

## **EFFECTS OF GIBBERELLIC ACID (GA<sub>3</sub>) ON THE GROWTH, FRUIT YIELD AND QUALITY OF STRAWBERRY (*FRAGARIA* × *ANANASSA*) IN A SUB-TROPICAL ENVIRONMENT**

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### **ABSTRACT**

*Strawberry (Fragaria x ananassa) is a widely adapted small fruit grown from the low-altitude of tropics and subtropics to high-altitude in cold continental areas. Despite its nutritional benefits, little research has been undertaken to facilitate its wide scale production in sub-tropical environments. This experiment was laid down in a Randomised Complete Block Design (RCBD) with four replications. Its aim was to determine the effects of different concentrations (0, 25, 50, 75 ppm) of gibberellic acid (GA<sub>3</sub>) on growth, yield and quality of strawberry. The results revealed that plants treated with 75 ppm GA significantly (P<0.05) had more growth in number of leaves, petiole length, plant spread and leaf area. Less leaf nitrogen (1.78%) and chlorophyll content index (25.01) were observed in GA treated plants with 75 ppm. Control plants sprayed with distilled water took more days to produce first flower (62.25 days) and fruit (69 days) as compared to GA treated plants which also produced fruits with more weight and yield which increased with increasing concentrations. Titratable acidity of the fruits was significantly (P<0.05) lower in plants treated with 75 ppm GA<sub>3</sub> and had higher vitamin C (71.88 mg/g) and total soluble solids (6.90° Brix). Results from GA treated plants at 50 ppm were the best in this experiment but more research needs to be done in the open field as this one was done in a lathhouse with plants grown in containers; differences might be observed in the field and there is need for more research on strawberry and bioregulators in the different ecological areas of Swaziland.*

**Keywords:** strawberry, gibberellic acid, yield, quality, subtropical environment

### **INTRODUCTION**

The cultivated strawberry is a hybrid plant between two American species, *Fragaria chiloensis* of western North and South America and *Fragaria*

*virginiana* of eastern North America (Lolaei *et al.*, 2012). The commonly cultivated strawberry is *Fragaria* × *ananassa*. The hybridization of the two species occurred around 1850 in France and hundreds of varieties have been selected and named since then. Although other *Fragaria* species are also cultivated, this hybrid is the primary source of commercially produced strawberries. Strawberries are very nutritious fruits and excellent sources of vitamin C, antioxidants and flavonoids. The strawberry fruit is very low in calories, saturated fat, cholesterol and sodium. It is also a good source of folate and potassium, and a very good source of dietary fibre, and manganese and ellagic acid, a nutraceutical that has powerful anti-oxidant and anti-carcinogen properties (Thiele, 1986).

Commercial strawberries are successfully grown in a broad range of climates including temperate, grassland, Mediterranean and subtropical regions (Hancock, 2000), however, production of strawberries in the sub/tropics is not uniform due to poor winter chilling (Darnell *et al.*, 2003) which results in poor flower initiation, poor fruiting and low total yields. There is a need to find suitable plant bioregulators like gibberellins which have been used in research trials for strawberry production in the temperate regions and had shown to have a positive effect on growth and yield. The objective of this study was to determine the effects of gibberellic acid applications on the growth, fruit yield and quality of strawberry fruits in a subtropical environment.

## **MATERIALS AND METHODS**

### **Experimental site**

The research was carried out at the University of Swaziland (UNISWA), Faculty of Agriculture, Luyengo Campus, at the Horticulture Department Farm lathhouse (black 50% netshade; Mahalaxmi, India) between September 2014 and February 2015. The farm is located at Luyengo, Manzini Region, on the Middleveld agro-ecological zone, 26°0 32' S – 31°0 14' E (Murdock, 1970), and the average altitude of this area is 750 m above sea level. The annual mean precipitation is 980 mm with most of the rain falling between October and March (MOAC, 2004).

### **Treatments and experimental design**

The research was done using the garden strawberry runners and they were obtained from a local farmer. Strawberry runners were planted in a potting

media comprising of sand, soil and compost mixture in a 1:1:1 ratio, in 3.5 liter polythene bags. The bags were spaced at 30 by 35 cm in the lathhouse. Gibberellic acid (GA<sub>3</sub>) was used as the treatment with three concentrations of 25, 50, 75 ppm and distilled water was used as the control treatment. The experiment was laid down in a randomised complete block design (RCBD) with four replications. The plants were watered thrice weekly depending on the weather and weeds were removed when they appeared.

### **Treatment preparations**

One litre solutions of GA<sub>3</sub> at 25, 50 and 75 ppm (90% pellets, Abbott Laboratories, North Chicago, Illinois, USA). A stirrer and rod were used to help dissolve the gibberellic acid pellets. All the treatment concentrations were applied by spraying using a 5 litre knapsack sprayer to a runoff onto the plant leaves once two weeks after transplanting.

### **Vegetative growth parameters**

Plant growth parameters for strawberry were measured from four plants of randomly selected and tagged plants, at intervals of 2, 4, 6 8 and/or 10 weeks after transplanting (WAT) during the experimental period. Leaf number was counted using the triplicate leaves per randomly tagged plant per treatment and the total number was recorded. The leaf petiole length and plant spread were measured using a 30 cm ruler per tagged plant per treatment concentration; the total number was recorded.

