. The required area was marked for experiment and land was again prepared thoroughly by spading to bring a fine tilth suitable for Cape gooseberry cultivation. A basal dressing of well rotten farm yard manure at the rate of two tractor trolleys full load per acre was applied and

was thoroughly incorporated in the soil. The sub-plots were then divided into different blocks according to the layout plan.

2.2 Nursery Bed

Seeds were sown on the raised bed with suitable mixture of garden soil and well rotten farm yard manure. Germination started visible after 9 d of sowing. The seedlings were ready for transplanting after a month of germination. Seedlings attained a height of 5-6 cm at the time of transplanting.

2.3 Seedling Transplanting

Seedlings were transplanted in the field during afternoon which was done manually in each sub-plot according to the layout plan with a planting distance of 50 cm × 50 cm, i.e., row to row and plant to plant, respectively. The plot size was maintained 2.00 m in both sides with accommodation of 16 plants per plot. To overcome the shock of transplanting, the transplanted seedlings were irrigated immediately with the help of a watering rose can. This practice was continued up to 7 d in both morning and evening hours.

2.4 Treatment Details

The colchicine concentrations used were 0.10% (C_1), 0.20% (C_2) and 0.40% (C_3) for the duration 12 (H_1), 24 (H_2) and 36 (H_3) hours for each concentration with seedling apex dip method (M_1), cotton plug method (M_2) and lanolin paste method (M_3).

2.5 Preparation of Chemicals

2.5.1 Colchicine Solution

Colchicine solutions of different concentrations were prepared in distilled water. For making 0.10%, 0.20% and 0.40% concentration, 100, 200 and 400 mg of colchicine was dissolved in separate glass beaker respectively in small quantity of absolute alcohol and then transferred to 100 mL measuring flask and distilled water was added to make required volume. The care was taken to keep the solution in dark place.

2.5.2 Lanolin Paste

The required amount of colchicine was measured and transferred to a Petri dish containing the required quantity of melted lanolin. Then it was mixed thoroughly with the help of a glass rod. The paste was allowed to cool down before application.

2.6 Methods of Treatment

2.6.1 Seedlings Apex Dips Method

Apex of one month old seedlings was dipped in known concentration of colchicine for a specific period. Roots of seedlings were protected by wrapping cotton swab. Water was poured on roots after some interval with the help of a dropper.

2.6.2 Cotton Plug Method

Small quantity of cotton was soaked in aqueous solution of colchicine of different concentrations by the help of glass rod. Soaked cotton of different concentrations was applied over growing apex of young and established seedlings for required duration. Treatments were repeated by dripping the solution with the help of a dropper after short interval.

2.6.3 Lanolin Paste Method

The paste containing different concentrations of colchicine was applied to the growing point of seedlings. The hairs and scales were removed from the growing point prior to application.

2.7 Observations Recorded

2.7.1 Floral Character

2.7.1.1 Period of Flower Bud Emergence

Each plant was closely watched for this purpose and as soon as the first flower bud emerged the date of its first appearance was recorded. The time required for appearance of flower bud was calculated from the date of transplanting. Five (5) flower buds were selected in each plot for calculation of the period of flower bud emergence and it was represented in days.

2.7.1.2 Duration of Anthesis

The five flower buds were tagged in each treatment from selected plants. The period required from the

date of first appearance of bud to full anthesis of flower was calculated and expressed in days.

2.7.1.3 Flower Size

The five fully opened flowers were taken. They were measured cross wise with the help of a scale and average was worked out.

2.7.1.4 Pollen Viability

It was assessed on the basis of stainability in acetocarmine (1%). The deeply stained ones were registered as viable while poorly stained ones were registered as non viable.

2.7.2 Fruiting Characters

2.7.2.1 Duration of Fruit Set

The time taken from date of anthesis to fruit set (pin head size) was noted and period required for fruit set was worked out and average was calculated in days.

2.7.2.2 Maturity Periods of Fruits

The time taken for maturity from the date of fruit set, i.e., a pin head stage was calculated for each tagged flower and the average period required for maturity was recorded in days. The maturity period of fruit was determined by light yellow colour of the calyx.

2.7.2.3 Fruit Size

The ripe fruits from each treatment were tagged and their length and breadth were measured with the help of vernier calipers and the average was worked out in centimeter.

2.7.2.4 Fruit Weight

The 10 fully matured fruits were weighed with the help of a physical balance. It was recorded from each tagged plant of each replication and the average was calculated each time.

