Nutrient, Mineral and Lycopene Content of Tomato (Solanum Lycopersicon L.) Grown with Different Extracts of Neem (Azadirachta Indica L.) and Sunflower (Tithonia Diversifolia Hemsl.)

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Abstract: - The effect of extracts of neem (Azadirachta indica) and sunflower (Tithonia diversifolia) extracted differently with 30% ethanol+70% water and 30% methanol+70% water on mineral, vitamin and lycopene content of tomato fruit was studied. The study was carried out at the screenhouse of faculty of Agriculture, Obafemi Awolowo University, Ile-Ife in south west Nigeria. The study consisted of five treatments which are tomato plant alone (control), neem leaves extracted with (30% ethanol+70% water) + tomato plant, neem leaves extracted with (30% methanol + 70% water) + tomato plant, sunflower extracted with (30% ethanol + 70% water) + tomato plant and sunflower leaves extracted with (30% methanol + 70% water) + tomato. Each of the treatments was in triplicate and arranged in randomized complete block design. Harvesting was done between 9 and 14 weeks after planting. The fruits from each treatment were analyzed in the laboratory using appropriate methods to get the value for vitamin A, B1, B2, calcium, sodium, zinc and lycopene. The result shows that vitamin B1, calcium and sodium was significantly higher in treatment with Sunflower (30% methanol + 70% water) + tomato plant which had a mean value of 0.48 mg/100g, 10.26 mg/100g and 0.018 % for vitamin B1, calcium and sodium respectively. Control treatment had the highest value for vitamin A mean value of 2953.86 µg/100g which was significantly higher than treatments with extracts of sunflower leaves (30% ethanol + 70% water) + tomato plant and sunflower (30% methanol + 70% water) + tomato plant with mean value of 2819.22 μg/100g and 2833.74 μg/100g respectively. Treatment with sunflower leaves extract (30% methanol + 70% water) + tomato plant which had a significantly high value for vitamin B1, calcium and sodium also had a significantly high value for zinc with mean of 3.84 mg/kg while lycopene had value that was not significantly different from one another across the treatments. Vitamin B2 was significantly higher in the control treatment with mean value of 0.21mg/100g.

Keywords: tomato; lycopene; nutrient; mineral; neem extract and sunflower extract

I. INTRODUCTION

Tomato belongs to the family solanaceae (night shade) and it one of the most important food crops. Tomato is a

native of Andes in Western South America. Wild tomato can grow in wide varieties of habitats, from near sea level to 3300 m in elevation (Rick, 1973; Taylor, 1986, Peralta et al., 2001). It had become a major condiment in many dishes across the globe. The wide acceptance cannot be too far from its rich taste, and as well as its rich nutritional content as it's a supplier of vitamins and other nutrients that human body needs to function properly as well as it rich lycopene content (Canene-Adams, 2005). Lycopene is an antioxidant that is capable of protecting humans from many types of cancer. Adequate consumption of tomato can also reduce the risk of osteoporosis and cardiovascular disease. However, lycopene production in tomato is relatively low when compared to chemical synthesis alternatives (Levin et al., 2004). In order to study the ability of plant extracts on nutritional richness of tomato, botanical extracts from neem and sunflower were used to determine the effect on Vitamin B1, B2, A, Calcium, Sodium, Zinc and Lycopene. Extract from neem and sunflower have the ability to improve growth and yield in tomato (Salami et al., 2017; Salami et al., 2018).

II. MATERIALS AND METHODS

2.1 Tomato Production

Sterilized soil was prepared and filled into pots at the screenhouse of Faculty of Agriculture, Obafemi Awolowo University, Ile-Ife. UC-82 tomato seeds were purchased from the National Horticultural Research Institute, Ibadan, Nigeria. Seeds were raised in the nursery for a period of three weeks before it was transplanted to already prepared pot containing 5kg of sterilized soil. Extracts of neem and sunflower leaves were made by collecting the two botanicals separately at their site of abundance within the campus of Obafemi Awolowo University. The leaves were air-dried separately for four days and grinded to powder with the aid of electric blender. The powdered neem and sunflower were extracted differently with 30% ethanol and 70% water, 30% methanol and 70% water by soaking one gram of the powdered botanicals in them for 24

hours and filtered with whatman filter paper. Each extracts of 100 mills was added to the centre of the pot before transplanting of tomato. Each treatment was in triplicate and arranged in randomized complete block design.