The number of runners and the runner plants were recorded for the various treatments. Leaf area was measured by taking leaf length and breadth using a 30 cm ruler for all the treatments. The measurements were multiplied together with a correction factor (L x W x 0.75) (Edje and Ossom, 2009). Leaf nitrogen (%) was determined through the digestion and distillation processes (AOAC, 1990). Chlorophyll content index was measured from randomly selected plant leaves using a chlorophyll content meter (CCM-200, Opti-Sciences; Chicago, Illinois, USA) which gives a chlorophyll content index (CCI) value as an estimate for leaf chlorophyll. Leaves were placed in-between the CCM lever and a CCI value was displayed on the screen and recorded; readings were taken fortnightly from the fourth week to the tenth one.

### Fruit parameters

Days to first flowering and fruit formation, and the number of flowers and fruits per plant were recorded for all the treatments. Fruits were weighed using a balance (HCB602H, Adam Equipment, Johannesburg, South Africa). A refractometer (Master.53T Brix~53%, Tokyo, Japan) was used to measure the total soluble solids in the fruits. Fruit juice was extracted by squeezing the fruit, a drop of the juice was put on the prism which was then closed to allow taking of the readings. Fruit yield was calculated by multiplying the number of flowers, number of fruits and the fruit weight per concentration and treatment. Titratable acidity in fruits was measured by firstly weighing 50 g of fruits for the treatments using procedures from AOAC (1990). Ascorbic acid was determined the procedures from the University of Canterbury (2014).

### Data analysis

Collected data was subjected to analysis of variance (ANOVA) using Statistical Analysis Software (SAS, 1990). Means, where statistical differences were detected, were compared using Duncan New Multiple Range Test (DNMRT) at P=0.05 (Gomez and Gomez, 1984).

## RESULTS

### Temperature and relative humidity

The average temperatures and relative humidity during the experimental study are shown below in Table 1.

Table 1. Average temperatures and relative humidity

Month	Temperature (°C)		Relative humidity (%)	
	Minimum	Maximum	Morning	Afternoon
September	15	28	70	49
October	14	24	87	66
November	17	26	85	73
December	17	26	89	71
January	17	28	83	87
February	17	35	89	74

### Vegetative growth

Leaf number of strawberry plants was significantly ( $P < 0.05$ ) different in all the concentrations. Plants sprayed with GA<sub>3</sub> at 50 ppm significantly produced more leaves (Figure 1) than the other concentrations; the number of leaves reduced at 75 ppm while control plants had the least values. Long petioles were observed in plants treated with 75 ppm of GA (Figure 2) followed by 50, 25 ppm and control plants. There was an increasing trend in leaf number and petiole length for all the concentrations which was evidently observed every fortnight after transplanting with week eight having the best results for leaf number and week ten for petiole length.

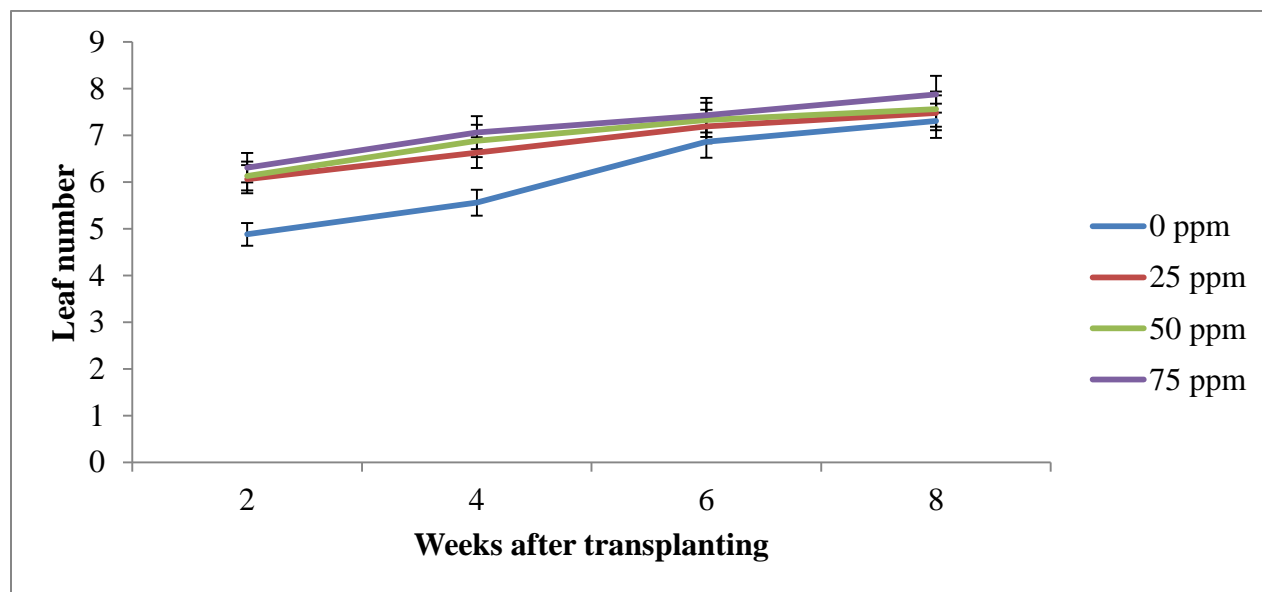


Figure 1. Effects of gibberellic acid on leaf number of strawberry. Bars are standard error (S.E.) below and above the mean

Significant differences ( $P < 0.05$ ) were observed for plant spread in all concentrations with GA treated plants at 75 ppm having the best (Figures 3). Control plants had the least plant spread followed by plants applied with 25 and 50 ppm GA<sub>3</sub>. The increase in plant spread was observed with every increase in treatment concentration and the number of weeks after transplanting. Plants sprayed with gibberellic acid at 75 ppm had significantly ( $P < 0.05$ ) more leaf area (Figures 4) compared to other concentrations with control plants having the least leaf area. The increase in plant spread and leaf area was due to the high number of leaves and leaf

petioles which were spreading outwardly and hence creating bushy plants. The triplicate leaves of strawberry also made a huge increase in plant spread and leaf area as it can be compared to plants with single leaves.

