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The Influence of Exogenous Hormone on the Flowering and Fruiting of Strawberry (*Fragaria x ananassa* Duch)

Isam Abdulbaset Haidar Al-madhagi [1],*, Sayed M. Zain Hasan [1], Aziz bin Ahmad [2], Abdullah M.Zain[1] and Wan Abdullah bin Yusoff [3]

[1]Faculty of Agrotechnology and Food Science, University Malaysia Terengganu (UMT), 21030 Kuala Terengganu, Malaysia.
[2]Faculty of Science and Technology, (UMT) University Malaysia Terengganu (UMT), 21030 Kuala Terengganu, Malaysia.
[3]Malaysian Agricultural Research and Development Institute (MARDI), Cameron Highlands, Pahang, Malaysia
*Corresponding Authors: isam.madhagi@gmail.com

Abstract
The influence the exogenous hormone on flowering and fruiting of strawberry was conducted at the Malaysian Agricultural Research and Development Institute (MARDI) station in Cameron Highlands. Factorial experiment with three replications was designed in randomized completed block design (RCBD) where two factors, cultivars and exogenous hormone were used. Two strawberry cultivars, Camarosa and Camaroga *cvs*., were grown under green house and treated with 0 and 50 ppm of auxin (IBA), gibberellins acid (GA3) or cytokinin (6-BA) either singly or in combination by foliage application. The results showed that, there are significant different between the mean of flowering parameter, percentage of fruit set and average of fruit weight among the two cultivars. The result also showed that the number of the flowers per plant was greatly increased by about 138% comparing with the control plants when treated with 50 ppm GA3. 6-BA and IBA shows an antagonized effect on the role of GA3 in producing the flowers. The percentage of fruits set had increased by about 97.24 % and 81.5 % after treated with IBA and GA3, respectively. Combined application of GA3+6BA had increased the fruit weight by 33.85% compared with the control plants. The result also indicated that these two cultivars of strawberry response differently to the exogenous hormones in the producing flowers and fruits. The first bud of the plant treated with IBA+6-BA formed to flower as compared with the plants treated with GA3. The result obtained from of the present study could be used to control the flowering and fruiting of strawberry.
Keywords: Strawberry, exogenous hormone, flowering, fruiting.

1. Introduction

Strawberry plant is belongs to the Rosaceae family. The commercial variety of strawberry (Fragaria x ananassa Duch.) has cultivated in about 75 countries (FAO, 2007) . FAO statistics service shows that, the world production of strawberry has been increased from 1961 to 2007 at 384.64% almost or nearly 2,902,142 tons and the cultivated area have been increased by 173.35% or approximately 163.185 hectares. However the total world production and acreage are estimated in 2007 as about 3,824,678 tons, 254,027 hectares, respectively.

The weight or sizes of strawberry fruit are depended on the number of Achenes. The fruit site on the inflorescence and the completion of pollination and fertilization (Gene *et al*, 1989) adding to the number of fruits. The application of gibberellic acid (GA3) showed different result on fruit weight, for example 75 ppm GA3 produce highest fruit set but has reduced the average fruit weight (Sharma and Singh, 2009). Meanwhile other study reported that the application of GA3 had improved the weight and size of strawberry fruits (Mohammad et al., 1990; Montero *et al*., 1998). In the other hand, average fruit weight was less affected by 0-15 ppm GA3 sprayed in strawberries *cv*. Sequoia, Chandlers, Douglas and Selva cultivars (D'Anna and Accardi, 1990).

Banday *et al*., (2005) reported that the application of 25 ppm GA3 at the flower bud formation stage in two strawberry cultivars 'Confitura' and 'Brighton' resulted in the maximum length, diameter and size of fruits , while application of 40 ppm GA3 were required had minimized the number of days to maturation. No influenced of the BA (benzyl adenine) on flowering and fruiting of strawberry. Kano and Asahira, (1978) suggested that the auxin is more essential than gibberellin and the carpel is the site of production of cytokinin which suppresses growth and ripening of the strawberry.

Study by Kirschbaum, (1998) on Sweet Charlie' *cv.* has found that the fruit weight was highest in the control plant without any significant with 6-BA, (6-BA + GA4+7) or IBA+KIN+GA and the lowest weight in GA3 only. Meanwhile the number of marketable fruits / plant was highest in IBA+KIN+GA treatment. It was that about 15.7 fruit /plant. Redchief *cv.* applied with BA at 0, 50, 125 or 250 ppm , had no effect on mean fruit weight (Archbold and Strang (1986).