3. Results

3.1 Flowering Characters

3.1.1 Period of Flower Bud Emergence

The data pertaining to the period of flower buds emergence in days as affected by the application of different methods of colchicine in various concentrations and for variable periods are presented in Table 1. In the first year (2017-2018) of experiment the more span of 53.00 d was taken in bud emergence from the date of transplanting by the virtue of the treatments $C_1M_2H_1$ and $C_1M_1H_1$ and it was statistically found at par with the treatments $C_2M_1H_1$, $C_3M_1H_1$, $C_1M_1H_3$, $C_1M_2H_3$ and $C_2M_2H_3$ with value of 52.00, 51.00, 51.00, 52.00 and 51.00 d, respectively, whereas the minimum time of 46 d was taken in control ($C_0M_0H_0$).

During the second year (2018-2019) of investigation, more or less similar results were obtained as previous year. The maximum of 54.67 d for the first emergence of flower bud was also recorded with the treatment $C_1M_2H_1$ and it was found statistically at par with the treatments $C_1M_1H_1$ and $C_1M_2H_3$ with numerical value of 54.00 d and 53.00 d, respectively. The minimum of 48 d was recorded by the control ($C_0M_0H_0$).

The pooled data of the both (2017-2018 and 2018-2019) also exhibited the similar trends of the both years. The maximum time of 53.83 d for the first emergence of flower bud was taken by the treatment $C_1M_2H_1$ and minimum of 47.00 d by the treatment control ($C_0M_0H_0$). The treatments $C_1M_1H_1$ and $C_1M_2H_3$ have value of 53.50 d and 52.50 d, respectively, for the first emergence of flower bud, which was found statistically at par with the treatment $C_1M_2H_1$.

These observations indicated that the period of flower bud emergence was delayed with the application of colchicine.

3.1.2 Duration of Anthesis

A close examination of the data in Table 1 indicated that the application of colchicine had responded well to enhance anthesis period in the treatments, during both the years (2017-2018 and 2018-2019) of investigation.

In the first year (2017-2018) of investigation the anthesis period after emergence of flower bud was found maximum of 19.33 d with the treatment $C_1M_2H_1$ followed by and at par with the treatment $C_1M_2H_3$, whereas the minimum time of 13.67 d was taken for anthesis by the treatment control ($C_0M_0H_0$).

During the following year (2018-2019) of experiment the similar results were also produced under the different treatments as previous year. This year also showed the maximum of 20.33 d for anthesis of flower buds with the effect of the treatment $C_1M_2H_1$ whereas the minimum time of 14.67 d was taken for anthesis in control ($C_0M_0H_0$).

The pooled results of the both years (2017-2018 and 2018-2019) showed the similar trends as previous both years. The maximum time of 19.83 d was taken for anthesis of flower buds with the effect of treatment $C_1M_2H_1$ (colchicine at 0.10% for 12 h with cotton plug method) which was found significantly superior over the rest of the treatments. The minimum time of 14.17 d was observed by the effect of control ($C_0M_0H_0$).

Thus, in the light of above findings, it can be concluded that the treatments delayed the anthesis as compared to control by the application of colchicine.

3.1.3 Flower Size

The flower size in respect of diameter (cm^2) of Cape gooseberry under the different treatments was measured and results have been presented in Table 1.

A critical examination of the data indicated that application of colchicine had marked effect on flower diameter of Cape gooseberry during the both years (2017-2018 and 2018-2019).

In the first year (2017-2018) of investigation, the maximum flower size 2.93 cm^2 was recorded in the treatment $C_1M_2H_1$ followed by the treatment $C_1M_1H_1$ with flower size 2.87 cm^2, whereas the minimum of 2.37 cm^2 was observed under the control ($C_0M_0H_0$).

During the next year (2018-2019) of experiment, the maximum flower size of 3.00 cm^2 was recorded by the treatment $C_1M_2H_1$, which was noticed statistically at

Table 1 Effect of colchicine on floral behavior of Cape gooseberry.