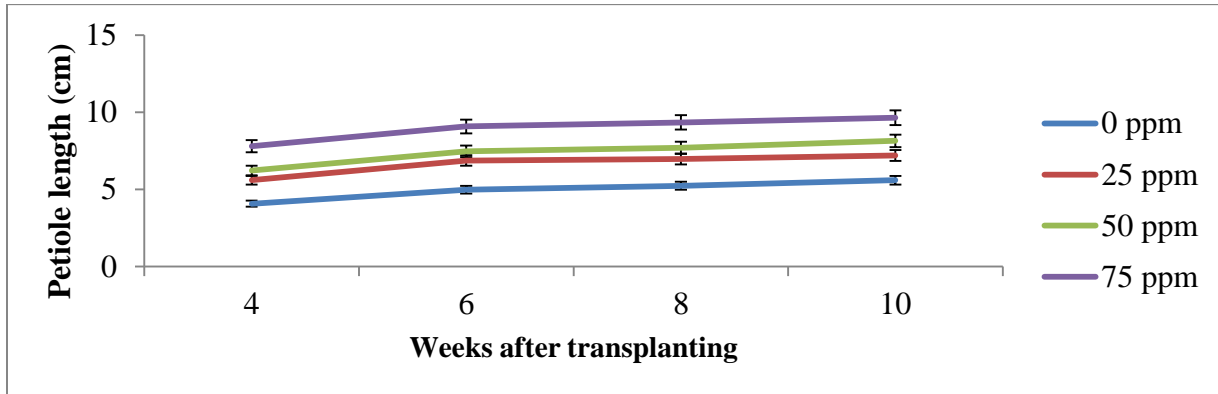


Figure 2. Effects of gibberellic acid on petiole length of strawberry. Bars are standard error (S.E.) below and above the mean

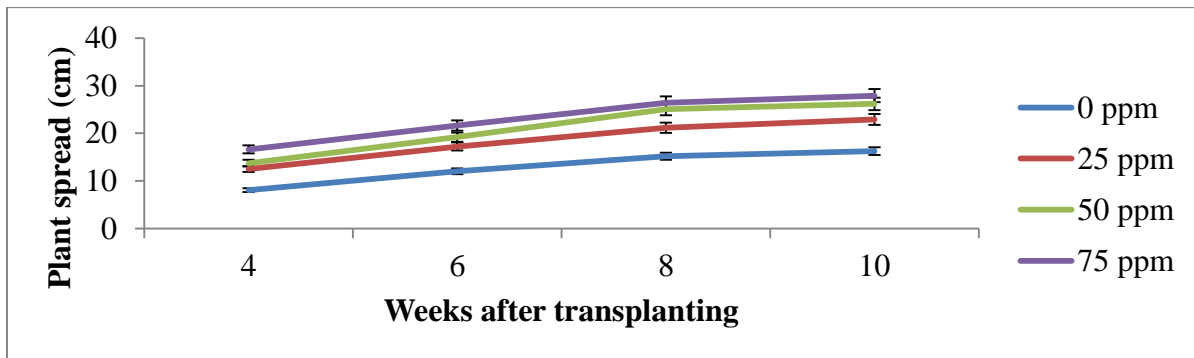


Figure 3. Effects of gibberellic acid on plant spread of strawberry. Bars are standard error (S.E.) below and above the mean

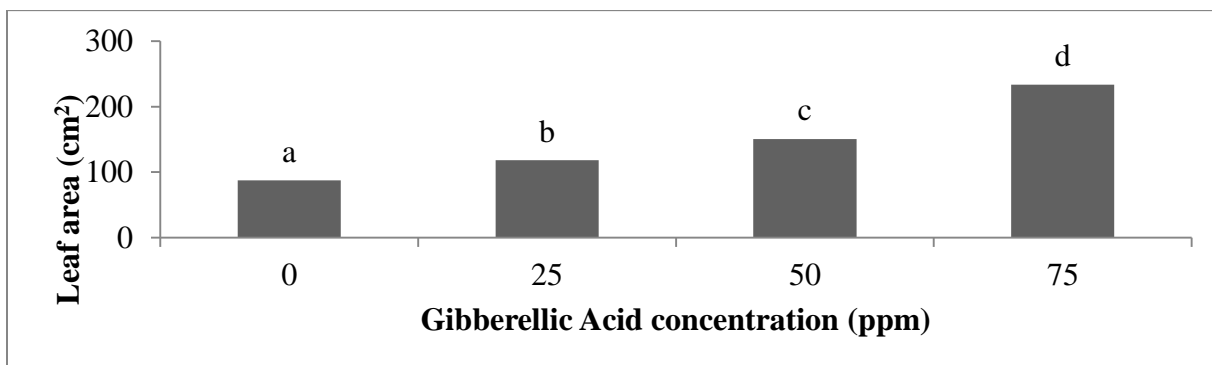


Figure 4. Effects of gibberellic acid on leaf area of strawberry. Bars followed by same letter not significantly different. Mean separation by DNMRT at P=0.05