The number of inflorescence may due to the number of bud conform to hold flower. Application of GA3 had increased the number of flower buds and open flowers (Paroussi et al 2002). However, the effect of GA3 was depended on the growth stage; GA3 applied about a month before the natural appearance of the flower buds hastened flowering (Honda, 1972). In addition application of GA3 led to reduce the time needed for inflorescence emergence (Paroussi et al., 2002). The number of inflorescences in three day-neutral strawberry cultivars (Summer Berry, Miyoshi and Enrai) were not affected by 50 ppm GA3, 50 ppm benzyladenine (BA) or 50 ppm GA3 + 50 ppm BA. The foliar spray of 50 mg/ L IAA , 50 mg/L 6-BA or both had increased the flower numbers (Shan *et al*., 2007). 6-BA + Ethephon was reported to decrease the number of flowers/plant , and a combination of 6-BA+GA4+7 produced the highest number of total flower / plant , while GA3 increased the number of the flower on the first month only, and the 6-BA only led to decrease it (Kirschbaum, 1998). Number of blossoms, in Raritan was increased by BA at 250 and 500 ppm on Raritan *cv.* (Weidman and Stang, 1983) .GA3 application in spring, after floral initiation increases the rate of production of open flowers (Tafazoli and Vince-Prue, 1978). Dale *et al*., (1996) found that the daughter plants derived from plants sprayed with growth regulators showed an increase of yield up to about 10%. Furthermore, gibberellic acid GA3 applied at 75 ppm has increased the harvesting period (Sharma and Singh, 2009).

D'Anna and Accardi,(1990) have observed that the cultivars Sequoia, Chandlers, Douglas and Selva applied with 0-15 ppm GA3 were highly sensitive to GA3 on the period of flower bud emission, floral stem elongation, the period of fruit ripeness and the total yields. Meanwhile the average weight of fruit was less affected by GA3. Gibberellin treatment led to a fall in the number of young fruit set per inflorescence in Festival'naya *cv.*, but caused no change on the character of Krasavitsa Zagor'ya *cv*. fruits (Agafonov *et al*., 1978). GA3 may decrease the yield significantly due to the production of malformed and button or nubbin berries, but the albinism incidence remained unaffected or reduced the day to harvesting (Sharma and Singh, 2009). The same result with different concentration was found by Mohammad *et al*., (1990) when applied with GA3 at 0, 10, 25, 50 or 100 ppm on strawberry *cv*. Murree, found that the longest productive period of strawberry was 41 day, which was obtained with 50 ppm GA3. The application of hormone significantly improved the growth, yield and quality of strawberry. NAA, in comparison with GA3 proved to be superior in improving yield and quality of strawberry, except vitamin C and acidity. Highest yield of 362.5 g plant was recorded with NAA at 15 ppm (Mir *et al*., 2004).

In the present study, the effect of exogenous hormone, on flowering and fruiting of strawberry Camarosa and Camarosa *cv*. was investigated

2. Materials and methods

2.1. Experimental plants

The experiment was conducted at the Malaysian Agricultural Research and Development Institute (MARDI) Station in Cameron Highland, Malaysia. Located altitude of 10,000 m above sea level, latitude 4°28'6.75"N and longitude 101°23'6.83"E. Two strawberry cultivars, Camarosa and Camaroga, were used. The uniform plantlets runner cuttings obtained from the adult plants were rooted in nursery. Rooting plantlets of 3.5 leaves stage were planted in 15 cm diameter plastic pots containing mixed growing media of coco peat and Perlite at 9:1 ratio. Three types of hormone, Auxin; indol-3-butyric acid (IBA) (Merck KGaA, Darmstadt, Germany)], cytokinin; N6-benzyladenine (6-BA) (R & M , UK)] and Giberralic acid (GA3) (Merck-Schuchardt CHG, 85662 Hohenbrunn, Germany)] each with two levels of concentrations, 0 and 50 ppm (Single or combination as total eight treatment) were treated once on the plants via foliage application. Each treatment contains three replications, each unit experiment containing 18 plants were involved. The experiment was arranged in a factorial randomized completed block design (RCBD) with exogenous hormone and cultivars as. All plants were placed under plastic rain-shelter, watered and fertilized through the fertigation system. Pest and disease control were made, and the temperature and humidity, were recorded daily by using Thermo-Hygrograph. All parameters: number of flower / inflorescence or per plant (No.F/inflo, No.F/plant) number of inflorescence /plant, number of fruits /plant, Fruit weight (g/fruit), percentage of fruit set (%), and number the crown and runner at the first opining flower were recorded.

