

## **EFFECTS OF NITROGEN NUTRIENT ON THE PHOTOSYNTHETIC PIGMENTS ACCUMULATION AND YIELD OF *SOLANUM LYCOPERSICUM***

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### **ABSTRACT**

*This study investigated photosynthetic pigment accumulation and yield of *Solanum lycopersicum* so as to ascertain the maximum concentration of nitrogen needed for optimum production. Seeds of *S. lycopersicum* tagged with VG-TH-017 were firstly raised in nursery bed. At the end of 28th day after sowing, the seedlings with uniform height were transplanted into experimental pots with 4 seedlings per pot under greenhouse. All the experimental pots were 40 in total, 4 levels of nitrogen ( $KNO_3$  and  $NH_4NO_3$ ) treatment (n, N, 5N, 10N) with 10 replicates. All the plants in the four treatments received 200ml of distilled water at 6a.m. in the morning every day. At 6p.m. in the evening, 100 ml of the differential treatments were applied. The photosynthetic pigments were determined spectrophotometrically. The number of flowers and fruits per plant per pot were counted and recorded. The fruit lengths and fruit diameters in each treatment were determined with the use of a Vernier Caliper. The fruits biomass were also determined. The 10N-plants and 5N-plants had leaves with deep-green colouration indicating an increase in chlorophyll content as well as an increase in the photosynthetic capacity. The highest number of flowers and early flowering discovered in 10N-plants and 5N-plants. The best yield was obtained in the treatments for the 5N-plants in which the concentration of nitrogen in the nutrient solution had been increased to a factor of 5. It is therefore suggested that when the seeds of tomato plants VG-TH-017 are to be grown, the 5N treatment is the most suitable level of application.*

**KEY WORDS:**  $KNO_3$ ,  $NH_4NO_3$ , Pigment, VG-TH-017, spectrophotometer.

### **INTRODUCTION**

Plants take up essential elements in the environment either from the soil through their roots and from the air through their leaves (mainly carbon and oxygen). Plants are often thought of as passive in relation to the environment. However this is not always a valid assumption since many plants clearly manipulate their environment in a fashion that tends to make certain nutrients more readily available. Plants adapt their growth rate and metabolism to the availability of ion or nutrient status present based on differences in growth rate, root distribution, phase of development, and efficiency of nutrient uptake and utilization. They are able to cope and adjust to inadequate supply sometimes, for the short time, without any visual deficiency symptoms, excessive supply or total nutrient depletion. Plants especially multicellular

ones require nutrient mobility for proper allocation of nutrient ions and translocation for the movement of assimilates between organs, through tissues and within cells (Gregersen *et al.*, 2008).

Nitrogen is an essential macro-element and 4th abundant plant element. It is a major constituent of several of the most important plant substances. Nitrogen compounds comprise 40% to 50% of the dry matter of protoplasm, and it is a constituent of amino acids, the building blocks of proteins (Swan, 1971). Nitrogen is utilized by plants from the soil by their roots majorly in the forms of nitrate ( $\text{NO}_3^-$ ); and nitrate is the only anion used by plants in large amounts. Another important form is ammonium ( $\text{NH}_4^+$ ). Nitrogen as an essential constituent of chlorophyll plays a major role in utilization of absorbed light energy and photosynthetic carbon metabolism (Huang *et al.*, 2004); but influences growth and utilization of sugars more than it influences photosynthesis through a reduction in chlorophyll.

Chlorophyll content is an important factor to determine the photosynthesis rate and dry matter production (Ghosh *et al.*, 2004). In a study on the relationship between chlorophyll content and nitrogen concentration in terms of leaf unit area, it was shown that the former has a high correlation with the latter (Karimi, 2001). Ghosh *et al.* (2004) reported an increase in chlorophyll content under stress due to the application of nitrogen fertilizer. Chlorophyll content is of particular significance to precision in agriculture as an indicator of photosynthetic activity.

