MUTAGENIC EFFECTS OF COLCHICINE ON THE MORPHOLOGY AND YIELD OF THREE TOMATO (S*OLANUM LYCOPERSICUM L.*) ACCESSIONS

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ABSTRACT

The mutagenic effect of Colchicine on the morphology and yield of tomato was investigated. Seeds of three tomato accessions (NG/MR/May/09/005-SouthWest, NG/SA/07/10/002-NorthEast and NG/CHU/Jun/0201-SouthEast) were presoaked in four different concentrations of Colchicine (0.1mM, 0.3mM, 0.5mM, 1.0mM) and 0.0mM as control. The domestic demand for tomato extremely exceeds the supply with over 2-3 million metric tons annually thereby involving Nigeria into importing about 300,000 metric tonnes worth over $360 million dollar yearly. The experiment was laid in Randomized Complete Block Design in five replications. The morphological parameters used to characterize the mutants were plant height, number of leaves/plant, leaf length, number of branches, number of days to first flowering, fruit weight, germination percentage and number of fruit/plant. The results revealed significant difference (p<0.05) in some of the morphological traits assessed when compared with the controls, except in the number of branches and plant height, where the effect of the mutagen was not important (p>0.05). The morphological and yield parameters of the southwest and northeast accessions of tomato as observed in this study, improved with decreasing concentration of Colchicine, while the southeast improved with increase in the concentration of Colchicine. It is thus recommended that, to improve growth and yield parameters in tomatoes, 0.1mM concentration should be employed to improve Southwest and Northeast tomato accessions, while 0.5mM or 1.0mM concentrations should be used in the Southeast tomato accessions.

Keywords: Accessions, Colchicine, Morphology, Tomato, Yield

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INTRODUCTION

Tomato (*Solanum lycopersicum*) is the edible, often red berry-type fruit of the nightshade or Solanaceae family. It develops from the ovary of the plant after fertilization and its flesh comprises the pericarp walls. The fruit contains hollow spaces full of seeds and moisture called locular cavities (Krishna, 2007). It is herbaceous, usually sprawling plant as its close cousins are tobacco, chilli peppers and eggplant. The tomato is consumed in diverse ways, such as an ingredient in many dishes, made into paste for canned tomato, ketchups, stew, sauces, salads, and drinks (Babalola et al., 2010).

Tomato production has long been overlooked by many Nigerians who choose to venture into agriculture for the first time. Tomato, an ignored vegetable crop in Nigeria, has remained a constant ingredient for the preparation of multiple meals in the country. Unfortunately, in nearly three decades, no tomato variety (Variant) has been released in Nigeria (Nwosu et al., 2014). About half of the harvests are lost due to spoilage and several other factors before and after they get to the market (Babalola et al., 2010). Some in-country efforts at improving the tomato crop include the studies on the development of regeneration protocols for local cultivars of tomato (Ajenifujah-Solebo et al., 2012). Therefore, the need to explore and enhance the production capabilities and potentials of long forgotten indigenous land races and other ecotypes has never been more urgent than now.

Mutation breeding technology has proved to be one of the effective ways of crop improvement. The use of chemical mutagens like Colchicine, has proved to be an efficient and effective tool for the improvement of the quality and yield of imported tomato variety (Roma VF) thus, indicating the possibility of isolating beneficial mutants with indigenous genotypes (Adelanwa & Habeeb, 2011). Reduction in losses due to spoilage in selected indigenous variants with traits could be achieved through mutation and selection, thus producing varieties with high yield robust fruits with long shelf life that will command high market value.

Colchicine is a powdery mutagenic agent used for inducing polyploidisation and inhibiting mitotic process for crop improvement (Mensah & Obadoni, 2006). The effects of mutagens in crops can be discovered through cytology which is the study of cells of living organisms. The cells of living organisms which come in contacts with mutagens often show the evidence of polyploidy as seen in forage sorghum cultivars (Maria & Sakunkan, 2018). Polyploidy is the condition in which a normally diploid cell or organism acquires one or more additional sets of chromosomes. Polyploidy in general tend to have larger cells compared to their diploid progenitors, retarded metabolism and growth, alter genomic relations between the nucleus and cell wall, and they also seem to have reduced number of cell division during development (Sander et al., 2019).