Treatment combination	Period of flower bud emergence (d)			Duration of anthesis (d)			Flower size (cm^2)		
	2017-2018	2018-2019	Pooled	2017-2018	2018-2019	Pooled	2017-2018	2018-2019	Pooled
T_1-$C_1M_1H_1$	53.00	54.00	53.50	17.33	18.67	18.00	2.87	2.93	2.90
T_2-$C_2M_1H_1$	52.00	52.00	52.00	16.00	15.67	15.83	2.60	2.53	2.57
T_3-$C_3M_1H_1$	51.00	52.00	51.50	15.00	16.00	15.50	2.53	2.50	2.52
T_4-$C_1M_1H_2$	50.00	51.00	50.50	15.67	17.67	16.67	2.67	2.70	2.68
T_5-$C_2M_1H_2$	49.00	50.00	49.50	15.00	15.33	15.17	2.57	2.53	2.55
T_6-$C_3M_1H_2$	48.00	49.00	48.50	15.33	15.00	15.17	2.50	2.50	2.50
T_7-$C_1M_1H_3$	51.00	51.00	51.00	15.67	16.33	16.00	2.53	2.57	2.55
T_8-$C_2M_1H_3$	49.00	50.00	49.50	15.33	15.67	15.50	2.50	2.53	2.52
T_9-$C_3M_1H_3$	50.00	51.00	50.50	15.33	15.00	15.17	2.47	2.53	2.50
T_{10}-$C_1M_2H_1$	53.00	54.67	53.83	19.33	20.33	19.83	2.93	3.00	2.97
T_{11}-$C_2M_2H_1$	50.00	51.67	50.83	15.67	16.67	16.17	2.57	2.53	2.55
T_{12}-$C_3M_2H_1$	48.00	49.00	48.50	15.00	16.00	15.50	2.50	2.57	2.53
T_{13}-$C_1M_2H_2$	50.00	51.00	50.50	15.00	15.33	15.17	2.53	2.53	2.53
T_{14}-$C_2M_2H_2$	50.00	50.00	50.00	14.33	15.67	15.00	2.47	2.57	2.52
T_{15}-$C_3M_2H_2$	49.00	50.00	49.50	15.33	15.67	15.50	2.43	2.53	2.48
T_{16}-$C_1M_2H_3$	52.00	53.00	52.50	18.33	19.00	18.67	2.77	2.87	2.82
T_{17}-$C_2M_2H_3$	51.00	51.00	51.00	15.33	15.33	15.33	2.57	2.53	2.55
T_{18}-$C_3M_2H_3$	49.00	52.00	50.50	14.67	15.00	14.83	2.53	2.57	2.55
T_{19}-$C_1M_3H_0$	49.00	50.00	49.50	15.00	16.00	15.50	2.50	2.53	2.52
T_{20}-$C_2M_3H_0$	49.00	50.00	49.50	15.33	15.00	15.17	2.47	2.57	2.52
T_{21}-$C_3M_3H_0$	49.00	49.00	49.00	14.33	15.33	14.83	2.40	2.53	2.47
T_{22}-$C_0M_0H_0$	46.00	48.00	47.00	13.67	14.67	14.17	2.37	2.47	2.42
SEm (±)	0.89	0.84	0.59	0.43	0.39	0.29	0.04	0.04	0.03
CD (p = 0.05)	2.54	2.40	1.66	1.23	1.11	0.82	0.13	0.10	0.08
CV (%)	3.09	2.86	2.87	4.79	4.18	4.55	2.97	2.45	2.58

par with the treatment $C_1M_1H_1$ with flower size 2.93 cm^2 whereas minimum flower size 2.47 cm^2 was exhibited by the treatment control ($C_0M_0H_0$).

The pooled results of the both years (2017-2018 and 2018-2019) showed the similar trends of the previous both years. The maximum flower size 2.97 cm^2 was recorded by the effect of treatment $C_1M_2H_1$, which was noticed statistically at par with the treatment $C_1M_1H_1$ with flower size 2.90 cm^2. The minimum flower size 2.42 cm^2 was registered in the control ($C_0M_0H_0$).

In view of the aforesaid results, it indicated that the application of colchicine had maximum potentiality to increase the flower size (diameter) in Cape gooseberry.

3.1.4 Pollen Viability

The calculated pollen viability percentage as influenced by the application of colchicine in different concentrations with various methods for variable duration is presented in Fig. 1.

During the first year (2017-2018) of experimentation the minimum of 40.33% pollen viability was obtained by the effect of treatment $C_1M_2H_1$ which showed superiority over the all treatments including control ($C_0M_0H_0$). The maximum pollen viability of 70.33% was recorded under the treatment control ($C_0M_0H_0$).