2.2. Statistical analysis

The data were analyzed as a factorial RCBD with an exogenous hormone factor (IBA, GA3, and 6-BA) and genetic factors (cultivars) factor. Analysis of variance (ANOVA) was used to determine the significant difference between means. The Least significant difference (LSD) and Multiple Range Test of Duncan for the significant separation of means was calculated by using the PC program GinStat 12.

3. Result

The variation among the cultivars on flowering and fruiting of strawberry is shown in fig 1A. It demonstrated

that the number of flowers per inflorescence was significantly higher with an average of 18.31% in cultivar Camaroga compared to about 9.61 in cultivar Camarosa (Fig 1A). Flower production in Camaroga *cv*. was also significantly higher with an average of 11.12% compared to Camarosa *cv*. Camarosa *cv*. has produced a significantly higher fruit weight (g/ fruit) with approximately 10.9 % compared to about 4.39 g/fruit in Camaroga *cv*. Cultivar Camarosa was also surpassed the Camaroga in term of the percentage of fruits set per plant, where the average fruit set of about 53 % was found in Camarosa as compared to about 44.6 % found in Camarosa (Fig 1B). However, the result showed that there were not significant different among the cultivars on number of inflorescence/ plant, number of runner and crown per plant at the beginning first flower opening and number fruits per plant.

Table1 is showing the effect of exogenous hormone on the flowering and fruiting of strawberry. IBA treatment increased the number of the flower per inflorescence, but with no significant different with GA3, 6-BA, control or a combination of all regulators. Whereas GA3 treatment increased the number of inflorescences per plant with about 97.39% compared to the control. However no significant different were found between GA3 and 6-BA or IBA application. A combination of GA3 and 6-BA reduced the number of inflorescence per plant.

The highest number of flowers per plant was obtained from GA3 treatment, which an increased about 138% compared to the control plant. However, no significant different was found on the number of flowers per plant between the control and combination of 6-BA and other hormone.

A binary combination of IBA with 6-BA or GA3 had significantly increased the number of fruits per plant with an average of 160.9 % and 142.77 %, respectively, compared to the control. The fruit weight (g/fruit) had increased by about 33.85% and 24.8 % compared to the control when applied with GA3+6-BA and 6-BA alone, respectively. Whereas there were no significant different observed on the fruit weight between treatments of IAB, GA3+6BA+IBA and control.

The percentage of fruits set had increased by 97.24 % and 81.5 % compared to the control when applied with IBA and GA3, respectively. However, 6-BA and GA3+6BA+IBA treatments difference the percentage of fruits set compared with the fruits set in the control plant.

The effect of exogenous hormone on the type of fist bud initiation was shown in Table 1. Plants applied with IBA+6-BA produced inflorescence and flower before producing runners, while plants applied with GA3 produced the highest number of runner with an average of about 2.33 runners per plant before producing flower. However, no significant was found among GA3 treatment and control on the number of crown per plant.

In overall, the interaction of exogenous hormone × cultivar had significantly increased the flowering and fruiting of strawberry (Table2). A significant high average number of flowers per inflorescence with about 14.81 flowers were obtained from Camaroga × IBA (Table 2). Whereas, the high number of flowers per inflorescence found in Camarosa *cv*. was in plants applied with GA3 alone. However, the number of flowers per inflorescence was in both cultivars was the lowest when sprayed GA3+IBA or IBA+6-BA.

The interaction effect of exogenous hormone × cultivar has also shown that the number of flowers per plant in Camaroga *cv.* was superb than Camarosa *cv*. The average number of flowers per plant in Camaroga × GA3 was reached nearly 65.54 flowers per plant as compared to those in Camarosa *cv*. which about 45.14 flowers per plant when applied with IBA alone (Table 2). However, GA3+IBA reduced the number of the flowers in Camaroga *cv*. only.

The effect of exogenous hormone × cultivar interaction on the number of inflorescence per plant is shown in Table 2. It displayed that the most significant higher number of about 5.22 inflorescence per plant was produced by Camarosa *cv*. when sprayed with IBA+6-BA and in Camaroga *cv*. when was sprayed with GA3. However the number of inflorescence in Camarosa *cv*. treated with IBA were similar to those found in Camaroga *cv*. sprayed with GA3.