- 2.2 Treatment layout
- B1- Tomato plant alone
- B2- Neem (30% ethanol+70% water) + tomato plant
- B3- Neem (30% methanol + 70% water) + tomato plant
- B4- Sunflower (30% ethanol + 70% water) + tomato plant
- B5- Sunflower (30% methanol + 70% water) + tomato plant
- 2.3 Collection of Fruits and Nutrient Content Analysis

Collection of tomato fruits begins from week 3 after planting all through to week 16.

For the analysis of vitamin C in the samples, determination was done on the same day of harvesting because of the instability of vitamin C. Tomato fruits to be used in the determination of calcium was finely chopped and vacuum oven dried at 70°C until constant weights were achieved. The dried sample was ground to fine powder and stored in airtight bottles in readiness for mineral analysis with respect to calcium. All the laboratory analysis was conducted at the land and water resources laboratory of Institute of Agricultural Research and Training, Obafemi Awolowo University, Moore Plantation, Ibadan, Nigeria.

2.4 Vitamin C determination

Vitamin C was determined by using the procedure as outlined by Nielsen (1998) Food Analysis Laboratory Manual Chapter 7 Vitamin C Determination by Indophenol Method and AOAC International Methods of Analysis vol 16 Method 967.21. 10 g of each of the samples with the exception of Valencia oranges was accurately weighed and ground using mortar and pestle with an additional of 20 ml of metaphosphoric acid acetic acid. The mixture was further ground and strained through muslin and the extract was made up to 100 ml with the metaphosphoric-acetic acid mixture. 5 ml of the metaphosphoric acid-acetic acid solution was pipetted into three of the 50 ml Erlenmeyer flask followed by 2 ml of the samples extract. The samples were titrated separately with the indophenol dye solution until a light rose pink persisted for 5 s. The amount of dye used in the titration were determined and used in the calculation of vitamin C content.

2.5 Determination of Vitamin B1

About 1.3g of the sample was weighed into a 150ml flask, 5ml flask of 5N HCI was added with 5mls of dichloro ethene. De-ionized water of 90ml of was also added. It was then put on the steam bath for 30 minutes so that all the Riboflavin in the sample could get dissolved into the solution. It was then allowed to cool and made up with water. The solution was then filtered and the first 20ml of the aliquot was discarded.

About 2ml of it was then pipetted into a 200ml volumetric flask and water was added to mark. Standard solution was prepared by dissolving 50gm of riboflavin in 500mls of distilled water and further dilution of mls into 150mols and 5ml into 100mols followed. Readings were taken on the flourometer and the calculation followed:

Sample reading X Standard weight X Dilution

Standard Reading Sample weight

= mg of riboflavin/gm

2.6 Determination of Vitamin B1 (Thiamine)

Vitamin B1 was determined by weighing 1g of first 20ml of Solanum lycopesicon sample into 100ml volumetric flask; 25ml of 0.1M H₂SO₄ was added and mixed by careful swirling. Additional 25ml of 0.1M H₂SO₄ was added to rinse any adhering sample particle off the flask. The flask was set in a boiling water bath to ensure a complete dissolution of the sample in the acid. The flask was shaken frequently in the first 5 minutes and subsequently every 5 minutes for 3 minutes. About 5ml of taka-diastase in 0.5M Sodium acetate solution was added and flask set in cold water to cool content below 50°C. The flasks was stopped and kept at a temperature between 45° C-50°C for 2 hours and thereafter made up to 100ml in ark after mixing thoroughly. The mixture was filtered through a No 42 Whatman filter paper, discarding the first 10ml and keeping the remaining. 10ml of the remaining filtrate was pipetted into a 50ml volumetric flask and 5ml of acid potassium chloride solution was added, shaking thoroughly to mix well. Standard Thiamine solution of range 10mg/ml was prepared from 100mg/ml stock and treated same way prepared from sample above. The absorbance of the sample as well as that of standards was read on a fluorescent UV spectrometer (Cecil A20model) at a wavelength of 285nm. Vitamin B¹ in mg/100g was calculated using the