In the subsequent year (2018-2019) of trial the trend of results was obtained more or less similar as previous year. The minimum pollen viability of 40.67% was observed in the treatment $C_1M_2H_1$ whereas the maximum pollen viability of 72.33% was noted under the treatment control ($C_0M_0H_0$).

The pooled results of the both years (2017-2018 and 2018-2019) also showed the more or less similar trends. The minimum of 40.50% was observed with the effect of the treatment $C_1M_2H_1$ whereas the maximum pollen viability of 71.33% was produced by the treatment control ($C_0M_0H_0$).

The results obtained for this character clearly indicated that application of colchicine had affected the pollen viability also.

3.2 Fruiting Characters

3.2.1 Duration of Fruit Set

The duration of fruit set was influenced by different treatments as presented in Table 2. During the first year (2017-2018) of investigation the maximum of 8.00 d was taken in fruit set after anthesis with the effect of the treatment $C_1M_2H_1$ followed by the treatment $C_1M_1H_1$ with the value of 7.33 d whereas the minimum of 4.33 d was taken by control.

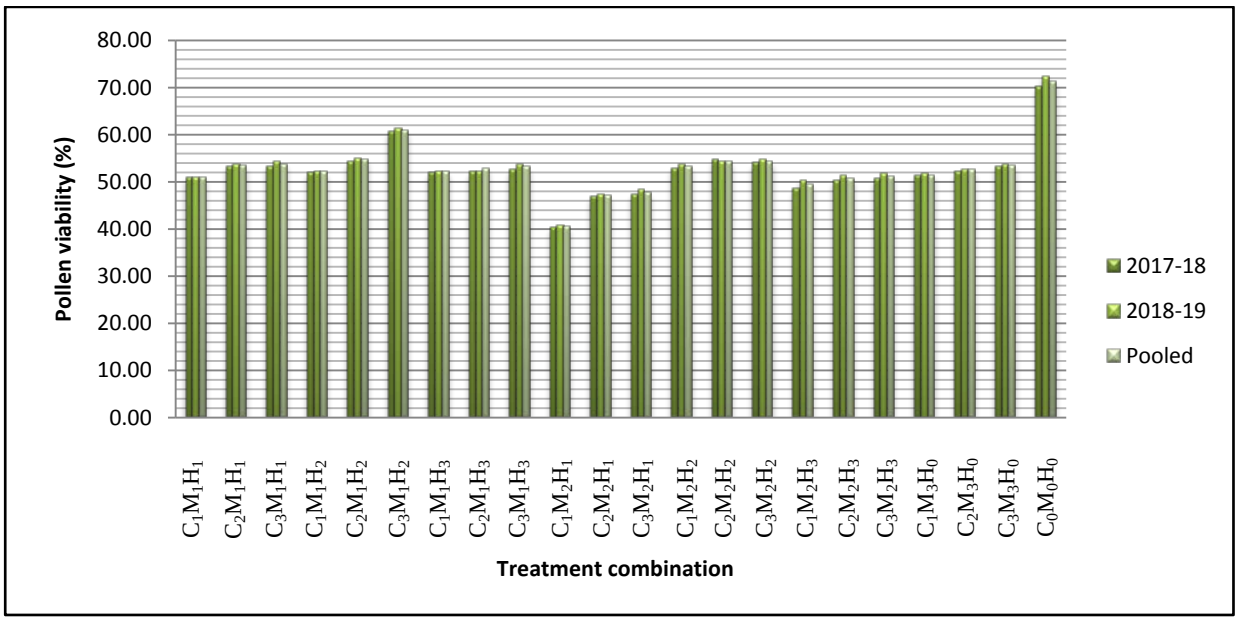

Fig. 1 Pollen viability (%) (CD (p = 0.05) 2017-2018: 5.22, 2018-2019: 5.58, pooled: 3.38).

Table 2 Effect of colchicine on fruit setting and fruit maturity of Cape gooseberry.