Therefore, this study becomes necessary to assess the mutagenic effect of Colchicine in effective improvement of this valuable and widely cultivated crop in Nigeria. Keeping all these facts in mind, the present research was designed to evaluate the effect of colchicine on the morphology and yield parameters of indigenous tomato accessions.

MATERIALS AND METHODS

Source of Materials

The three tomato accessions (NG/MR/May/09/005-South West, NG/SA/07/10/002-North East and NG/CHU/Jun/0201-South East) used for this study were collected from National Centre for Genetics Resources and Biotechnology (NAGRAB), Ibadan, and the chemical mutagen, (Colchicine) was obtained from Sigma Company, India.

Seed Viability and mutagenic treatment of the selected seeds were carried out in the Plant Biotechnology Laboratory of the Department of Agricultural Biotechnology, National Biotechnology Development Agency (NABDA), Abuja. Randomly selected seeds were divided into five groups of fifty seeds each. Each group was treated with four different concentrations (0.1mM(A), 0.3mM(B), 0.5mM(C) and 1.0mM(D)) of Colchicine respectively and the last being the control was soaked with ordinary distilled water. They were all subjected to these treatments for 24 hours. The seeds were tested for viability using the method of Stephen (2009). Prior to the treatment, the seeds were pre-soaked in distilled water to activate their bio-activity. The plated seeds were then observed for germination and the number of germinated seedling counted. The percentage germination was then calculated using the formula below;

$$\%\ \text{germination} = \frac{Total number of germinated}{\text{Total number of seeds used}} \times 100 \quad \text{(Stephen, 2009).}$$

Evaluation of Morphological and Yield Parameters

Five seeds from each treatment and control were sown in pots filled with sandy-loamy soil followed by regular watering in the screen house of NABDA. The seedlings were later thinned to one seedling per pot at 2 weeks after sowing (WAS) (Dauda & Falusi, 2011). Data such as leaf length, plant height at maturity, number of branches per plant, number of days to first flowering, fruit weight and number of fruit per plant were then collected using standard procedure (Dauda & Falusi, 2011).

Statistical Analysis

The morphological data collected was subjected to analysis of variance (ANOVA) to test significant differences at 5% probability level and Duncan multiple range tests were used to separate the means where there were differences.

RESULTS

Effects of Colchicine on the Germination Percentage of Tomato Accessions

The three accessions responded differently to the varying concentrations of Colchicine applied. In the Southwest (SW) accession, there was no significant difference ($p<0.05$) in the germination percentage for treatments A, C, D and the control (E) except for treatment (B) Colchicine concentration which had 53.33% germination percentage while all other treatments had 60.00% (Table 1). In the North east (NE) accession, there was high significant difference ($P>0.05$) among the treatments. The highest mean value (93.33%) was recorded in the A and C treatments. The lowest germination percentage (60.00%) was recorded in the D treatment which is significantly different from other treatments. Also, the control for Southeast (SE) accession had the highest mean germination percentage of 73.77%. The lowest germination percentage of 46.67% was recorded in B and C treatment which were significantly different from the other treatment.

Effects of Colchicine on the Number of Branches of Tomato Accessions

The results of the effect of Colchicine on the number of branches of tomato showed that treatment C produced the highest number of branches in all the three accessions (Table 1). In the southwest (SW) accession, C had the highest number of branches with a mean value of 1.60. This value was significantly different from the values of other treatments. The lowest number of branches 0.80 was recorded in the control E, which is significantly different from other treatments.

In the Northeast (NE) accession, treatment C produced the highest number of branches with a mean value of 1.60. This value was significantly different from the value of other treatments except for treatment D. The control (E) and B treatments had the least number of branches with mean value of 1.00. These values were significantly different from the other treatments. For the Southeast (SE) accession, treatment C had the highest number of branches (1.60) which is significantly different from other treatments except D. The least number of branches (0.80) was recorded in the control which was significantly different from all other treatments.