The tomato, *Solanum lycopersicum* is an herbaceous annual plant of the Solanaceae family. Taxonomically, tomato plant initially belongs to genus *Solanum* (Linnaeus, 1753) and was later moved to its own genus with the name *Lycopersicon esculentum* (Miller, 1768). Tomato is a rich source of nutrients, dietary fibers, vitamin A and C, minerals such as iron and phosphorus. It is consumed in large quantities directly as salads, cooked into soups or processed as sauce, juice, paste, ketchup, chutney, puree, etc. Tomato is a very important crop in terms of diet and economy worldwide. Physiological studies on the availability of nutrients and their relationship to the growth and developmental processes in plants has been of great interest. Hence, it is imperative to study the role of nitrogen in influencing the production and yield of tomato plant to expound a better understanding of its physiological responses to nitrogen stress that will ultimately improve the productivity and availability of tomato on a global scale. The specific objective of this research work was to determine the effect of different levels of nitrogen application on the photosynthetic pigment accumulation and yield of *Solanum lycopersicum*.

## **MATERIALS AND METHODS**

Seeds of *Solanum lycopersicum* (Tomato-Roma long variety) tagged with VG-TH-017 were only grown and utilized during the experiment. The seeds were collected from National Research Institute, Akure. This is to ensure pureness in breed and

restrict variations in results due to species origin and variety. The initial stages of germination of seedlings were carried out in a nursery bed of plastic bath containing bored holes at the bottom to allow for drainage of excess water in the course of their emergence. The primary aim of germination in the nursery bed was to enhance the growth and development of the young seedlings of *S. lycopersicum* as much as possible alongside with the protection of the delicate vegetative parts. To a large extent, many viable seedlings emerged. At the end of the 28th day after sowing, the established seedlings were transplanted.

The established seedlings were transplanted into 40 plastic pots with 4 plants per pot filled near brim with soil with perforated holes at its base for drainage of excess water during the course of the experiment. The transplanted plants were kept in the green house to prevent extraneous materials from interfering with the experiment. The 40 plastic pots were divided into four groups, each group representing a particular treatment containing 10 plastic pots each. In general, the treatment consists of a nutrient solution with nitrogen sources as  $\text{KNO}_3$  and  $\text{NH}_4\text{NO}_3$  which is either in deficit, moderate or optimum supply or in excess to the desired alteration for stress and was applied once every day at 6p.m. in the evening.

The first group served as the control experiment and received treatment with no nitrogen supply in the nutrient solution (i.e. the nitrogen sources  $\text{KNO}_3$  and  $\text{NH}_4\text{NO}_3$  in the nutrient solution were removed) throughout the run of the experiment. The treatment was tagged as “n”. The second group received treatment of complete nutrient solution (the standard concentration of nitrogen i.e.  $\text{KNO}_3$  and  $\text{NH}_4\text{NO}_3$  was supplied as required basically in the nutrient solution) and the treatment was labelled as “N”. The third group was supplied with a treatment of nutrient solution in which the concentration of nitrogen was increased with a factor of 5 (i.e.  $\text{KNO}_3$  and  $\text{NH}_4\text{NO}_3$  were 5 times more in the composition of the nutrient solution) and the treatment were marked as “5N”. The fourth group was treated with nutrient solution containing a 10 multiplier factor of the concentration of nitrogen sources and the treatment was tagged as “10N”.

The plastic pots were labelled in order to differentiate, identify and observe the effect of the differential treatment in each group and replicates. The average light intensity in the screen house was measured using a digital luxmeter TCX100 while the average temperature was measured with a thermometer. The average light intensity and average temperature throughout the run of the experiment was realized to be 11380Lux and 35.2<sup>o</sup>C respectively. All the plants in the four treatments received 200 ml of distilled water at 6a.m. in the morning every day. At 6p.m. in the evening, the differential treatments were applied. The first group received 100 ml of nutrient solution void of nitrogen sources in the evening every day. The second group received 100 ml of complete nutrient in the evening every day. The third group received 100 ml of nutrient solution in which the concentration of nitrogen sources have been increased

by 5 (5N) in the evening every day. The fourth group received 100 ml of nutrient solution in which the concentration of nitrogen sources have been increased by 10 (10N) in the evening every day. The nutrient solution was prepared in accordance with the modified long Ashton formula (Hewitt, 1952).

Sampling was done on a weekly basis starting from the 35th day after sowing which is the 1<sup>st</sup> week after the application of the differential treatments. Plants were selected at random from each of the four treatments.