Treatment combination	Duration of fruit set (d)			Maturity period of fruit (d)		
	2017-2018	2018-2019	Pooled	2017-2018	2018-2019	Pooled
T_1-$C_1M_1H_1$	7.33	8.67	8.00	57.67	59.00	58.33
T_2-$C_2M_1H_1$	5.33	5.67	5.50	55.67	53.67	54.67
T_3-$C_3M_1H_1$	5.00	6.00	5.50	54.67	53.00	53.83
T_4-$C_1M_1H_2$	6.33	7.67	7.00	56.33	55.33	55.83
T_5-$C_2M_1H_2$	5.33	6.33	5.83	54.33	52.00	53.17
T_6-$C_3M_1H_2$	5.33	5.67	5.50	54.33	51.33	52.83
T_7-$C_1M_1H_3$	5.33	6.33	5.83	55.00	53.67	54.33
T_8-$C_2M_1H_3$	5.33	5.67	5.50	54.33	53.33	53.83
T_9-$C_3M_1H_3$	5.00	6.00	5.50	54.00	53.67	53.83
T_{10}-$C_1M_2H_1$	8.00	8.67	8.33	58.67	60.33	59.50
T_{11}-$C_2M_2H_1$	5.33	6.00	5.67	56.67	54.00	55.33
T_{12}-$C_3M_2H_1$	4.67	6.33	5.50	55.67	54.33	55.00
T_{13}-$C_1M_2H_2$	5.33	6.33	5.83	54.33	53.67	54.00
T_{14}-$C_2M_2H_2$	5.00	6.33	5.67	53.67	52.67	53.17
T_{15}-$C_3M_2H_2$	5.00	5.67	5.33	53.33	52.67	53.00
T_{16}-$C_1M_2H_3$	6.33	7.33	6.83	57.33	58.67	58.00
T_{17}-$C_2M_2H_3$	4.67	6.33	5.50	55.67	54.67	55.17
T_{18}-$C_3M_2H_3$	5.33	5.33	5.33	52.67	54.33	53.50
T_{19}-$C_1M_3H_0$	5.00	6.33	5.67	55.00	54.33	54.67
T_{20}-$C_2M_3H_0$	4.67	6.00	5.33	55.33	53.33	54.33
T_{21}-$C_3M_3H_0$	4.67	5.67	5.17	55.67	52.67	54.17
T_{22}-$C_0M_0H_0$	4.33	4.67	4.50	45.33	45.00	45.17
SEm (±)	0.32	0.30	0.29	1.57	1.22	0.91
CD (p = 0.05)	0.91	0.86	0.81	4.49	3.48	2.56
CV (%)	10.21	8.33	12.05	4.97	3.92	4.12

In the next year (2018-2019) of observations the similar trend of the result was recorded as previous year. The maximum days in fruit set were exhibited by the treatments $C_1M_2H_1$ and $C_1M_1H_1$ by the same footing value of 8.67 d in fruit set after anthesis. The minimum of 4.67 d was taken in fruit set after anthesis in the treatment control.

The pooled data of the both years (2017-2018 and 2018-2019) exhibited the similar trends of both years. The maximum of 8.33 d after anthesis was taken by the treatment $C_1M_2H_1$ for fruit set which showed statistically at par with the treatment $C_1M_1H_1$ with the value of 8.00 d for fruit set. The minimum of 4.50 d was taken by the treatment control.

Thus, from the above findings, it is evident that the methods, concentrations and duration of treatments of colchicine affected the duration of fruit set in Cape gooseberry.

3.2.2 Maturity Period of Fruit

A careful scanning of the data in Table 2 reflected that the maturity period in days after fruit set was affected by application of colchicine in different concentrations for variable duration with various methods when it was compared to control. In the first year (2017-2018) of experimentation the maximum of 58.67 d span was observed in maturity after fruit set with the treatment $C_1M_2H_1$, which was found statistically at par with all of the other treatments except $C_3M_1H_3$, $C_2M_2H_2$, $C_3M_2H_2$, $C_3M_2H_3$ and control ($C_0M_0H_0$) with the value of 54.00, 53.67, 53.33, 52.67 and 45.33 d, respectively. The minimum 45.33 d was taken in control ($C_0M_0H_0$).

During the subsequent year (2018-2019) of the investigation more or less similar results were obtained as previous year. The maximum of 60.33 d was taken in fruit maturity after fruit set with the treatment

$C_1M_2H_1$ which was found statistically at par with the treatments $C_1M_1H_1$ and $C_1M_2H_3$ with values of 59.00 d and 58.67 d in fruit maturity, respectively. The minimum period of 45.00 d was counted under $C_0M_0H_0$.

The pooled result also showed the similar trend of the both years (2017-2018 and 2018-2019). The maximum of 59.50 d was recorded by the treatment $C_1M_2H_1$ and it was found statistically at par with the treatments $C_1M_1H_1$ and $C_1M_2H_3$ with value of 58.33 d and 58.00 d in fruit maturity, respectively. The minimum 45.17 d was noticed by the treatment control ($C_0M_0H_0$).

