MUTAGENIC EFFECTS OF SODIUM AZIDE (NaN₃) ON MORPHOLOGICAL CHARACTERISTICS ON TWO VARIETIES OF TOMATO (Solanum lycopersicum Mill)

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ABSTRACT
The impact of Sodium Azide on the germination and growth of two varieties of Tomato was investigated with the aim of inducing variability that could be exploited in the improvement of some quality traits in tomato plants. The seeds of two varieties of Tomato (Roma and UC82B) were treated with three different concentrations of Sodium Azide (0.1 mM, 1.0 mM, 2.0 mM and 0.0 mM as control). The result obtained revealed a highly significant differences (P≤0.01) in the effects of Sodium Azide on seedlings heights, height at maturity, fruit weight, pericarp thickness, number of leaves and pH (Potential of Hydrogen) Significant improvement (P≤0.05) was also recorded in fruit diameter, while no significance difference was found on root dry weight. The result revealed that Sodium Azide improve important quality traits in tomato. However, the response of variety UC82B to Sodium Azide was higher. More so, significant differences were observed between the seasons except in the number of leaves, fruit number, pericarp thickness and fruit diameter. It was concluded that 0.1 mM of Sodium Azide improve some quality traits of tomato that could be utilized in the improvement of tomato.

Key Words: Sodium Azide, Tomato, Mutation, Varieties, Morphology, UC82B, Roma.

INTRODUCTION
Mutations are the tools used by the geneticist to study the nature and function of genes which are the building blocks and basis for plant growth and development, thereby producing raw materials for genetic improvement of economic crops (Adamu et al., 2004). Induced mutations have great potentials and served as a complimentary approach in genetic improvement of crops (Mehandjiev et al. 2001). Various mutagenic agents are used to induce favorable mutations at high frequency which include ionizing radiation and chemical mutagens (Ahloowalia and Maluszynski, 2001). Induced mutations have been used to improve major crops such as wheat, rice, barley, cotton, peanut and cowpea, which are seed propagated (Ricardo and Ando 1998) have reported on the role of chemical mutagens in enhancing variability in higher plants. The mutants produced facilitate the isolation, identification and cloning of genes used in designing crops with improved yields, increase stress tolerance, longer shelf life and reduced agronomic input (Ahloowalia and Maluszynski 2001).

The cultivated tomato is herb with different growth habits comprising of erect, prostrate or climbing stems (Gill, 1992). The stem can be determinate or indeterminate with reddish to yellowish glandular hairs having monopodial branching system. The root is of tap-root system. The leaves are opposite and pinnately compound with lobed margins which are ovate to oblong and petiolated. The flowers form raceme inflorescence, perfect, actinomorphic and pentamerous with superior ovary. The fruit is a berry with smooth skin which is reddish to yellowish succulent at maturity and many seeded. The seeds are flat and kidney shaped (Johnson, 2005).

Tomato (S. lycopersicum Mill.) is a plant with variety of uses. The importance of the plant is mostly centered upon its edible fruit s (Mann et al., 2003). The fruit is rich in Alkaloid Tomatine, Lycopene, Carotene, Ascorbic acid and Vitamins (Gill, 1992). The seeds, leaves and skin can provide oil on extraction that can be used as cooking oil. The fruit is used for making soup and sausage (Johnson, 2005). Commercially, the sale of the fruits served as a source of income to peasant farmers and as a source of income to the governments. The ‘kwanar Gafan’ tomato market in Kano state serve as the centre for purchasing and supply of tomato to the nation; providing job opportunities to thousands of individuals in the area. The desirable characters usually sought in a tomato fruit include total soluble solids (TSS), pH, fruit size, fruit shape, and color of the fruits. Some other characters such as fruit firmness, thick pericarp, optimum pH and high TSS are important characteristics from a processing point of view (Dhaliwal et al., 2002).
The plant is of high medicinal importance as the fruit is a good remedy for preventing or inhibiting various forms of venous and arterial thrombosis and fibrin clots formation in veins. It is a valuable food mineral and vitamins particularly vitamin A and C. Studies have shown that people who ate tomatoes or tomato products may be at lower risks of some kind of cancer, particularly cancer of the prostate gland, lungs and stomach (Anonymous, 2009). The fruits are recommended for patients suffering from obesity, hormone replacement therapy, anti phospho-lipid syndrome, cancer or patients suffering from genetic and plasmic risk factors as well as those that had recently undergone surgical operations (Anonymous, 2010).

Research on the genetic control of fruit quality traits have been dominated by studies of the ripening process, since this is a specific developmental process in fruit greatly affects the evolution of several quality parameters, like colour, aroma, Soluble Solute Contents (SSC) and Titratable Acidity (TA). High SSC and TA are highly desirable not only in processing cultivars but also in fresh-market cultivars due to the important contribution of sugars and acids to the overall flavour and nutritional value of tomatoes (Foolad, 2007). Mutations are the tools used by the geneticist to study the nature and function of genes which are the building blocks and basis of plant growth and development, thereby producing raw materials for genetic improvement of economic crops (Adamu et al., 2004).