The photosynthetic pigments were determined spectrophotometrically. Fresh green tomato leaves were plucked from the differential treatments. 5.0g of these leaves were grinded in 20ml of 80% acetone and a piece of Na<sub>2</sub>CO<sub>3</sub> was added to prevent the degradation of chlorophyll to phaeophytin. The brew obtained was filtered using Whatman No 1 filter paper. Each coloured filtrate obtained from the four treatments were fed into separate cuvettes. The optical absorbance of the coloured solution from each treatments in the cuvettes were determined at two specified wavelengths, 647nm and 664nm respectively (Combs *et al.*, 1985). The chlorophyll contents were then determined as follows:

$$\text{Chlorophyll a} = 13.19(A_{664}) - 2.57(A_{647}); \text{Chlorophyll b} = 22.1(A_{647}) - 5.26(A_{664});$$

$$\text{Total chlorophyll} = 7.93(A_{664}) + 19.53(A_{647})$$

The quantitative yield of the plants in each treatment was determined. The number of flowers and fruits per plant per pot were counted and recorded. The fruit lengths and fruit diameters in each treatment were determined with the use of a Vernier caliper. The fruit fresh weights were determined on a weighing balance. The fruit dry weights were determined on a weighing balance after drying in Gallempamp oven at 80<sup>o</sup>C until a constant weight was achieved

Statistical analysis was carried out on SAS portable 9.13 software. Data collected from the study were subjected to analysis of variance (ANOVA) to test for differences among treatments and to investigate the effect of nitrogen levels on the photosynthetic pigment accumulation. The significant means were separated using LSD Fisher test at 0.05 confidence limit (alpha level) for the mean.

## RESULTS AND DISCUSSIONS

The chlorophyll “a” accumulation of the tomato plants subjected to different level of nitrogen stress did not follow a regular pattern from the beginning of the experiment to the end of the experiment (Fig. 1). The result of the ANOVA showed

**TABLE 1. The composition of nutrient solution**

| Nutrient              | Solution Composition  | Amount (g <sup>l</sup> <sup>-1</sup> ) |
|-----------------------|---|--|
| <b>Macronutrients</b> |   |  |
|                       | KNO <sub>3</sub>  | 0.216                                  |
|                       | NH <sub>4</sub> NO <sub>3</sub>   | 0.029                                  |
|                       | CaCl <sub>2</sub>   | 0.553                                  |
|                       | K <sub>2</sub> SO <sub>4</sub>  | 0.348                                  |
|                       | NaH <sub>2</sub> PO <sub>4</sub> .2H <sub>2</sub> O                               | 0.2104                                 |
|                       | MgSO <sub>4</sub> .7H <sub>2</sub> O  | 0.3801                                 |
|                       | Ferric Citrate  | 0.053                                  |
| <b>Micronutrients</b> |   |  |
|                       | MnSO <sub>4</sub>   | 0.003                                  |
|                       | CuSO <sub>4</sub>   | 3.537 X 10 <sup>-4</sup>               |
|                       | ZnSO <sub>4</sub> .7H <sub>2</sub> O  | 3.079 X 10 <sup>-4</sup>               |
|                       | H <sub>3</sub> BO <sub>3</sub>  | 0.0019                                 |
|                       | (NH <sub>4</sub> ) <sub>6</sub> Mo <sub>7</sub> O <sub>2</sub> .4H <sub>2</sub> O | 0.368 X 10 <sup>-4</sup>               |
|                       | CoSO <sub>4</sub> .7H <sub>2</sub> O  | 0.286 X 10 <sup>-4</sup>               |
|                       | NaCl  | 0.0058                                 |

that there was a significant difference in the chlorophyll “a” accumulation of the 10N-plants, 5N-plants, N-plants and n-plants ( $p>0.05$ ).

The chlorophyll “b” accumulation of the tomato plants grown under different nitrogen treatments did not follow a particular pattern during the period of the experiment (Fig. 2). The result of the ANOVA showed that the chlorophyll “b” accumulation of the 10N-plants, 5N-plants, N-plants and n-plants were significantly different from one another ( $p>0.05$ ).

The total chlorophyll accumulation of the tomato plants in each nitrogen treatment followed an irregular pattern throughout the course of the experiment (Fig. 3). The result of the ANOVA showed that the total chlorophyll accumulation of the 10N-plants, 5N-plants, N-plants and n-plants were significantly different from one another ( $p>0.05$ ).

