



## Influence of Some Biostimulants and Seaweed Extracts on Growth and Fruit Characteristics of Strawberry (*Fragaria x ananassa* Duch.)

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### Abstract

This study was conducted during (2013 -2014) season at Girda-Rash field, College of Agriculture, University of Salahaddin, Erbil governorate, Iraqi Kurdistan Region to study the effect of biostimulants as microbial inoculants (Fulzyme SP and Fulzyme Plus SP) and seaweed extracts as seamino on some vegetative growth and fruit characteristics of two short day cultivars of Strawberry 'Rubygem' and 'Florida Fortuna', the experimental units were arranged in split plot in a randomized complete block design with four replicates. The obtained results indicated that the 'Rubygem' cultivar was superior in the characteristics of leaf area and shoot dry weight, while no significant difference was obtained between the two cultivars in total chlorophyll content and root dry weight. On other hand 'Rubygem' cultivar recorded highest significant value in number of fruits per plant, fruit fresh weight and plant yield, however no significant difference was found between the two cultivars in fruit size. Fulzyme Plus SP application caused significant increase in the leaf area, total chlorophyll content, shoot dry weight and root dry weight, also application of Fulzyme Plus SP significantly increased the number of fruits per plant, fruit fresh weight, fruit size and plant yield, and the highest value in plant yield was recorded as a result of interaction effects of Rubygem x Fulzyme Plus SP.

### Introduction

Strawberry (*Fragaria x ananassa* Duch.) belongs to the family Rosaceae, the genus *Fragaria*, strawberry is known by its attractive and delicious fruit, which has an important role in human diet because of high antioxidants content. It is a rich source of vitamins and minerals with delicate flavors [1]. In 2012, the area cultivated with strawberry in the world reached (241,109 ha) representing a worldwide production of (4,516,810 tons) [2].

High yield with good quality of strawberry production depends on adequate mineral nutrition, weather terms and cultivars [3]. Cultivar choice is one of the most important factors for organic strawberry production as choosing the wrong cultivar inevitably leads to problems [4]. The strawberry 'Earlibrite' cultivar significantly overtopped in fruit fresh weight as compared with 'Sweet Charlie and 'Oso Grande' cultivars [5], also there were a wide variation among strawberry cultivars with respect to leaf area [6].

Recently researchers have started to give attention to the negative effects of using chemical fertilizers in environment and on human. Plant biostimulants are diverse substances and microorganisms used to enhance

plant growth [7], due to their effects which reduce fertilizers and other chemical compound application in agriculture, now, the biostimulants such as microbial inoculants and seaweed extracts use widely in agriculture [7] and [8].

The application of inoculants is seen as being very attractive since it would substantially reduce the use of chemical fertilizers and pesticides, and there are now an increasing number of inoculants being commercialized for various crops [9].

The presence of microorganisms in the soil is critical to the maintenance of soil function. Ability of the microorganisms to produce and release various metabolites affecting plant growth and health is considered one of the most important factors in soil [10]. Recent studies showed that a number of bacterial species mostly associated with plant rhizosphere, are found to be beneficial for plant growth, yield and fruit quality, they are called plant growth promoting rhizobacteria (PGPR). The PGPR have the potential to contribute in sustainable plant growth promotion [11]. PGPR as biofertilizers or microbial inoculants can be important components of an integrated nutrient management system [12].

Seaweed extracts (Seamino) increase plant resistance to pests and diseases, plant growth, yield and quality [13]. The application of seaweed extracts for different crops was a great importance due to containing high levels of organic matter, micro elements, vitamins and fatty acids and also rich in growth regulators such as auxins, cytokinins and gibberellins [14], also enhance plants resistance to diseases [15]. Foliar application of seaweed extracts as (soluamine) to strawberry plants caused significant increase in strawberry fruit size, number of fruits per plant and plant yield, while seaweed extracts as (marmarine) caused significant increase in fruit fresh weight [16]. Enzymes like protease, lipase, amylase, cellulase, lichenase and chitinase, enhance the biodegradation of macromolecules of the agricultural residues in the soil so that further microbial attack is speeded up [17].

The aim of this study was to investigate the effect of biostimulants as microbial inoculants (Fulzyme SP and Fulzyme Plus SP) and seaweed extracts as Seamino on some vegetative growth and yield characteristics of two short day strawberry cultivar 'Rubygem' and 'Florida Fortuna'.