Both percentage leaf nitrogen (N) and chlorophyll content index (CCI) were significantly ( $P < 0.05$ ) different with decreasing amounts for GA concentrations. Plants treated with 75 ppm of GA had the lowest leaf nitrogen (1.78%) while control plants had the highest value (Figure 5). Control plants had more leaf nitrogen as compared to GA treated plants which reduced with an increase in GA<sub>3</sub> concentration. Week ten after transplanting had the best leaf nitrogen and week 15 had the lowest. Chlorophyll content index (CCI) was highest in control plants (Figure 6) and lowest (25.01) at 75 ppm in GA treated plants; the CCI decreased for GA treated plants with increasing GA<sub>3</sub> concentrations; at week 8, the plants had the highest values which then decreased at week 10.

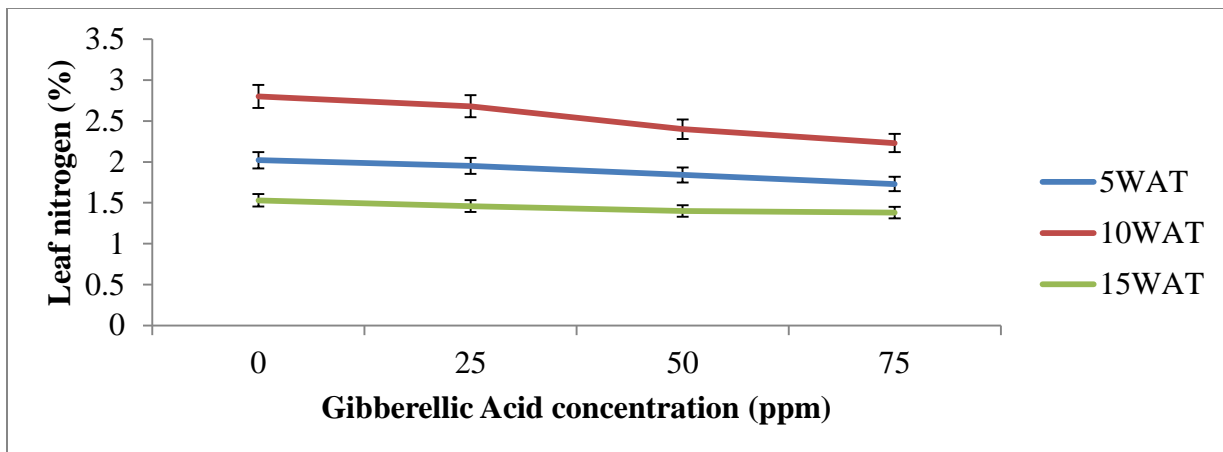


Figure 5. Effects of gibberellic acid on leaf N (%) of strawberry. Bars are standard error (S.E.) below and above the mean

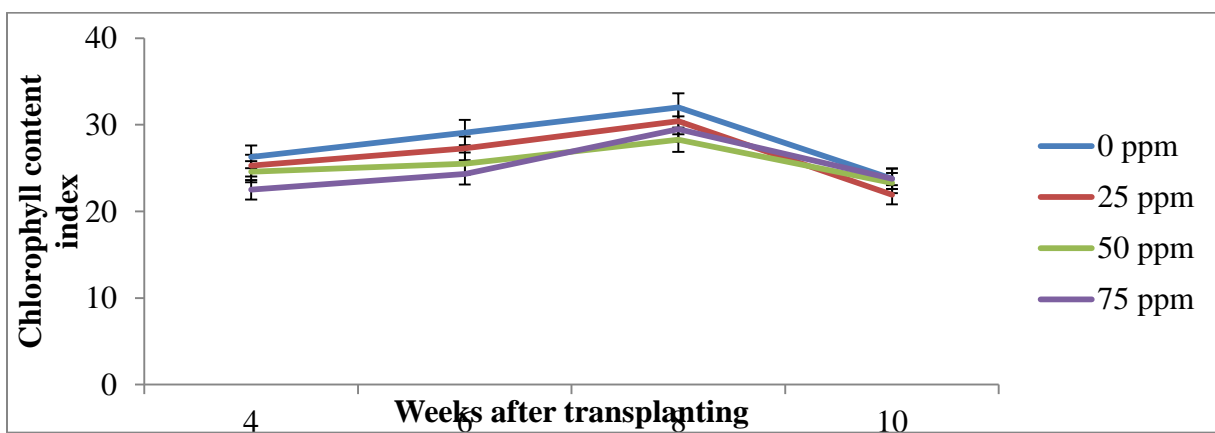


Figure 6. Effects of gibberellic acid on chlorophyll content index of leaves of strawberry. Bars are standard error (S.E.) below and above the mean

### Reproductive growth

There were significant ( $P < 0.05$ ) differences between treatments for the number of days to first flowering and fruit formation; and the number of flowers and fruits formed. The number of days to first flowering and fruiting decreased with each increase in concentration for GA (Figure 7). The GA treated plants took a few days (57) at 75 ppm to start producing flowers and fruits (63.5 days). Plants sprayed with 50 ppm of GA produced more flowers (17) and fruits (13.44) (Figure 8). Both flower and fruit numbers were increasing with increases in GA concentrations but fruit numbers were less than the flower numbers.