The highest number of flowers was recorded in the 5N-plants (Table 2). The result of the ANOVA showed that number of flowers of the 10N-plants, 5N-plants and N-plants were significantly higher from the n-plants ( $p>0.05$ ). The number of fruits of 5N-plants were significantly higher than the N-plants and n-plants but was not significantly different from the 10N-plants ( $p>0.05$ ). The n-plants recorded no number of fruits throughout the course of the experiment (Table 2).

The fruit length as well as the fruit diameter of the 10N-plants, 5N-plants and N-plants were not significantly different ( $p>0.05$ ). The fruit fresh weight as well as the fruit dry weight of the 10N-plants, 5N-plants and N-plants were not significantly different ( $p>0.05$ ).

In the present study of the photosynthetic pigment accumulation of *S. lycopersicum* as affected by different levels of nitrogen application, all the conditions needed for normal growth and development of the tomato seedlings were maintained except that some seedlings were exposed to nitrogen stress while others were supplied with normal nutrient application. Other environmental conditions were the same for all the seedlings throughout the experimental period. Any differences observed during the experimental course could therefore be attributed to the differential levels of nitrogen application that was introduced during the experiment.

The methodologies used for chlorophyll extraction in plant materials are almost always based on methods that destructively extract leaf tissue using organic solvents that include acetone (McKinney, 1941; Bruinsma, 1961), dimethylsulfoxide (DMSO) (Hiscox & Israelstam, 1979) methanol, N,N-dimethyl formamide and petroleum ether (Moran & Porath, 1980; Moran, 1982; Lichtenthaler & Wellburn, 1983; Inskeep & Bloom, 1985). The 10N-plants and 5N-plants had leaves with deep-green colouration indicating an increase in chlorophyll content as well as an increase in the photosynthetic capacity. Evidence shows that plants with a higher nitrogen supply develop greener leaves unlike plants growing in nitrogen-deficient conditions which

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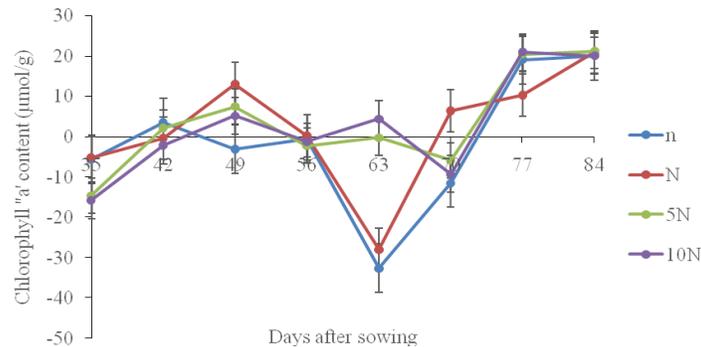
exhibit pale-green colouration of leaves or in extreme cases, uniform chlorosis (Kozlowki, 1985; Perry & Hickman, 1998).

The highest number of flowers and early flowering discovered in 10N-plants and 5N-plants might probably be due to the fact that nitrogen increases the biosynthesis of cytokinin in plants. The presence of cytokinin in the floral meristem induce floral buds to produce more numbers of flowers (Ding *et al.*, 2014). Bahnisch & Humphreys (1977) found out that additional nitrogen supply increase the fertility of the main stem and accelerated flowering.

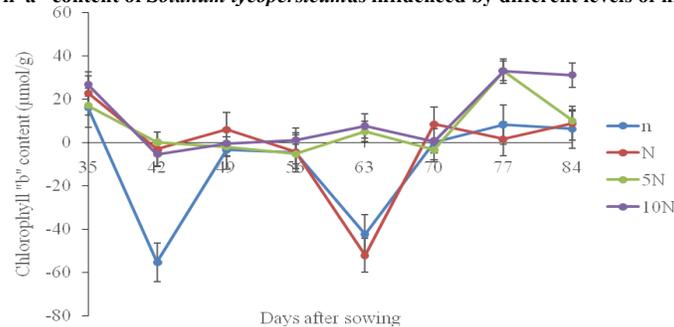
**TABLE 2.** Yield parameters of *Solanum lycopersicum* influenced by different levels of nitrogen application.