On the basis of the above findings it can be concluded that the application of colchicine with various methods in different concentrations and for variable period had capacity to extend maturity period of fruits in Cape gooseberry.

3.2.3 Fruit Size

3.2.3.1 Fruit Length

The data of both the years (2017-2018 and 2018-2019) of investigation were analyzed statistically and it was evident from Table 3 that the application of colchicine was effective in increasing fruit length over control.

During the first year (2017-2018) of the experimentation the maximum fruit length of 2.53 cm was observed with the effect of the treatment $C_1M_2H_1$ and statistically it was found superior over the remaining treatments. The next superior treatment was recorded 2.43 cm with $C_1M_1H_1$. The minimum fruit length 1.75 cm was noted under the treatment control.

In the next year (2018-2019) of the observation more or less similar trend of the results was calculated. The treatments $C_1M_1H_1$ and $C_1M_2H_1$ showed the same footing result of 2.57 cm fruit length in both treatments and statistically they were found most effective in enhancing the fruit length over the other treatments. The minimum length 1.82 cm was observed under the treatment control ($C_0M_0H_0$).

The combine pooled data of the both years (2017-2018 and 2018-2019) also approved $C_1M_2H_1$ as the most effective treatment having value of 2.55 cm. The minimum fruit length 1.78 cm was observed in the control ($C_0M_0H_0$).

In the view of aforesaid observations, it can be derived that application of colchicine with the treatment $C_1M_2H_1$ had the capacity to enhance the fruit length.

3.2.3.2 Fruit Breadth

The close study of the data in Table 3 transpired that application of colchicine was responsive in increasing the fruit breadth in Cape gooseberry. Considering the effect of different treatments in the first year (2017-2018) of observation it was evident that the treatment $C_1M_2H_1$ produced the fruits of maximum breadth 2.27 cm followed by 2.17 cm with the treatment $C_1M_2H_3$. The minimum 1.57 cm fruit breadth was recorded under control ($C_0M_0H_0$).

During the second year (2018-2019) of investigation the maximum of 2.33 cm fruit breadth was recorded by the effect of the treatment $C_1M_2H_1$ and it was found at par with the treatments $C_1M_2H_3$ and $C_1M_1H_1$ having value of 2.27 cm and 2.23 cm fruit breadth respectively whereas the minimum breadth 1.67 cm was under the control ($C_0M_0H_0$).

The pooled analyzed data of the both years (2017-2018 and 2018-2019) also showed the more or less similar trends as previous years. The treatment $C_1M_2H_1$ was found statistically superior with the value of 2.30 cm fruit breadth in comparison to other treatments except the treatment $C_1M_2H_3$ which gave the at par result of 2.22 cm. The control ($C_0M_0H_0$) showed the minimum value of 1.62 cm fruit breadth.

On the basis of above results, it can be concluded that application of colchicine 0.10% for 12 h or 36 h with cotton method produced the maximum fruit length and breadth.

3.2.4 Fruit Weight

A careful scrutiny of data in Table 3 reflected that fruits having more weight were obtained under plants treated with colchicine in both the years (2017-2018 and 2018-2019) of experimentation and as well in their combined analysis, too.

Table 3 Effect of colchicine on physical characters of Cape gooseberry.