Induced mutations have great potentials and served as a complimentary approach in genetic improvement of crops (Mehandjiev et al., 2001). Various mutagenic agents are used to induce favorable mutations at high frequency that include ionizing radiation and chemical mutagens (Ahloowalia and Maluszynski 2001). The mutant produced facilitate the isolation, identification and cloning of genes used in designing crops with improved yields, increase stress Tolerance, longer shelf life and reduced agronomic input (Ahloowalia and Maluszynski 2001).

Tomato suffers from several problems that include high disease incidence, pest infestations, and adverse effects of environmental stress e. t. c. that greatly affects its production (Encarta 2005). The results of a study on the effectiveness of Sodium Azide in inducing mutation in tomato are presented. This study aimed to investigate the mutagenic effects of Sodium Azide (NaN₃) on Morphological Characteristics of Two Varieties of Tomato (Solanum lycopersicum Mill).

MATERIALS AND METHODS
The research was conducted in (September, 2013) at the Green House of the Botanical Garden of the Department of Biological Sciences, Ahmadu Bello University Zaria, Kaduna State. (Lat11⁰ 12'N, Long 7⁰,37'E, Alt 550-700 m above sea level) (Anonymous, 2014).

Sources of the Seeds
Seeds of three varieties of cultivated tomato (Roma, UC82B) were collected from the Institute for Agricultural Research (I.A.R), Ahmadu Bello University Zaria, Nigeria. The UC82B was said to flourished more during dry season, while the Roma flourished more during rainy season.

Treatment and Experimental Design
The treatment used in the study is Sodium Azide and two varieties of tomatoes (Roma and UC82B) with three different concentrations of sodium azide (0.1mM, 1.0mM, 2.0Mm). These were laid out in a Completely Randomized Design (CRD) with three replications. The seeds of the three tomato varieties were soaked/treated with three different concentrations of Sodium Azide (0.1mM, 1.0mM, 2.0Mm) while 0.0mM as control.

Data Collection
Data were obtained for seedling height, height at maturity, diameter of the fruits, thickness of pericarp, number of leaves, fruit weight, pH(Potential of Hydrogen) value and Root dry weight.

Data Analysis
All the data obtained were analyzed using Analysis of Variance. And the means were separated using Duncan’s Multiple Range Test. (DMRT).

Seedlings Height (cm)
The heights of the plants per treatment per variety were determined after eight weeks of planting by holding the highest leaf erect and the heights measured in centimeters

Height at Maturity (cm)
The heights of the successful grafts were determined after the emergence of the first flower by holding the highest leaf erect and the heights measured in centimeters

Number of Leaves/Plant
The number of leaves produced per plant was counted and recorded for each treatment after the emergence of the first flower.

Thickness of Pericarp (mm)
The pericarps of the fruit of the different treatments were measured using screw gauge in millimeters and recorded.
Diameter of the Fruits (cm)
The diameter of the fruits per treatment was determined by screw gauge and recorded. pH-values of the Fruit Juice
The pH values of the fruit juices were determined using pH-meter and recorded. 6 fruits were randomly harvested from each replication and were used for pH determination.

Roots Dry Weights (g)
The weights of the roots per treatment were taken in grams using a balance machine and recorded.

RESULTS
The results from the combined analysis of variance on the effects of mutation on some selected traits of three tomato varieties are presented in (Table 1). The results showed highly significant difference (P≤0.01) in the effect of concentrations of sodium azide on almost all the selected tomato traits except on fruit diameter; where the effect is significant (P≤0.05) and on root dry weights; no significant differences was found. Similarly, highly significant difference (P≤0.01) was found among the varieties in terms of the selected traits only in height at maturity and fruit diameter, where significant difference exist (P≤0.05). But no significant difference was found among the varieties in pericarp thickness. More so, highly significant difference (P≤0.01) was found among the treatments (mutation) in terms of seedlings height, fruit weight, and number of leaves, however, no significance difference was observed on pH(Potential of Hydrogen), root dry weight, height at maturity, pericarp thickness and fruit diameter.