Effects of Colchicine on the Plant Height of Tomato Accessions

The result of the effects of Colchicine on the plant height of tomato accessions at Week 8 is shown in Table 1. There was significant difference between the plant heights for all the treatments in SW Accession. The highest height of (30.70 cm) was recorded in treatment C and the lowest (15.24 cm) was recorded in the control. For NE Accession, the control had the highest plant height (34.64 cm). This value was significantly different from other treatments except treatment A. The lowest (15.68 cm) was found in treatment C which is significantly different from all other treatments. There was significant difference between the plant heights of all the treatments in SE Accession. The highest (30.10 cm) was recorded in treatment D while the lowest (11.28 cm) was found in treatment A.

Effects of Colchicine on the Leaf Length of Tomato Accessions

The result of the SW Accession exposed to 0.1 mM (A) colchicine showed the highest leaf length (12.10 cm) while the least (8.08 cm) was recorded in treatment E (control). For NE Accession, treatment A recorded the highest leaf length (10.84 cm), this value was significantly different from other treatments except treatment B. A significantly low leaf length (5.48 cm) was recorded in treatment C. There was significant difference between the leaf lengths of all the treatments of SE Accession. The highest (11.20 cm) was recorded in treatment D while the least (6.88 cm) was recorded in treatment A.

Effects of Colchicine on the number of leaves of Tomato Accessions

The results indicated that in SW accession, there was no significant difference between the treatments at week 2. The same number of leaves (4.00) were recorded in treatment B and C as the highest number of leaves. At week 8, the highest number of leaves (12.60) was recorded in treatment C, this value was significantly different from other treatments. On the other hand, the lowest number of leaves (8.20) was recorded in the control which was also significantly different from other treatments. However, in the NE Accession, the results showed that there was significant difference among all the treatments with the highest number of leaves (15.00) recorded in treatment A and the lowest (9.80) recorded in treatment C. SE accession with treatment D had the highest number of leaves (11.40), this value was significantly different from other treatments except the control (E). A significant lower number of leaves were observed in treatment A (8.20) as well.

Effects of Colchicine on the Number of Days to First Flowering

The results of the effect of Colchicine on the number of days to first flowering are shown in Table 2. The results revealed that the control E had the highest mean number of days to first flowering (79.40) in SW Accession. The least (65.00) was recorded in treatment C which was significantly different from other treatments but statistically the same with treatment A. In the NE accession, 79.20 was recorded as the highest number of days to first flowering in the control. This value was significantly different from other treatments. On the other hand, treatment C had lowest number of days to first flowering (66.00).

Also, the control E for SE accession had the highest mean number of days to first flowering (80.20) which is significantly different from other treatments. Treatment A had the lowest number of days to first flowering (67.80), this value was significantly different from other treatments apart from C. The control (treatment E) in all the three accessions had the highest number of days to first flowering as compared to other treatments.

Effects of Colchicine on the Fruit Weight of Tomato Accessions

The fruit weight recorded in the SW accession, for all the treatments were higher than the control (E), with treatment C having a significantly higher fruit weight (9.89 g). The lowest fruit weight (6.04 g) was recorded in the control which is significantly different from other treatments except treatment D (Table 2). In NE Accession, treatment C had the highest fruit weight (7.36 g), which is significantly different from other treatments, except A. The lowest fruit weight at treatment B was recorded as 5.70 g, this value was significantly different from other treatments except E. There were significant differences between the fruit weight for all the treatments in SE Accession with treatment C having the highest fruit weight (9.70 g) and the lowest (4.54 g) recorded in treatment E.

Effects of Colchicine on the Number of Fruits per plant of Tomato Accessions

The result of the effect of Colchicine on the number of fruits per plant as shown in Table 2, revealed that in the SW Accession, treatment B had the highest number of fruit per plant (7.53), this value was significantly different from other treatments except for treatment A (6.80). A significant low number of fruit per plant (3.07) was recorded in treatment D. In the NE accession, treatment A had a significantly higher number of fruit per plant (8.60) and the least (3.00) was recorded in treatment D, which is significantly different from other treatments. In SE Accession, a significantly higher number of fruits per plant were observed in treatment C (4.60). On the other hand, a significant low number of fruit per plant (2.60) was recorded in treatment A.