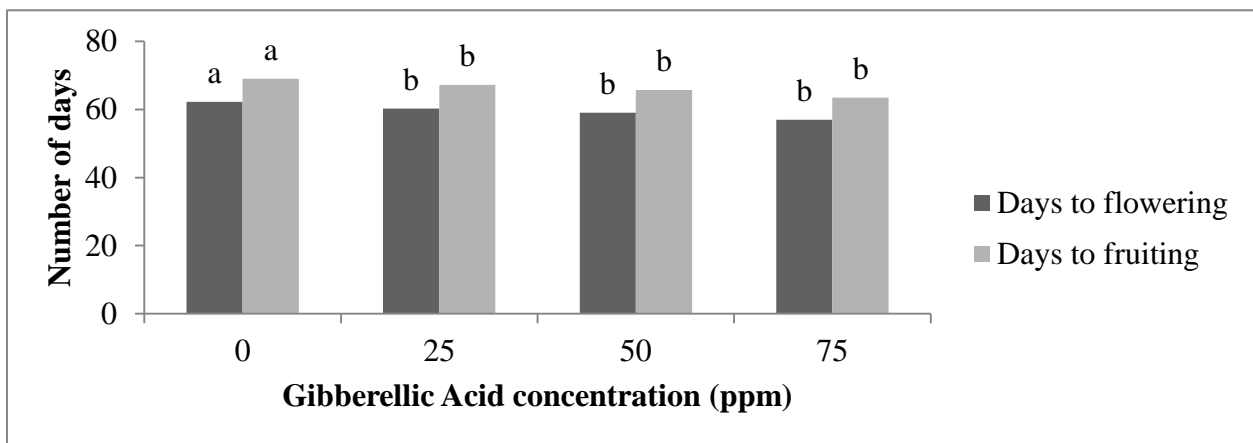


Figure 7. Effects of gibberellic acid on the number of days to first flowering and fruiting of strawberry. Bars of similar shade followed by same letter not significantly different. Mean separation by DNMRT at  $P = 0.05$

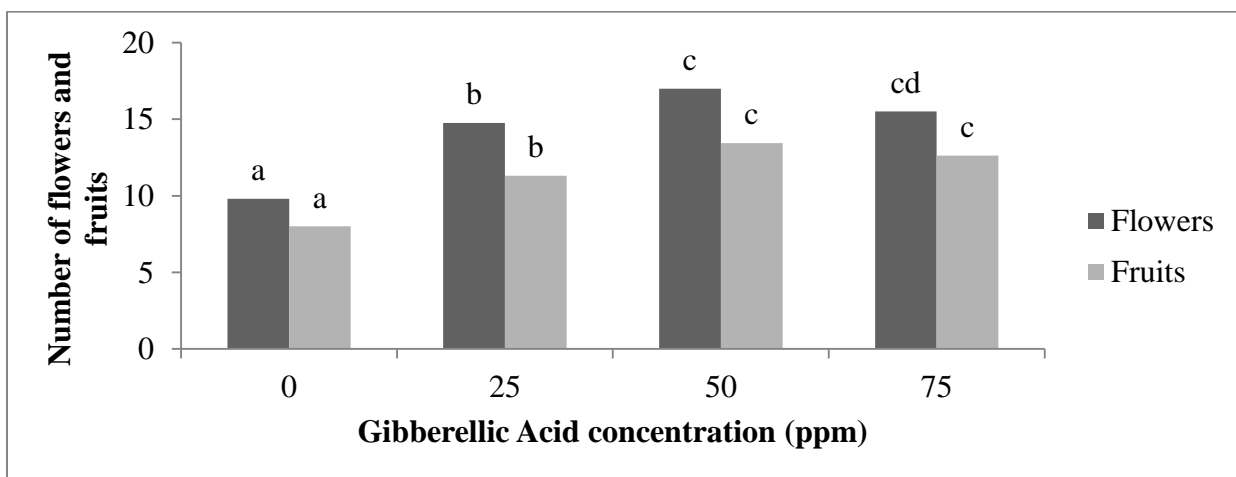


Figure 8. Effects of gibberellic acid on the number of flowers and of strawberry. Bars of similar shade followed by same letter not significantly different. Mean separation by DNMRT at  $P = 0.05$



Fruit weight increased with increased concentrations of GA<sub>3</sub> with control fruits having the least weights. Fruits from plants treated with GA had higher weight (3.31g) at 75 ppm (Figure 9). Yield from GA treated plants was significantly higher (Figures 10). However, GA treated plants produced higher yield (668.49g per plant) at 50ppm as compared to other treatments. Fruit yield was directly associated with the number of plant leaves, leaf area, number of flowers and fruits produced and the fruit weight. Treatments with high values for the above mentioned parameters are expected to have higher yields; this was observed in GA sprayed plants.