 $Vitamin\ B1\ in\ mg/100g =$

Absorbance x Ave. Gradient x Dilution Factor

Wt. of sample

2.7 Determination of Vitamin B2

About 1.3g of the sample was weighed into a 150ml flask, 5ml flask of 5N HCI was added with 5mls of dichloro ethene. De-ionized water of 90ml of was also added. It was then put on the steam bath for 30 minutes so that all the Riboflavin in the sample could get dissolved into the solution. It was then allowed to cool and made up with water. The solution was then filtered and the first 20ml of the aliquot was discarded. About 2ml of it was then pipetted into a 200ml volumetric flask and water was added to mark. Standard solution was prepared by dissolving 50gm of riboflavin in 500mls of distilled water and further dilution of mls into 150mols and 5ml into 100mols followed. Readings were taken on the flourometer and the calculation followed:

Sample reading X Standard weight X Dilution

Standard Reading Sample weight

= mg of riboflavin/gm

2.8 Determination of Vitamin A

Vitamin A was determined by weighing 2g of sample into a flat bottom reflux flask. 10ml of distilled water was added. shaked carefully to form a paste. 25ml of alcoholic HOH solution was added and a reflux condenser attached. The above mixture was heated in water bath for 1 hour with frequent shaking. The mixture was cooled rapidly and 30ml of water was added. The hydrolysate obtained was transferred into a separatory funnel. The solution was extracted three times with 250ml quantities of chloroform. About 2g anhydrous Na₂S04 was added to the extract to remove any traces of water. The mixture was then filtered into 100ml volumetric flask and made up to mark with chloroform. Standard solution of B-carotene Vitamin A of range 0-50 μ g/ml with chloroform were made by dissolving 0.003g of standard B-carotene in 100ml of chloroform. Absorbances of sample and standards were read on the Spectrophotometer (Metrohm Spectronic 21D Model) at a wavelength of 328nm.

Vitamin A (μ g/100g) =

Absorbance of sample x Dilution Factor

Weight of Sample

2.9 Determination of Calcium and Sodium

The ash of *Solanum lycopersicom* fruit sample obtained was digested by adding 5ml of 2 MHCL to the ash in the crucible and heat to dryness on a heating mantle. 5ml of 2 MHCL was added again, heat to boil, and filtered through a Whatman No. 1 filter paper into a 100ml volumetric flask. The filtrates was made up to mark with distilled water stopper and made ready for reading of concentration of Calcium, Potassium and Sodium on the Jenway Digital Flame Photometer (PFP7Model) using the filter corresponding to each mineral element.

The concentration of each of the element was calculated using the formula:

%Ca, and %Na =

Meter Reading (MR) x Slope x Dilution factor

1000

$2.10\ Determination\ of\ Lycopene\ Content\ in\ tomato$

Lycopene was determined using the low hexane extraction method (Fish *et al.*, 2002). Approximately 0.6g was weighed into 40 ml of amberscrew-top that contained 5 ml of 0.05% (w/v) butylated hydroxytoluene in acetone, 5 ml of 95% USP grade ethanol, and 10 ml of hexane. The sample was stirred on a magnetic stirring plate during sampling. The sample was then extracted on an orbital shaker at 180 RPM for 15 mins on ice. After thorough shaking, 3 ml of deionized water was

added and then further shaken for another 5 minutes on ice. It was then left for for 5 min at room temperature for phase separation. The absorbance of the upper, hexane layer was measured in a 1 cm path length quartz cuvette at 503 nm blanked with hexane. The lycopene content of the sample was then determined using absorbance at 503 nm and the sample weight (Beerth *et al.*, 1959; Fish *et al.*, 2002).