DISCUSSION

The result of the effects of the doses of Colchicine on the germination percentages of tomato accessions indicated an independent response to the mutagen. However, the result of the southwest and southeast accession shows that the control produced the highest germination percentages of 60% and 73.33% respectively and this was in agreement with the findings of Mensah and Obadoni (2007), who reported the reduction in germination percentage with increase in doses of Colchicine on sesame seeds. The delay in seedling emergence could be attributed chiefly to physiological disturbance, which might have probably affected some biological pathway in the treated seeds. This might have delayed seedling emergency and finally reduced the germination percentage. It is probable that the seeds soaked in higher concentration of Colchicine have to readjust in response to the treatment thus delaying the time it took the seed to emerge (Brisibe et al., 2011). On the other hand, the result of the Northeast accession showed high significant difference in germination percentage with increase in Colchicine treatment. This was in agreement with the work of Adelanwa and Habeeb (2011), who reported that tomato variety Roma VF yielded highly significant difference on germination percentage when treated with Colchicine. The result of the effects of different concentrations of Colchicine on the number of branches of tomato accessions shows that treatment C produced a significantly higher number of branches (1.60) in all the three accessions. However, this result is in contrast with the findings of Adelanwa and Habeeb (2011), who reported 7.2 number of branches in treatment D of colchicine, compared to the control (7.60). The increase in the morphological parameters of plants may be attributed to the artificial induction of mutation by Colchicine leading to the alteration of plant genome integrated by environmental signals, perhaps by increasing the rates of cellular division and expansion at their meristematic regions (Uno et al., 2001).

The emergence of early flowering mutants in this study indicates that treatment A and C were effective and is in agreement with the work of Nura et al. (2013), who reported early flowering Colchicine-induced mutants in Sesame (*Sesamum indicum* L.). Similarly, Ravindra et al. (2019) reported a decrease in the number of days for flower bud emergence due to the effect of the mutagen in *Physalisperuviana* L. (Cape gooseberry).

The increase in the plant height of treated tomato seeds with increasing concentration (0.5 mM and 1.0 mM) of Colchicine observed in this study is in agreement with the findings of Adelanwa and Habeeb (2011), who observed significant high plant height at treatment D concentration in tomato (*Lycopersicon esculentum*). However, Nura et al. (2013) observed significant higher plant height at low concentration (treatment A) in Colchicine-induced mutants in Sesame (*Sesamum indicum* L.)**.** Therefore, appropriate dose of Colchicine can significantly increase the height of plants. The mutagen might have probably influenced the activities of cytokinin which is of great importance in the fundamental processes of plant development including cell division and morphogenesis (Deikman and Ulrich, 1995).

A significant increase in the leaf length (12.10 cm and 10.84 cm), as observed in SW and NE accessions, respectively, was at treatment A. This result is however, in agreement with the findings of Ravindra et al. (2019) who observed a significant higher leaf length of Cape gooseberry (12.13cm) at 0.1% Colchicine treatment.

A pronounced variation was observed in the number of leaves of induced tomato seeds at the different concentrations of Colchicine in the South West accession. This result reveals that Colchicine has the ability to increases the number of leaves in tomato plant as well as crops that have vegetative values. A similar result was reported by Nura et al. (2013). They observed pronounced variations in the number of leaves of two varieties of Colchicine-induced Sesame, with the highest number of leaves recorded at treatment 0.1mM. Abiola et al. (2014) also observed a significant difference in the number of leaves of plants from treated seeds (11.08-4.98). This result is however contrary to the findings of Adelanwa and Habeeb (2011) who observed no variations in the number of leaves of colchicine-induced tomato types tested. Furthermore, Nura et al. (2011) reported increase in leaf number and area among mutants of jute. As reported by Lockhart et al. (1996), the increase in leaf number and area provides an increase in the surface area for gaseous exchange which considerably affects the process of photosynthesis.