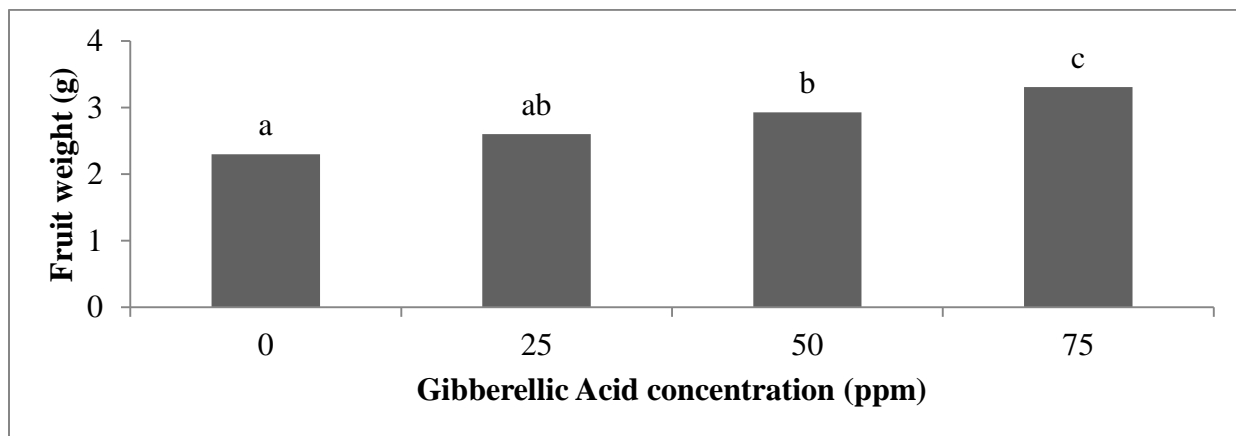


Figure 9. Effects of gibberellic acid on fruit weight of strawberry. Bars followed by same letter not significantly different. Mean separation by DNMRT at P=0.05.

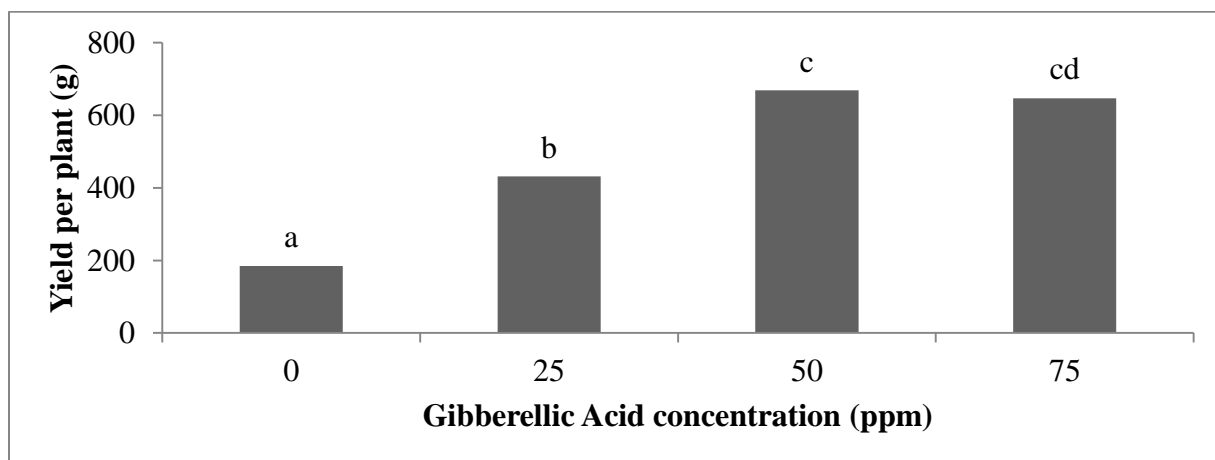


Figure 10. Effects of gibberellic acid on fruit yield per plant of strawberry. Bars followed by same letter not significantly different. Mean separation by DNMRT at P=0.05

### Fruit quality

Total soluble solids of fruits increased with each increase in concentration of GA, while their titratable acidity was decreased with increasing concentrations (Figure 11 and 12).. Fruits from GA treated plants (75 ppm) had the highest vitamin C (Figure 13) followed in decreasing order by fruits from 50 ppm, 25 ppm and control plants. Significant differences ( $P < 0.05$ ) in total soluble solids, fruits' titratable acidity and vitamin C content were observed in all concentrations.

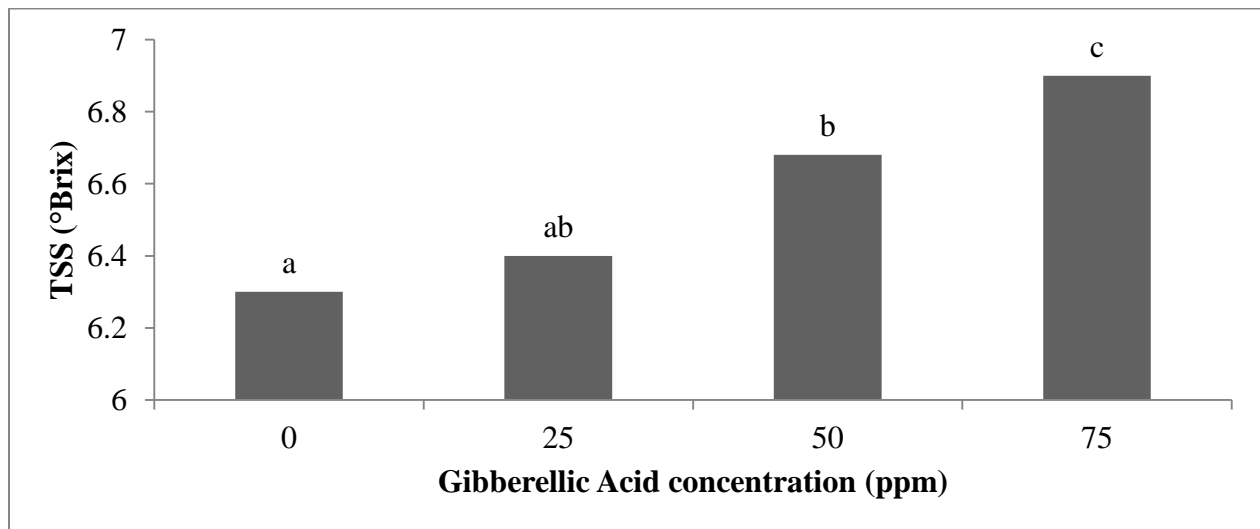


Figure 11. Effects of gibberellic acid on the TSS of fruits. Means (each bar) with different letters are significantly different ( $P < 0.05$ )