Treatment combination	Fruit length (cm)			Fruit breadth (cm)			Fruit weight (g)		
	2017-2018	2018-2019	Pooled	2017-2018	2018-2019	Pooled	2017-2018	2018-2019	Pooled
T_1-$C_1M_1H_1$	2.43	2.57	2.50	2.13	2.23	2.18	8.20	8.10	8.15
T_2-$C_2M_1H_1$	2.17	2.23	2.20	2.03	2.07	2.05	7.10	7.17	7.13
T_3-$C_3M_1H_1$	2.13	2.17	2.15	1.93	1.97	1.95	7.05	6.97	7.01
T_4-$C_1M_1H_2$	2.23	2.32	2.28	2.07	2.13	2.10	7.45	7.90	7.68
T_5-$C_2M_1H_2$	2.08	2.13	2.11	1.93	2.03	1.98	7.30	7.17	7.23
T_6-$C_3M_1H_2$	2.07	2.13	2.10	1.87	1.97	1.92	7.20	7.07	7.13
T_7-$C_1M_1H_3$	2.17	2.23	2.20	1.97	2.07	2.02	7.30	7.13	7.22
T_8-$C_2M_1H_3$	2.08	2.17	2.13	1.93	2.03	1.98	7.25	7.17	7.21
T_9-$C_3M_1H_3$	2.07	2.13	2.10	1.87	1.97	1.92	7.15	7.07	7.11
T_{10}-$C_1M_2H_1$	2.53	2.57	2.55	2.27	2.33	2.30	8.70	9.07	8.88
T_{11}-$C_2M_2H_1$	2.17	2.23	2.20	2.03	2.07	2.05	7.05	7.23	7.14
T_{12}-$C_3M_2H_1$	2.13	2.17	2.15	1.97	2.03	2.00	7.05	7.20	7.13
T_{13}-$C_1M_2H_2$	2.13	2.13	2.13	1.97	2.03	2.00	7.00	7.17	7.08
T_{14}-$C_2M_2H_2$	2.07	2.17	2.12	1.93	2.03	1.98	6.95	7.13	7.04
T_{15}-$C_3M_2H_2$	2.08	2.13	2.11	1.93	2.03	1.98	6.80	7.07	6.93
T_{16}-$C_1M_2H_3$	2.27	2.43	2.35	2.17	2.27	2.22	8.70	8.33	8.52
T_{17}-$C_2M_2H_3$	2.17	2.23	2.20	1.97	2.07	2.02	7.15	7.17	7.16
T_{18}-$C_3M_2H_3$	2.07	2.13	2.10	1.93	2.03	1.98	7.15	7.07	7.11
T_{19}-$C_1M_3H_0$	2.17	2.20	2.18	1.93	2.03	1.98	7.05	7.13	7.09
T_{20}-$C_2M_3H_0$	2.13	2.17	2.15	1.87	1.97	1.92	7.05	7.03	7.04
T_{21}-$C_3M_3H_0$	2.08	2.17	2.13	1.83	1.93	1.88	6.95	7.03	6.99
T_{22}-$C_0M_0H_0$	1.75	1.82	1.78	1.57	1.67	1.62	5.92	6.00	5.96
SEm **(±)**	0.03	0.03	0.02	0.03	0.04	0.03	0.34	0.38	0.23
CD (p = 0.05)	0.08	0.10	0.07	0.09	0.10	0.08	0.97	1.09	0.64
CV (%)	2.12	2.70	2.76	2.83	3.05	3.55	8.11	9.08	7.71

Considering the effect of different treatments on the first year (2017-2018) of experimentation, it was evident that $C_1M_2H_1$ and $C_1M_2H_3$ produced fruits with maximum weight of 8.70 g, which was found on the same footing value, whereas 8.20 g fruit weight in $C_1M_1H_1$ was found at par with these treatments. The minimum fruit weight of 5.92 g was recorded under the treatment control ($C_0M_0H_0$).

In the second year (2018-2019) of investigation, more or less similar trend of the results was observed as in previous year. The maximum fruit weight of 9.07 g was recorded in the treatment $C_1M_2H_1$ which was found statistically at par with the treatments $C_1M_1H_1$ and $C_1M_2H_3$ with the values of 8.10 g and 8.33 g fruit weight, respectively. The minimum of 6.00 g was noted under the treatment control ($C_0M_0H_0$).

The pooled analysis of the two years data (2017-2018 and 2018-2019) showed that treatments differed significantly in increasing the fruit weight. The maximum of 8.88 g fruit weight was noted in the treatment $C_1M_2H_1$, which was found statistically at par to $C_1M_2H_3$ with the value of fruit weight 8.52 g. The minimum fruit weight of 5.96 g was observed under the treatment control ($C_0M_0H_0$).

On the basis of the results obtained for this attribute it can be inferred that the treatment $C_1M_2H_1$ produced the heaviest fruits in comparison to other treatments. The next effective treatment in this regard was found $C_1M_2H_3$ with the bigger fruit weight.

4. Discussion

In regards to delay in flower bud emergence (53.83 d) and anthesis (19.83 d) of colchicine treated plants in comparison to control (47.00 d and 14.17 d),

it was indicated in the present observations that application of colchicine extended the period of flower bud emergence and duration of anthesis as compared to untreated plant. It happened due to bigger stomata cells and a decrease in stomata number per unit area in tetraploids. In recent year studies, most of researchers examined the size of the stomata cells, and reported bigger cells in polyploids. The results obtained are in conformity with the findings of Gu *et al.* [6] in *Zizyphus jujuba* Mill. cv. Zhanhua, Tulay and Unal [7] in *Vicia villosa* roth, Kushwaha *et al.* [8] in *Chrysanthemum carinatum* L. and Luo *et al.* [9] in rubber plant.