The number of fruits per plant had also increased significantly by using exogenous hormone (Table.2). The highest number of fruit of about 32.17 fruits per plant was obtained from Camarosa *cv*. treated with IBA+6-BA.But in Camaroga *cv*. when it was sprayed with GA3 alone with an average of about 23.33 fruits. However, Camarosa *cv*. sprayed with IBA had no significant difference to those Camaroga sprayed with GA3. Additionally, Camarosa *cv*. sprayed with GA3 produce similar number of fruits with those produce in Camaroga *cv*. sprayed with IBA+6-BA.

The highest average of fruit weight was about 7.30 g/fruit. It was obtained from Camarosa *cv*. sprayed with GA3+6BA. While in Camaroga *cv*. the highest weight was about 5.88 g/fruit when sprayed with 6-BA alone. However, the fruit of the control in Camarosa *cv*. had no significant different with Camaroga *cv* sprayed with 6-BA.

Exogenous hormone increased the percentage of fruits set in the both cultivars. The highest percentage was about 78.95 and 67.68 in Camarosa × IBA and Camaroga × GA3 respectively. However, the fruit of Camarosa × IBA+6-BA gave the similar percentage of fruit set with the Camaroga × GA3.The highest number of crown at

the beginning of first flower opening was obtained in Camaroga *cv* × GA3+IBA, where it was about two crowns per plant, but was no significant difference with Camarosa × IBA, and spraying with GA3+6-BA. The plants sprayed with GA3 produced the first flower earlier with about 2.3 runners per plant.

4. Discussion

The result of the present study clearly indicated that the GA3 had increased the number of inflorescence per plant, number of flowers per plant and number of flowers per inflorescence. However; GA3 plays an important role in promotion of the flowers (John *et al.,* 2008).

GA3 increases the lateral bud (Chesworth *et al* ,1998). This was observed via the plants treated with GA3 which produced about two runners before producing flowers. It was also found that GA3 increased the number of runners per plant (data not show). Acutely under the tropical condition, strawberry is not initiating the flowers bud during the autumn (the end of previous life-cycle) as like strawberry grown in colder area. But it is initiation and formation of the flowers at the same time. This may be able to explain an important of GA3 in initiating and increasing the flower of strawberry under the tropical condition.

However the combination of GA3 and IBA or 6-BA led to reduce the number of flowers per plant. It may be due to the antagonized effect of 6-BA and IBA to the role of GA3 on flower production. GA3 also increased the number of the flowers per plant. This may be due the increase of the number of inflorescence, where positive correlation was about 0.615 ($P>0.01$). IBA+6-BA treatment increased the number of fruits per plant. But reduce the average of fruits weight (g/fruit).This study is in an agreement with pervious reported by Paroussi et al.,(2002b),where the GA3 is not effect on the yield of Camarosa *cv*. only but in Camaroga *cv*. GA3 treatment was the best treatment increasing the number and fruit weight of strawberry

Percentage of fruit set was lower under the tropical condition. This may be due to the high humidity (above 90%) of the planting area. It may have effect on the pollen germination and additionally the raining weather reduces the insect and bees pollination activity. However, this percentage was largely enhanced when treated with IBA alone but with no significant different when treated with GA3 alone or in combination of IBA and other hormone. This may be related to the important role of auxin on fruiting of strawberry. Auxins in the pollen ensure that a rapid burst ovary growth, accompanied by abscission of the stamen and petals, usually follows after pollination, Auxins increased the fruit set in fruits containing many ovules (Chesworth *et al*., 1998).

Cytokinin and Auxin may be inhibited or antagonized the role of gibberellins. This was also found in this study via the negative significant on flowering parameter and number of runner per plant at the bingeing of first flower opening. But no effected with combination of cytokinin and gibberellins on fruit weight compared to those plants treated with GA3 and 6-BA alone. However the GA3 enhanced the 6-BA on percentage of fruit set per plant. However IBA enhanced the GA3 on the number of fruits per plant. These results are similar to those in tomato, where 6-BA has affected on GA3 in response to hypocotyls length, and GA3 effect on 6-BA in response to Anthocyanin production (Fleishon et al., 2011). As it was suggest Arabidopsis that cytokinin has no effect on gibberellin responses to the percentage of fruit set (Greenboim-Wainberg et al., 2005). Furthermore IBA inhabited the GA3 response on fruit set.

6-BA and IBA expressed antagonized effect to each other. BA and IBA have enhanced the number of fruits per plant on Camarosa *cv*. This might because the increase of percentage of fruit set per plant in Camarosa cultivar. Also it was found when the two hormones applied together increased the fructose level in Camarosa *cv.* (data not show).