The highest fruit weight for the three varieties was observed in 0.5 mM while the least was observed in the control 0.0 mM of Colchicine. This result therefore shows that Colchicine's ability to double the chromosomes, enable an increase in the fruit weight of tomato plants.

This result is similar to the findings of Mensah et al. (2013) who observed significant high weight in *Sesame indicum* L. treated with colchicine.

Also, there were variations in the number of fruit per plant in the induced tomato at the different concentrations of Colchicine, with increase in the number of fruit per plant in the treated accessions. Similar to this result, Tammu et al. (2021) observed a significant increase in the number of fruit per plant of C*apsicum annuum* L. in Colchicine treated plants. As reported, there was an average number of fruits of about two to three times higher than control plants.

CONCLUSION

The effect of colchicine on the agro-morphological and yield parameters of tomato investigated in this study such as number of days to first flowering, leaf size, fruit weight and number of fruit/plant, indicates beneficial effect as there was increase in the parameters compared to the control. The study shows that it is shown that the gigas effect of Colchicine can be achieved in grain crops as well as vegetable crops to enhance high yield production and crop improvement in plant breeding.

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APPENDICES

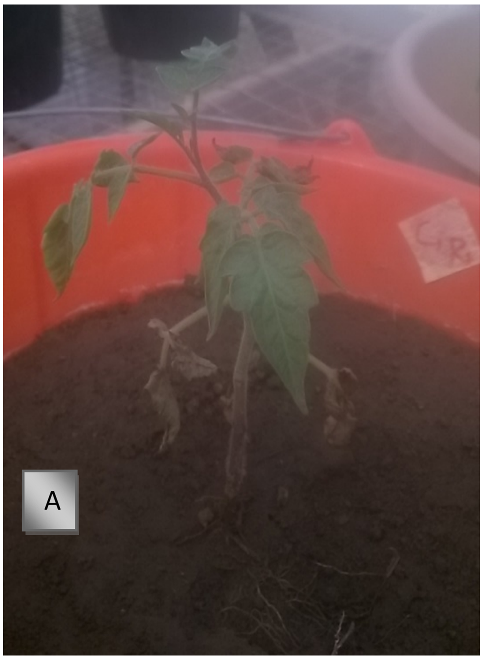

Figure 1A: Control Tomato plant
Treated

Figure 1B: Plant height of Colchicine
Tomato at 8 Weeks after Planting.

Source: Field Photographs

Table 1: Effects of Colchicine on the Morphological Parameters of Tomato accessions