The ultimate size of flowers (2.97 cm^2) was considerably larger in colchicine treated plants than untreated plant (2.42 cm^2). It might be due to accumulation of more food materials in colchicine treated plant through larger size of leaf resulting in large flower in Cape gooseberry. The rate of photosynthesis depends upon the size of leaf and chlorophyll content. These results obtained are in agreement with the findings of Ascough and Staden [10] in *Watsonia lepida*, Gu *et al.* [6] in *Zizyphus jujuba*, He *et al*. [11] in *Chrysanthemum*, Kwon *et al.* [12] in *Prunella vulgaris* for *albiflora* Nakai, Manzoor *et al.* [13] in *Gladiolus grandiflorus*, Muhammad *et al.* [14] in watermelon and Shao *et al*. [15] in pomegranate.

It was observed that the pollen grain viability (40.50%) in Cape gooseberry flowers was decreased over untreated plant (71.33%). The minimum viability percentage was caused by the application of aqueous solution of colchicine at 0.10% for 12 h with cotton plug method followed by 0.20% for 12 h with cotton plug method. The main cause behind these findings was reduction of pollen fertility in colchicine treated plant due to multivalent association during synapsis of chromosome and the consequent production of unbalance gametes. Meiotic abnormalities are also one of the causes of low pollen grain viability. The viability in auto as well as allopolyploids was influenced not only by the presence or absence of multivalent but also by other genetic factors. The similar observations were earlier reported by Biswas and Bhattacharyya [16] in French bean, Glowacka *et al*. [17] in *Miscanthus* species, Kadota and Niimi [18] in Japanese pear, Omidbaigi *et al.* [19] in dragonhead, Shao *et al*. [15] in pomegranate, Sohoo *et al.* [20] in chickpeas and Zhang *et al.* [21] in *Trollius chinensis* Bunge.

It is evident from the present investigation that application of different colchicine treatments extended time of fruit set (8.33 d after anthesis) and maturity of fruit (59.50 d after fruit set) in comparison to untreated plant (4.50 d and 45.17 d, respectively). It may be due to the physical injury at initial stage as well as also due to the genetical imbalance caused by colchicine chemical. This may also be due to large flower and fruit size, which required more time thereby delaying fruit maturity. The similar findings were recorded by Gu *et al.* [6] in *Zizyphus jujuba* Mill. cv. Zhanhua, Kushwaha *et al.* [8] in *C. carinatum* L. and Luo *et al.* [9] in rubber plant.

Increase in fruit weight (8.88 g) in comparison to control (5.96 g) is attributed to increase in cell size; cell number and volume of inter cellular space in the flesh which has enabled the maximum accumulation of water and food substances by the lower application of colchicine. Further, mixoploidy induced by colchicine treatment in cannabis showed better growth as compared to tetraploid plants. It is well known that polyploidy leads to an increase in organ size, which may be caused by changes in activities of cell division and expansion as the result of the duplication of gene loci and increase in nuclear deoxyribonucleic acid (DNA) content. Further, increase in fruit weight due to colchicine application indicates that colchicine concentrations bring about certain change in the metabolism of fruit, which are reflected in more accumulation of food constituents in the fruit and thus increase fruit weight of individual fruit as well as fruit yield per plant. It was supported by the researchers

Amiri *et al.* [22] in Datura, Glowacka *et al*. [17] in *Miscanthus* species, Hayashi and Yoshida [23] in *Glycine max*, Lindayani *et al.* [24] in *Zingiber ofjicinale*, Liu *et al*. [25] in pumpkin, Manawadu *et al.* [26] in radish, Nezhad and Mansouri [27] in *Dunaliella salina*, Sugiyama [28] in two grass species of *Lolium* and Vijayalakshmi and Singh [29] in cluster bean.

5. Conclusions

In the view of aforesaid observations, it can be safely concluded that among the different treatments lower concentrations of colchicine for minimum duration with cotton plug method performed better in respect of delayed flower bud emergence, anthesis and fruit set, maturity of fruits, larger flower size and more fruit weight than the control. Further, it was also observed that lower pollen viability, less number of fruits per plant with colchicine treated plant was obtained than the untreated plant. But due to higher value of fruit weight in the colchicine treated plant showed the more fruit yield per plant in comparison to control.

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