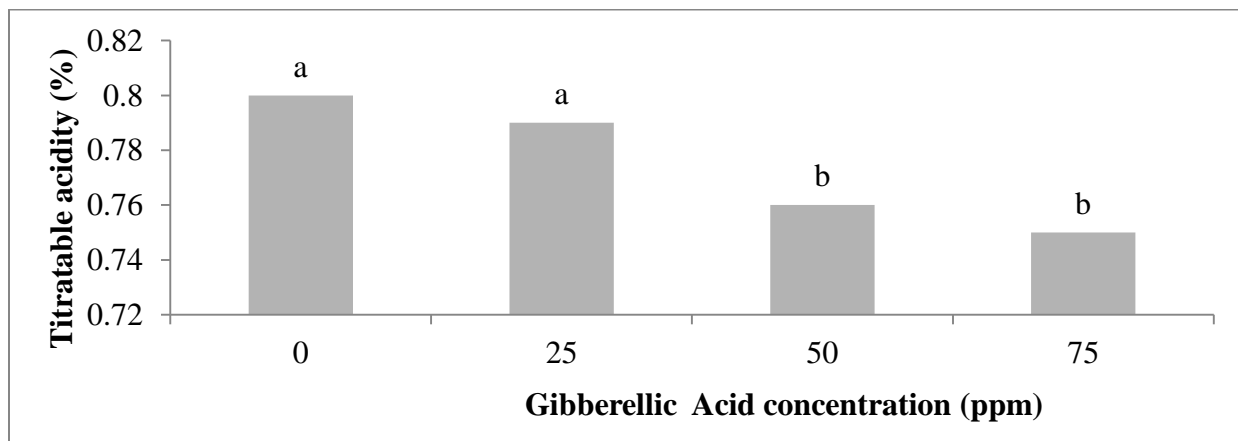


Figure 12. Effects of gibberellic acid on the titratable acidity of fruits. Means (each bar) with different letters are significantly different ( $P < 0.05$ )

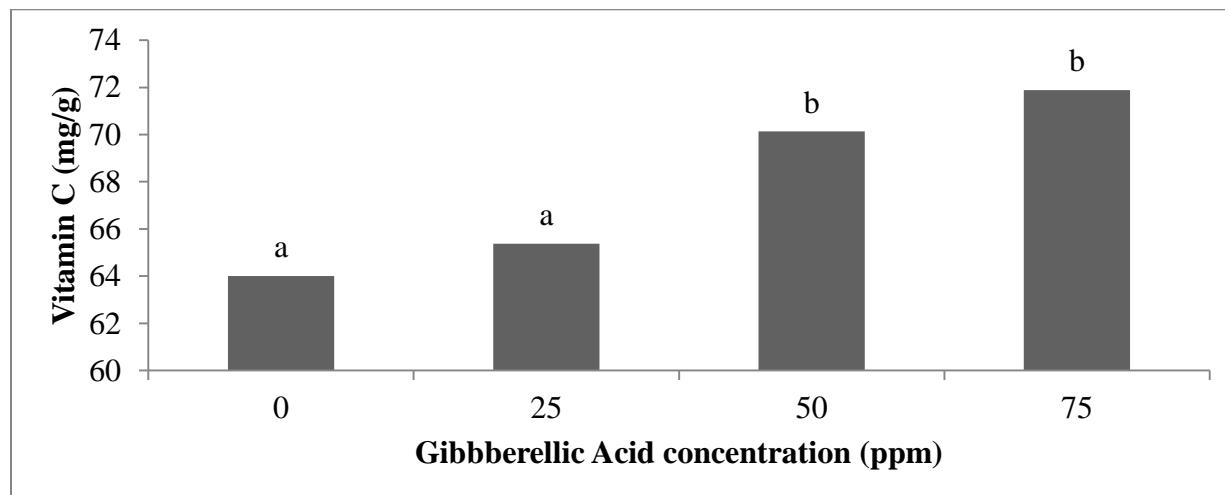


Figure 13. Effects of gibberellic acid on the vitamin C of fruits. Means (each bar) with different letters are significantly different ( $P < 0.05$ )

## DISCUSSION

Gibberellins regulate growth by causing cell elongation in the plant system. This is due to the fact that GA increases cell division, elongation and a corresponding increase in the epidermal and parenchyma's cell length (Saima *et al.*, 2014). This is in agreement with the results obtained in this study where there was an increase in leaf number and petiole length after the plants were sprayed with different GA concentrations. The increase in leaf number per plant by  $GA_3$  could possibly be explained by  $GA_3$  reducing the plastochron, which is the time between successive leaf initiation events (Emongor and Ndambole, 2011).

Increased plant spread after  $GA_3$  treatment may be due to the increased length of certain internodes which were either in the process of elongation at the time of treatment or were differentiated soon thereafter (Lolaei *et al.*, 2013). Nanda and Purohit (1965) reported the enhancement of growth by  $GA_3$  in relation to the mobilization of reserve starch. Due to enhanced mobilization by  $GA_3$ , large amounts of food material are available over a shorter period, causing a spurt in the growth processes (Nanda and Purohit, 1965) and this resulted in GA sprayed plants having a greater plant spread. Results from Paroussi *et al.* (2002) are in agreement with those of this study, as application of gibberellic acid increased leaf area of the strawberry plants.