Accession/ Concentration (mM)	Germination %	Number of Branches	Plant height @ week 8 (cm)	Leaf Length (cm)	Number of Leaves
Accessions SW					
0(E)	60.00 ± 0.00^{b}	0.80 ±0.37^{a}	15.24± 4.80^{a}	8.08 ± 1.63^{a}	8.20 ± 1.16^{a}
0.1(A)	60.00 ± 11.55^{b}	1.20±0.20^{ab}	26.74± 1.39^{d}	12.10 ± 0.52^{c}	11.20± 0.66^{d}
0.3(B)	53.33 ± 6.67^{a}	1.20±0.37^{ab}	24.46± 7.66^{c}	9.10 ± 1.47^{ab}	9.20 ± 1.96^{b}
0.5(C)	60.00 ± 11.55^{b}	1.60±0.24^{b}	30.70± 5.62^{e}	11.60± 1.26^{b}	12.60 ± 2.14^{e}
1.0(D)	60.00 ± 11.55^{b}	1.20± 0.20^{ab}	18.86± 4.99^{b}	8.58 ± 1.11^{a}	10.80 ± 2.11^{c}
Accessions NE					
0(E)	73.33 ± 6.67^{ab}	1.00 ±0.45^{a}	34.64± 5.97^{d}	8.74 ± 1.13^{b}	12.80 ± 2.22^{c}
0.1(A)	93.33 ± 6.67^{c}	1.20±0.37^{ab}	34.48± 4.78^{d}	10.84± 1.57^{d}	15.00 ± 2.98^{e}
0.3(B)	86.67 ± 6.67^{b}	1.00±0.32^{a}	29.38± 4.95^{c}	10.20± 0.90^{d}	11.80 ± 2.06^{b}
0.5(C)	93.33 ± 6.67^{c}	1.60±0.24^{b}	15.68± 4.67^{a}	5.48 ± 0.82^{a}	9.80 ± 1.02^{a}
1.0(D)	60.00 ± 20.00^{a}	1.60± 0.24^{b}	24.76± 7.23^{b}	9.06 ±1.24^{c}	13.60 ± 1.96^{d}
Accession SE					
0(E)	73.33 ± 6.67^{c}	0.80 ±0.20^{a}	16.74± 6.51^{b}	9.96 ± 1.92^{c}	11.00 ±1.38^{d}
0.1(A)	46.67 ± 13.33^{a}	1.00±0.32^{ab}	11.28± 2.86^{a}	6.88 ± 0.86^{a}	8.20 ± 0.58^{a}
0.3(B)	53.33 ± 6.67^{ab}	1.20±0.37^{ab}	17.90± 5.79^{c}	7.78 ± 2.03^{ab}	9.20 ±1.66^{b}
0.5(C)	66.67 ± 17.64^{b}	1.60± 0.40^{b}	19.42± 7.29^{d}	8.76 ± 2.06^{b}	10.00 ±1.76^{c}
1.0(D)	53.33 ± 6.67^{ab}	1.40± 0.24^{b}	30.10± 6.03^{e}	11.20± 1.20^{d}	11.40 ± 0.75^{d}

Values followed by the different superscript down in a column are significantly different $p<0.05$

Table 2: Effects of Colchicine on the Yield Parameters of Tomato Accessions

Accession Concentration (mM)	Number of Days to First Flowering	Fruit weight (g)	Number of Fruit per Plant
Accession SW			
0 (E)	$79.40 \pm 1.29^{d*}$	$6.04 \pm 0\ .47^{a}$	4.67 ± 0.73^{ab}
0.1(A)	66.60 ± 1.25^{a}	7.13 ± 0.45^{ab}	6.80 ± 1.10^{b}
0.3(B)	68.80 ± 3.01^{b}	7.17 ± 0.96^{ab}	7.53 ± 1.48^{b}
0.5(C)	65.00 ± 1.67^{a}	9.89 ± 0.72^{b}	4.80 ± 0.66^{ab}
1.0(D)	75.60 ± 2.73^{c}	6.91 ± 0.79^{a}	3.07 ± 0.56^{a}
Accession NE			
0 (E)	79.20 ± 1.71^{d}	6.14 ± 0.94^{a}	4.60 ± 1.40^{b}
0.1(A)	67.40 ± 1.91^{ab}	7.32 ± 0.36^{b}	8.60 ± 2.06^{d}
0.3(B)	71.20 ± 2.96^{b}	5.70 ± 1.19^{a}	7.00 ± 1.95^{c}
0.5(C)	66.00 ± 1.52^{a}	7.36 ± 1.25^{b}	4.20 ± 1.02^{b}
1.0(D)	73.60 ± 2.98^{c}	6.68 ± 1.46^{ab}	3.00 ± 1.05^{a}
Accession SE			
0(E)	80.20 ± 1.32^{d}	4.54 ± 0.54^{a}	3.40 ± 1.0^{ab}
0.1(A)	67.80 ± 1.66^{a}	5.580 ± 0.87^{ab}	2.60 ± 0.51^{a}
0.3(B)	70.80 ± 3.12^{b}	8.60 ± 2.30^{c}	3.20 ± 0.86^{ab}
0.5(C)	68.60 ± 3.17^{a}	$9.70 \pm .63^{d}$	4.60 ± 1.17^{b}
1.0(D)	76.20 ± 3.18^{c}	6.50 ± 1.89^{b}	3.20 ± 1.20^{ab}

*Values followed by the different superscript down in a column are significantly different $p<0.05$