The effects of PGR are cultivar-dependence which might be due to the genetic different of the two cultivars. Camarosa *cv*. gave a greater number of flowers per inflorescence when applied with IBA but Camarosa *cv*. gave the higher number when applied with GA3. While, Camarosa *cv*. treated with IBA+6-BA produced the higher number of fruits per plant and increased the percentage of fruit formation. Camaroga *cv*. was produced the higher number of fruits when treated with GA3 alone. This study could be explains the response of cultivars to the exogenous hormone that dependent on the required hormone by the plant and genotype factor it was related to the surrounding condition of the plant and hormone level or antagonistic interactions among of endogenous hormone (Gray, 2004).

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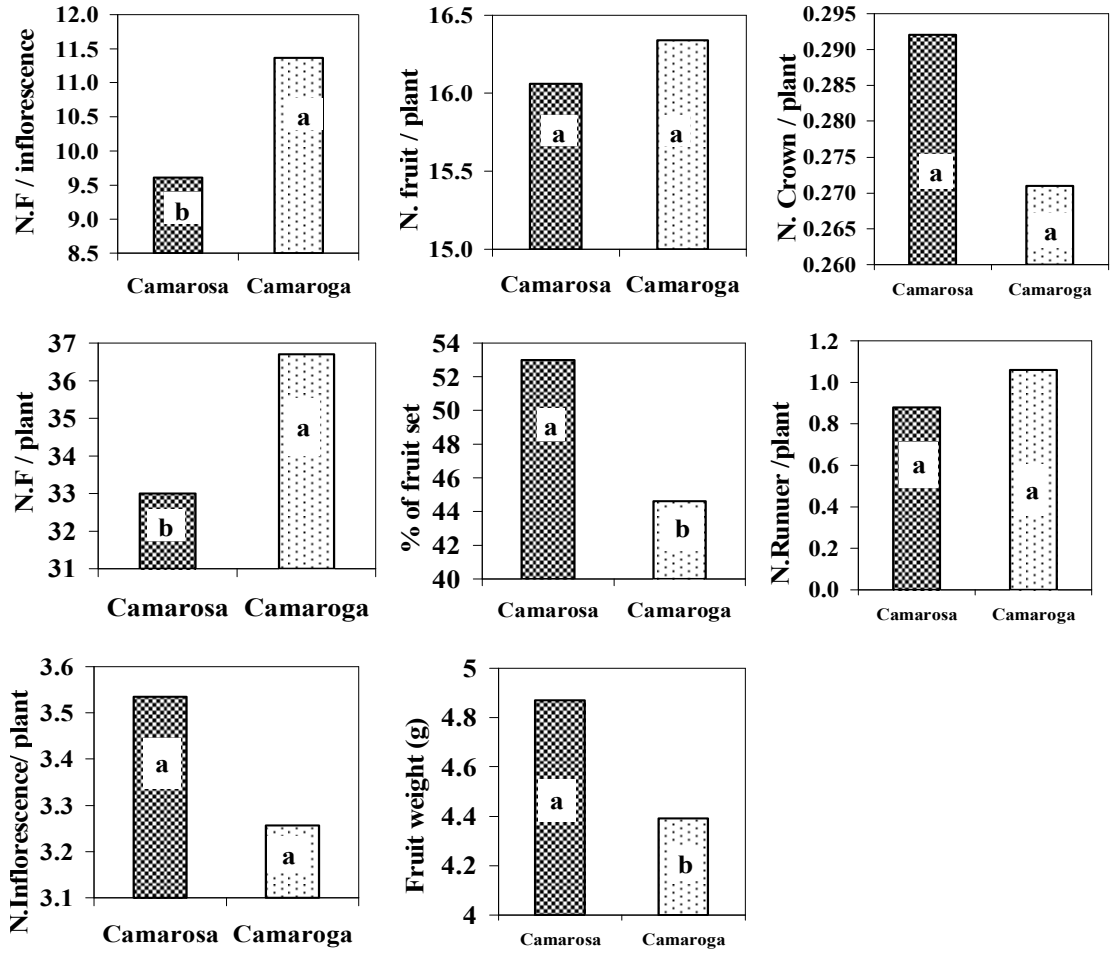

Figure 1 means compression of the effect of the strawberry cultivars on flowering and fruiting

Table 1 Means compression of the effect of exogenous hormone on the flowering and fruiting of strawberry