Table 1: Mean Squares for the Effects of Sodium Azide on Two Varieties of Tomato

| Sources of Variation | Df | Seedlings Height (cm) | Height at Maturity (cm) | Number of Leaves | Fruit Weight (g) | Pericarp Thickness (mm) | Fruit Diameter (cm) | pH | Root DW (g) |
|----------------------|----|-----------------------|------------------------|------------------|-----------------|------------------------|---------------------|----|------------|
| Replication          | 2  | 269.79**              | 88.41**                | 27.83**          | 55.92**         | 0.01**                 | 0.12**              | 0.01** | 3.31**     |
| Concentration        | 2  | 1459.65**             | 3716**                 | 300.33**         | 383**           | 0.53**                 | 0.28*               | 0.92** | 3.42**     |
| Variety              | 1  | 599.37**              | 270.39*                | 226.86**         | 564.34**        | 0.07**                 | 0.20*               | 0.42** | 7.38**     |
| Error                | 142| 17.17                 | 56.22                  | 3.21             | 7.53            | 0.05                   | 0.06                | 0.01   | 1.11       |

Keys: ns= No significant difference  * = Significant difference (P≤0.05) **= Highly significant difference (P≤0.01)

The result of the responses of the two varieties of tomato to sodium Azide is presented in (Table 2). The result showed that, variety UC was found to be better in almost all the selected traits, except on thickness of pericarp where the performance was low. However, variety Roma was found to be better than of the UC in pericarp thickness (0.28 mm) that produced 55-64 seeds per fruit having fruit diameter of 0.22-0.31 cm.

The variety UC also produced the highest number of fruits which are larger in size (0.31 cm in diameter), 16.11g weight, containing 64 seeds/fruit. But in terms of pericarp thickness, the response of the variety Roma is the best.

Table 2: Responses of the Two Tomato Varieties to Sodium azide

| Variety | Seedlings Height (cm) | Height at Maturity (cm) | Number of Leaves | Fruit Weight (g) | Pericarp Thickness (mm) | Fruit Diameter (cm) | pH | Root DW (g) |
|---------|-----------------------|------------------------|------------------|-----------------|------------------------|---------------------|----|------------|
| Control | 19.37d                | 31.27d                 | 11.64d           | 9.93b           | 0.21c                  | 0.17c               | 4.07d | 10.03g     |
| UC      | 27.72a                | 43.13d                 | 16.58b           | 16.11e          | 0.26e                  | 0.21e               | 4.29a  | 2.50b      |
| Roma    | 26.05b                | 40.38b                 | 13.56b           | 10.79c          | 0.28d                  | 0.22d               | 4.21b  | 2.11b      |
| Mean    | 25.29                 | 40.97                  | 14.55            | 12.94           | 0.27                   | 0.25                | 4.21   | 2.17       |

N.B: *a* Means within the columns with the same letter(s) are not significantly different (P≤0.05) using (DMRT)
DISCUSSION
The differences observed in most of the quantitative and qualitative traits among the mutants showed significant improvements in the selected traits. Although there were few traits with no significant differences in responses to the applied treatments; the ability of the mutants to germinate faster after one and two weeks of planting when compared with the controls showed that the mutagenic treatments induced increased enzymatic activities, which could be responsible for the early germination. This finding is in agreement with the findings of Mensah et al. (2007) who reported a decrease in germination with increase in the dose of chemical mutagens. In the present investigation, plant heights and leaf number decreased with increasing concentration of sodium azide. This finding conformed to the earlier report by Ahloowalia and Maluszynski (2001) that, the viable mutants observed are mainly dependable measure of genetic effect in mutagen. The increased in the number of leaves and plant heights due to sodium azide treatments was also in conformity with the work of Adamu and Aliyu (2007) who reported increased in growth and yield parameters of tomato due to sodium azide treatments.

More so, the improvement in the growth and yield components of tomato due to sodium azide treatments demonstrate the effect of mutation on the growth and yield of plants. This is in agreement with the work of Adamu et al. (2002) who reported the effect of gamma rays on Groundnut. Sheeba et al. (2005) who reported the effect of gamma rays and EMS on Sesanum indicum L. on seedling survival, was reduced significantly with an increase in dosage levels of both mutagens. However, in contrast, Sasiet et al. (2005) showed that all plant mutant types registered lower yields compared to their parents in the study of the effects of diethylsulphate and EMS on Okra (Abelmoschus esculentum (L.) var. MDU-1).

The increased in fruit quality (such as pericarp thickness, juice pH and fruit weight) and fruit number due to induced mutagenesis by Sodium Azide signifies the vital role played by the mutagen in improving the quality traits of tomato.

CONCLUSION
The effect of Sodium Azide was found to be beneficial in improving certain qualitative traits of tomato varieties. Variety UC is therefore recommended for processing industries. More so, Induced mutation using various concentrations of sodium azide technique were employed singly and in combination on the two varieties of tomato with the aim of improving the growth and yield parameters of the plants in both the wet and dry seasons.

It was concluded that, Sodium Azide via mutation improves some important quality traits of tomato that are of high economic value and possible recommendations made. It was also concluded that, 0.1mM concentration of sodium azide (NaN₃) is significant in inducing variability that could be exploited in the improvement of highly economic crops like tomato. It was also concluded that, the mutants of tomato can be grown all the year round (both during the rainy and dry seasons).

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