Leaf nitrogen and the chlorophyll content for GA sprayed plants decreased with an increase in GA concentration. This suggests that chlorophyll synthesis was enhanced by lower GA<sub>3</sub> concentrations and higher concentration of GA<sub>3</sub> could have had a negative effect on chlorophyll synthesis. According to a study by Moneruzzaman *et al.* (2011), the chlorophyll readings in wax apple leaves from all the treated branches were the highest in the 20 mg/L GA<sub>3</sub> treated branch, followed by control and 50 mg/L GA<sub>3</sub>, whereas GA<sub>3</sub> 100 mg/L treated branch showed the lowest value.

Nitrogen values ranged from 1 to 3%, which are within reported ranges for strawberry leaf tissues (Casteel, 2011). Peng *et al.* (1993) found that most within-species variation in relationships between CCI and N could be explained by differences in leaf thickness and how they perceive light; these relationships can be improved by calculating specific leaf weight (Peng *et al.*, 1993) or considering N on an area, rather than dry weight basis. The results indicated that leaf N and CCI contents of strawberry cultivars showed great variability but in a linear pattern which can probably be used to conclude that CCI provides a linear approximation of total N.

Chlorophyll meter values are based on light absorption by the leaf chlorophyll at specific spectral bands and they may be affected by the degree of yellowness or greenness of the leaf. Chlorophyll content has been used as a direct indication of plant health and condition; it can also be used to manage nutrient optimization programmes that both improve crop yield and help protect the environment (Casteel, 2011). Changes in chlorophyll content can occur as a result of nutrient deficiencies, exposure to environmental stress, and exposure to certain herbicides and differences in light environment during growth (shading).

Strawberries require cold temperatures to initiate flowering but this can be bypassed by use of GA in hot environments hence reducing the time needed before flower induction and fruit formation. Results from this study have shown that GA treated plants at 75 ppm took minimum days (57 and 63.5) to initiate flowering and fruit formation respectively. This was in agreement with reports from Kasim *et al.* (2007) and Paroussi *et al.* (2002) who reported that gibberellic acid initiated early flowering, number of flowering trusses per crown and thus early fruit development and harvesting. Studies done by Singh and Singh (2006) and Ozguven and Kaska (1990) revealed that gibberellic acid caused the production of large numbers of flowers with

rapid elongation of the peduncle, leading to full development of flower buds having all reproductive parts functional which increases fruit set and number of berries per plant. Fruits produced in all treatments were less than the number of flowers; this might have been due to flower abortion, drop and/or fruit drop.

The increase in fruit weight for all the treated plants could possibly have been due to increasing cell size and/or cell numbers. El-Kosary (2009) reported an increase in weight after spraying 'Samany' and 'Zaghloul' date palm cultivars with GA at 50, 100 and 150 ppm four weeks from pollination. The increase in fruit yield due to GA<sub>3</sub> application observed in the current study was attributed to the enhanced vegetative growth (leaf area and leaf number per plant), dry matter accumulation and increase in yield components (number of flowers and fruits, and fruit weight) [Huang and Huang, 2005]. Yield capacity of strawberry plants can be influenced by more complex environmental factors other than flowering. Chilling effect as a temperature regulation method is indirect and mediated through flowering time, carbohydrate reserve, vegetative vigor (Darnell *et al.*, 2003) and photoperiod (Hytonen *et al.*, 2004).

Fruits with higher contents of total soluble solids usually have low titratable acidity as evidenced by the results of this study. In a study by Lolaei *et al.* (2013), GA reduced TSS and increased TA for strawberry cultivar Selva and Queen Elisa. Huang and Huang (2005) reported that application of growth regulators like auxin and gibberellins can significantly increase the total soluble contents of the fruit in citrus species. Early foliar spray treatments containing gibberellic acid significantly lower the concentration of the bitter flavonoid naringin and fruit acid in fruit tissues while having no changes in TSS. Salicylic acid has also been observed to increase total soluble solids content and not affect the titratable acidity of fruits (Fariduddin *et al.*, 2003). Favell (1998) concluded that titratable acidity which is mainly citric acid can be used as an appropriate marker for monitoring quality changes in fruits and vegetables during transportation, storage and processing.

Vitamin C or ascorbic acid is important for general health and its consumption is advised by health practitioners. Vitamin C content increased in strawberry fruits due to application of gibberellins (Ozguven and Yilmaz, 2002; Paroussi *et al.*, 2002). Application of gibberellins was beneficial to

green tea quality, increasing vitamin C content by 18% (Liang *et al.*, 1996). Ascorbic acid is also in high contents in immature fruits as it was in our control fruits. Nagy (1980) reported that immature citrus fruits contained the highest concentration of vitamin C whereas ripe fruits contained the least.

## CONCLUSIONS

Plant bioregulators can be used to improve growth and production of strawberry in a subtropical environment. Gibberellic acid applications led to increased vegetative and reproductive growth of strawberry; growth increased with each increase in the gibberellic acid concentration with 75 ppm giving the best results. Fifty (50) ppm of GA gave the best results for the reproductive growth. Fruits from the highest GA concentration had less titratable acidity while vitamin C and total soluble solids were high.

## ACKNOWLEDGEMENTS

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