2.11 Statistical Analysis of Data

The data were subjected to descriptive statistical methods and the means were separated using error bar.

III. RESULTS

The result of vitamin B2 content of the harvested tomato fruit shows that treatment 5 that is 30% methanol + 70% water (Sunflower extract) +Tomato has the highest value for vitamin B2 in all the harvested fruits which is significantly higher than all other treatments with extracts of neem and sunflower with value of 0.48 mg/100g while the lowest value for vitamin B2 was recorded in treatment with 30% ethanol + 70% water (Sunflower extract) + Tomato which had a value of 0.26 mg/100g (Fig. 1). Treatment B1 which was the control treatment consisted a value of 0.43 mg/100g which was significantly higher than treatments 2, 3 and 4 with 30% ethanol + 70% water (Neem extract) + Tomato, 30% methanol + 70% water (Neem extract) + Tomato and 30% ethanol + 70% water (Sunflower extract) + Tomato (Fig. 1).

For vitamin A, the highest level was recorded in the control treatment with value of 2953.86 ug/100g which was significantly higher than the two treatments which contained extracts of sunflower which are 30% ethanol + 70% water (Sunflower extract) + Tomato and 30% methanol + 70% water (Sunflower extract) + Tomato (Fig. 2). The least value for vitamin A was recorded in treatment 5 which consisted of 30% methanol + 70% water (Sunflower extract) +Tomato with mean value of 2833.74 µg/100g (Fig. 2). Vitamin B1 had the highest value in treatment 5 which contained 30% methanol + 70% water (Sunflower extract) + Tomato with mean value of 0.48 mg/100g which was significantly higher than the mean of all other treatments (Fig. 3). Treatment 3 had the lowest mean value of 0.35 but its insignificantly different from treatments 2 and 4 (Fig. 3). Vitamin C was significantly higher in treatment 5 with 30% methanol + 70% water (Sunflower extract) +Tomato (Fig. 8). The mineral content of the fruit revealed that treatment 5 which was the treatment containing 30% methanol + 70% water (Sunflower extract) + Tomato had a significantly higher value for calcium, sodium and zinc (Fig. 4, 5 and 6) with mean of 10.26 mg/100g, 0.018 % and 3.84 g/100g respectively while the least value was in the control treatment with mean of 3.18 mg/100g, 0.014% and 3.05 g/100g respectively for calcium, sodium and zinc.

Lycopene content of the harvested tomato revealed that there was an insignificant difference between the content in treatments 1, 2, 3 and 5. Although, there was a significant

reduction in the lycopene content of treatment 4 compared to the control treatment (Fig. 7).

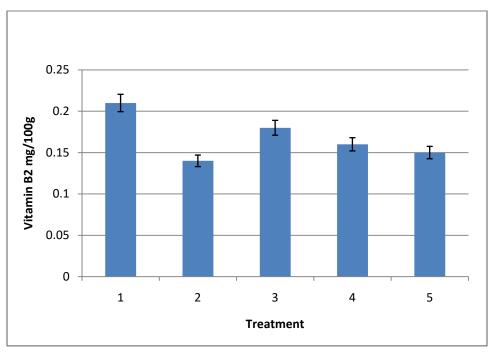


Fig. 1 Vitamin B2 content of harvested tomato fruit

Legend: 1-Tomato alone; 2- 30% ethanol + 70% water (Neem extract) + Tomato
3- 30% methanol + 70% water (Neem extract) + Tomato; 4- 30% ethanol + 70% water (Sunflower extract) + Tomato; 5- 30% methanol + 70% water (Sunflower extract) + Tomato