| Nitrogen treatments | Number of flower   | Number of fruit     | Fruit length (mm)  | Fruit Diameter (mm) | Fruit Fresh Weight (g) | Fruit Dry Weight (g) |
|---------------------|--------------------|---------------------|--------------------|---------------------|------------------------|----------------------|
| n                   | 6.20 <sup>b</sup>  | 0.00 <sup>c</sup>   | 0.00 <sup>b</sup>  | 0.00 <sup>b</sup>   | 0.00 <sup>b</sup>      | 0.00 <sup>b</sup>    |
| N                   | 21.90 <sup>a</sup> | 18.00 <sup>b</sup>  | 50.10 <sup>a</sup> | 41.20 <sup>a</sup>  | 42.92 <sup>a</sup>     | 2.15 <sup>a</sup>    |
| 5N                  | 24.70 <sup>a</sup> | 46.00 <sup>a</sup>  | 47.70 <sup>a</sup> | 40.90 <sup>a</sup>  | 49.15 <sup>a</sup>     | 2.80 <sup>a</sup>    |
| 10N                 | 22.50 <sup>a</sup> | 32.00 <sup>ab</sup> | 49.73 <sup>a</sup> | 38.83 <sup>a</sup>  | 45.61 <sup>a</sup>     | 2.47 <sup>a</sup>    |
| Mean                | 18.83              | 24.00               | 36.88              | 30.23               | 34.42                  | 1.86                 |
| LSD                 | 8.53               | 15.37               | 0.46               | 0.69                | 10.22                  | 1.08                 |

Means with the same letter within the same column are not significantly different at P > 0.05.



**FIG. 1.** Chlorophyll "a" content of *Solanum lycopersicum* influenced by different levels of nitrogen application.



**FIG. 2.** Chlorophyll "b" content of *Solanum lycopersicum* influenced by different levels of nitrogen application.

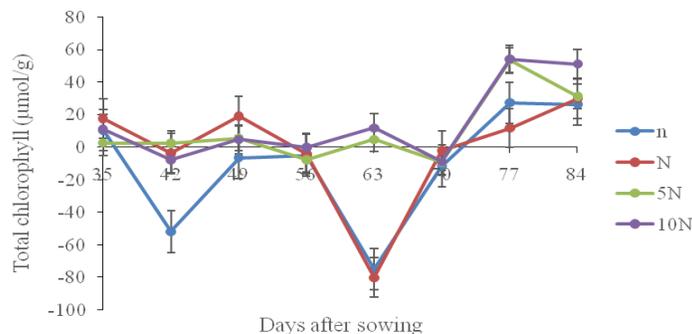


FIG. 3. Total chlorophyll content of *Solanum lycopersicum* influenced by different levels of nitrogen application

The observed higher yield in the 5N-plant are in conformity with the research of Lal (1992) who found that higher yield were responsible for better development of fruits, increased uptake of nutrients in the plants leading to enhanced chlorophyll content and carbohydrate synthesis, higher accumulation of photosynthates and their distribution to the developing ovules. Similar findings have been reported by Kulvinder & Srivastava, (1988) that higher yield might be due to a greater amount of carbohydrates which were translocated to fruits in plants grown at optimal levels of nitrogen. The yield in the 10N-plants was observed to be lower than the 5N-plants and agreed to the report of Adams *et al.* (1978) who reported that optimum nitrogen application to the soil produces high fruit yield and improve fruit quality, whereas, excessive application leads to luxuriant development of vegetative parts of the plant at the expense of reproductive growth (Tisdale *et al.*, 2003). It might due to the fact that the increased vegetative growth of the plants accustomed to high supply of nitrogen becomes indisposed to divert some of its nutrient for fruit development and rather choose to the pattern of continuous vegetative growth without regression. The n-plants produced no fruit as a result of nitrogen-deficiency which reduced the branching of the plants making them to have an overall appearance of short, spindly plants that are more concerned with recuperating rather than reproduction phases. Too little nitrogen encourages abortion of tomato flowers since the plant cannot produce enough food the support itself (Picken, 1914).

### CONCLUSIONS

The best yield was obtained in the treatments for the 5N-plants in which the concentration of nitrogen in the nutrient solution had been increased to a factor of 5. It is therefore suggested that when the seeds of tomato plants ((Tomato-Roma long variety) are to be grown, the 5N-plants treatment is the most suitable level of application for shorter cultivation periods and higher yield because they could flower early and produce an optimum amount of good quality fruits.

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