	No.F / inflo	N. fruit / plant	N. Crown / plant	Fruit weight (g)	N.F / plant	% fruit set	No. Runner /plant	N. Inflo / plant
GA3	11.93 a	17.67 ab	0.00 b	4.91 abc	54.87 a	58.06 a	2.33 a	4.167 a
6-BA	10.25 ab	15.44 bc	0.00 b	5.40 ab	38.19 bc	40.65 bc	0.33 bcd	3.722 ab
IBA	12.46 a	19.50 ab	0.50 ab	3.65 d	41.58 b	63.10 a	0.67 bcd	3.584 ab
GA3+6BA	10.49 ab	15.17 bc	0.25 ab	5.79 a	28.43 de	56.21 a	1.42 ab	2.722 cd
GA3+IBA	8.83 b	21.17 ab	1.00 a	4.39 bcd	32.10 cd	47.57 ab	1.50 ab	3.639 ab
IBA+6-BA	8.56 b	22.75 a	0.33 ab	4.60 bcd	28.42 de	52.88 ab	0.00 d	4.00 a
GA3+6BA+IBA	9.91 ab	9.17 cd	0.17 ab	3.93 cd	32.21 cd	39.73 bc	1.33 abc	3.222 bc
control	11.47 ab	8.72 d	0.00 b	4.33 cd	22.97 e	31.99 c	0.17 cd	2.111 d
L.S.D	**1.339**	**6.075**	**0.8165**	**0.961**	**7.557**	**14.24**	**1.123**	**0.6175**

Table 2 Means compression of the effect of the interaction between exogenous hormone and cultivars on the flowering and fruiting of strawberry

		Exogenous hormone								
Parameters	cultivars	GA3	6-BA	IBA	GA3+6BA	GA3+IBA	IBA+6-BA	GA3+6BA+IBA	control	L.S.D
N.F / inflo	Camaroga	11.22 abc	11.01 abc	14.81 a	11.33 abc	9.93 bc	9.74 bc	11.79 abc	11.11abc	3.787
	Camarosa	12.64 ab	9.49 bc	10.1 bc	9.65 bc	7.72 c	7.39 c	8.03 c	11.83 abc	
N. fruit / plant	Camaroga	23.33 abc	16.22bcde	15.33 cde	17.67 bcd	25.33 ab	13.33 de	10.83 de	8.67 de	8.591
	Camarosa	12.00 de	14.67 cde	23.67 abc	12.67 de	17.00 bcde	32.17 a	7.50 e	8.78 de	
N. Crown / plant	Camaroga	0.00 b	0.00 b	0.00 b	0.17 b	2.00 a	0.00 b	0.00 b	0.00 b	1.1547
	Camarosa	0.00 b	0.00 b	1.00 ab	0.33 b	0.00 b	0.67 b	0.33 b	0.00 b	
Fruit weight (g)	Camaroga	5.25 bc	5.88 ab	3.38 def	4.29 cde	4.44 bcde	4.35 bcde	4.76 bcd	2.75 f	1.359
	Camarosa	4.57 bcde	4.92 bcd	3.92 cdef	7.30 a	4.34 bcde	4.86 bcd	3.11 ef	5.91 b	
N.F / plant	Camaroga	65.54 a	40.40 bc	38.02 bcd	31.31 cdefg	37.29 bcde	18.81 h	39.33 bc	22.86 gh	10.688
	Camarosa	44.20 b	35.98bcdef	45.14 b	25.54 efgh	26.92 defgh	38.03 bcd	25.10 fgh	23.08 gh	
% of fruit set	Camaroga	67.68 ab	44.61 bcd	47.26 bcd	56.41 bc	55.53 bc	40.00 cd	28.02 de	17.17 e	20.14
	Camarosa	48.44 bcd	36.69 cde	78.95 a	56.00 bc	39.62 cd	65.76 ab	51.43 bc	46.82 bcd	
N. runner /plant	Camaroga	2.33 a	0.67 ab	1.00 ab	1.83 ab	1.33 ab	0.00 b	1.33 ab	0.00 b	1.588
	Camarosa	2.33 a	0.00 b	0.33 b	1.00 ab	1.67 ab	0.00 b	1.33 ab	0.33 b	
N. Inflo/ plant	Camaroga	4.833 a	3.667 bcde	2.612 fgh	2.778 cefgh	3.778 bcd	2.778 cdefgh	3.333 cdef	2.278 gh	0.8733
	Camarosa	3.5 cdef	3.778 bc	4.556 ab	2.667 efgh	3.5 cdef	5.222 a	3.111 cdefg	1.943 h