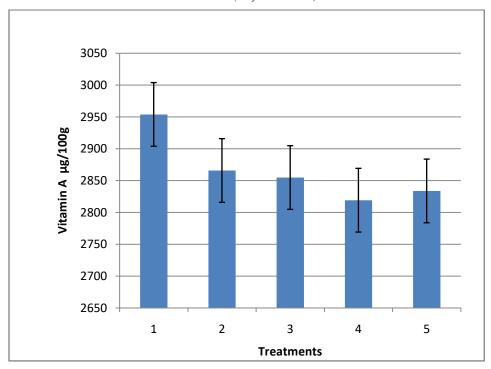


Fig. 2 Vitamin A content of harvested tomato fruit

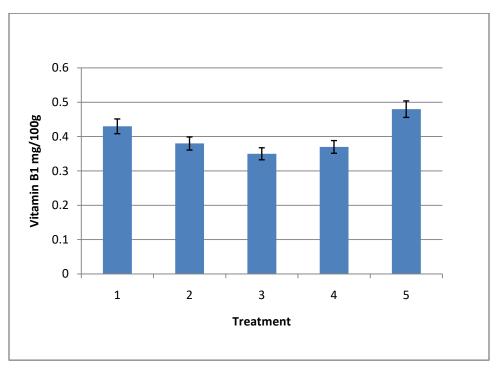


Fig. 3 Vitamin B1 content of harvested tomato fruit

Legend: 1-Tomato alone; 2- 30% ethanol + 70% water (Neem extract) + Tomato

 $3-30\%\ methanol+70\%\ water\ (Neem\ extract)+Tomato;\ 4-30\%\ ethanol+70\%\ water\ (Sunflower\ extract)+Tomato;\ 5-30\%\ methanol+70\%\ water\ (Sunflower\ extract)+Tomato$

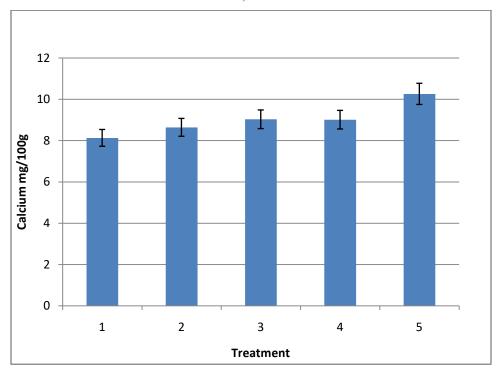


Fig. 4 Calcium content of harvested tomato fruit

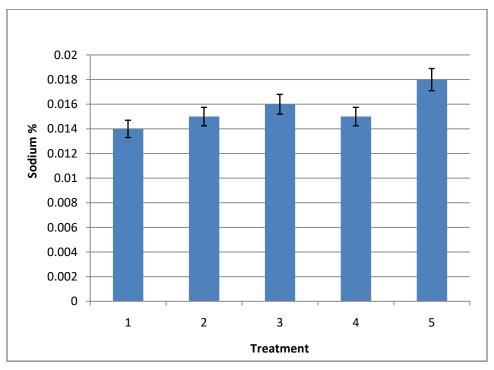


Fig. 5 Sodium content of harvested tomato fruit

Legend: 1-Tomato alone; 2- 30% ethanol + 70% water (Neem extract) + Tomato

3-30% methanol + 70% water (Neem extract) + Tomato; 4-30% ethanol + 70% water (Sunflower extract) + Tomato; 5-30% methanol + 70% water (Sunflower extract) + Tomato

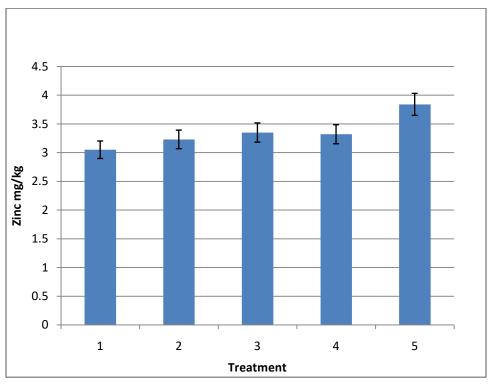


Fig. 6 Zinc content of harvested tomato fruit

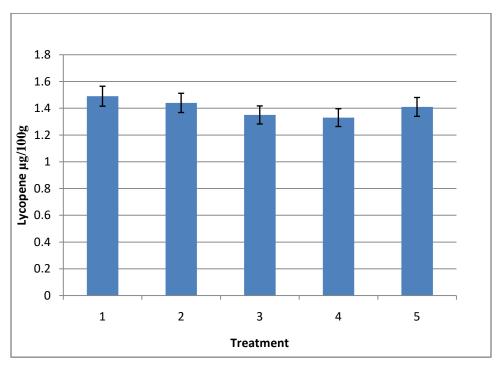


Fig. 7 Lycopene content of harvested tomato fruit

 $Legend: 1\text{-}Tomato\ alone;\ 2\text{-}\ 30\%\ ethanol+70\%\ water\ (Neem\ extract)+Tomato$

 $3-30\%\ methanol+70\%\ water\ (Neem\ extract)+Tomato;\ 4-30\%\ ethanol+70\%\ water\ (Sunflower\ extract)+Tomato;\ 5-30\%\ methanol+70\%\ water\ (Sunflower\ extract)+Tomato$

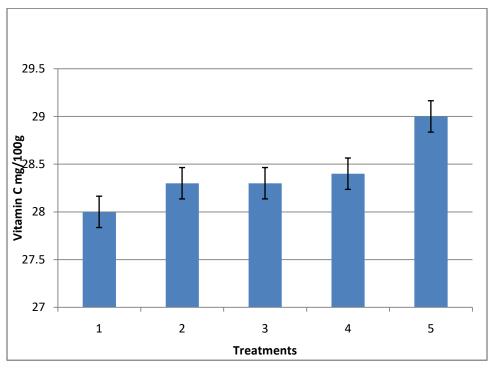


Fig. 8 Vitamin C content of harvested tomato fruit

 $Legend: 1\text{-}Tomato\ alone;\ 2\text{-}\ 30\%\ ethanol+70\%\ water\ (Neem\ extract)+Tomato$

 $3-30\%\ methanol+70\%\ water\ (Neem\ extract)+Tomato;\ 4-30\%\ ethanol+70\%\ water\ (Sunflower\ extract)+Tomato;\ 5-30\%\ methanol+70\%\ water\ (Sunflower\ extract)+Tomato$

IV. DISCUSSION

Sodium content of tomato fruit across the treatment revealed that sodium was higher in all the treatment when compared to the sodium level of the control. Although, the difference was insignificant except in methanolic sunflower extract. Sodium content of the harvested tomato fruit was significantly lower than the standard of 45.8mg/100g (Tindel, 1983). Zinc level was higher than those reported by Gundersen et al., (2001). The zinc concentration was also in agreement with Ordonez-Santos et al., (2011). The zinc level was reportedly higher than the standard of 0.14 mg/100 as the control without any extract had 3.05mg/100g. The zinc level in the fruits was significantly higher than the standard zinc level of 0.14mg/100g (Tindel, 1983). The insignificant difference in the lycopene content between the control and other treatment with extracts agrees with Kapoulas et al., (2011) who stated that no consistence effect of the farming system had been reported to influence the level of lycopene in tomato. However, the lycopene level in all the treatments and the control were lower than standard content of 12mg/100g (Liana et al., 2009). It was also observed that treatment with methanolic sunflower extract had a robust increase in sodium, calcium and Vitamin B1 content which was higher than the control. It was also observed that the Vitamin A content was significantly lower than the standard of 380mg/100g (Tindel, 1983). However, Riboflavin level (Vitamin B2) and Thiamin (Vitamin B1) was significantly higher than the standard content per 100g of fresh tomato fruit which are 0.01mg/100g and 0.06mg/100g respectively in all the treatments including the control (Tindel, 1983).

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