### Materials and Methods

This study was conducted during (2013 -2014) season at the lath house of Girda-Rash field, College of Agriculture, University of Salahadin, Erbil governorate, Iraq which is located on the latitude ( $36^{\circ} 07' 14.36''$  N), and longitude ( $44^{\circ} 00' 48.33''$  E) and at an altitude of 410 m above the mean sea level. Soil of the experimental field was loam in texture having pH: 7.9, E.C.:  $0.15 \text{ dSm}^{-1}$ , organic material: 0.5%, total Nitrogen: 0.054%, available phosphorus:  $3.6 \text{ mg.kg}^{-1}$  and available potassium:  $96.5 \text{ mg.kg}^{-1}$ . Soil was thoroughly ploughed and manual raised beds with 0.75 m width and 0.2 m height were prepared, and 0.3 m distances were left between plants, all necessary cultural practices such as weed control, fertilizer application and surface irrigation were followed uniformly for all the plots and treatments during the study period. There were two factors 1. Two strawberry short day cultivars; 'Rubygem' and 'Florida Fortuna', 2.a- Fulzyme SP, which contain (*Bacillus subtilis* & *Pseudomonas putida*:  $2 \times 10^{10}$ / grams) and enzymes of (Protease, Amylase, Chitinase & Lipase) was applied to the soil at planting time at a rate of 1.0 L/ha and was repeated after five weeks, b- Fulzyme Plus SP, which contain (*Bacillus subtilis* & *Pseudomonas putida*:  $2 \times 10^{10}$ / grams), enzymes of (Protease, Amylase, Chitinase & Lipase) and growth regulators (0.3 % Gibberellin, & Cytokinin) was applied to the soil at planting time at a rate of 1.0 L/ha and was repeated after five weeks, c- Seaweed Extract as Seamino (Seaweed Extract plus amino acids, the composition is ( $\text{N} \geq 3\%$ ,  $\text{P}_2\text{O}_5 \geq 4\%$ ,  $\text{K}_2\text{O} \geq 8\%$ , amino acid  $\geq 10\%$ , seaweed extract  $\geq 18\%$ ) was sprayed to the plants at a rate of 10L/ha two times, before and after flowering respectively, d- Untreated plants as Control. Ten plants were planted in each experimental units on 2<sup>nd</sup> of October 2013.

### Measurements

Six plants were selected randomly from each experimental unit to measure the vegetative growth, yield and fruit characteristics:

### Vegetative growth characteristics

Four fully expanded leaves were taken randomly from each selected plants after the harvesting period, to determine the leaf area (cm<sup>2</sup>) through photographing the leaves on previously weighted A4 white paper then the photograph area was cut out weighed to obtain the corresponding area for each (g) of A4 paper then the leaf area [18].

$$\text{Leaf area (cm}^2\text{)} = \frac{\text{Area of A4 paper (cm}^2\text{) x cut part weighed of plant leaf (g)}}{\text{Wight of (A4) paper (g)}}$$

Total chlorophyll content was measured at the end of the harvesting period for ten leaves selected randomly from each of the selected plants using a chlorophyll meter (SPAD-502. Minolta Co. Ltd.) to find an average of total chlorophyll content. At the end of the experiment, shoot and root dry weights (g) were recorded for the selected plants after washing and oven drying at 70°C until the weight fixed.

### Yield and fruit characteristics

Fruits were harvested at mature red stage for the selected six plants in each experimental unit at 2–3 day intervals during the harvesting period (from 2nd April 2014 to 29th May, 2014), the average number of fruits were counted, and the fruits were weighed to determine the average fruit weights (g) , and plant yield was determined as an average of summing up total fruit weight(g) produced from the six selected plants, and the fruit size was measured by method of measuring water displacement in a scaled cylinder [19].

The Experimental units were arranged as split plot in a randomized complete block design with four replicates. Main plots were assigned to two strawberry cultivars and sub-plots were assigned to biostimulants as microbial inoculants (Fulzyme SP and Fulzyme Plus SP) and Seamino and control, the observations were taken and results were compiled for treatment comparison. The data were statistically analyzed with computer using SAS system (2005) and the significant differences between treatment means were tested with Duncun's Multiple Range Test (P≤0.05) [20].

## Results and discussion

### Vegetative growth characteristics

The data from table (1) show that the ‘Rubygem’ cultivar was superior in the characteristics of leaf area (68.17 cm<sup>2</sup>) and shoot dry weight (13.63 g) as compared with ‘Florid Fortuna’ cultivar, while no significant difference was obtained between the two cultivars in total chlorophyll content and root dry weight. The significant differences observed may be due to the genetic variability between the two cultivars [21] and [22]. These results are in agreement with those recorded by [23] and [24].

The data from the same table also show that the application of Fulzyme Plus SP caused significant increase in the total chlorophyll content (42.11 SPAD), at the same time the obtained data indicate that the application of Fulzyme SP and Fulzyme Plus SP resulted in increasing the leaf area, shoot dry weight and root dry weight but no significant differences were found between them.

The effect of Fulzyme Plus SP may be due to the accumulation effect of the Fulzyme SP Plus contents, which contains the PGPR as *Bacillus subtilis* and *Pseudomonas putida*, Enzymes and (0.3 % Gibberellin, & Cytokinin) . The PGPR can promote plant growth and development either directly and indirectly. Direct stimulation includes biological nitrogen fixation, producing phytohormones like auxins, cytokinins and gibberellins, solubilizing minerals like phosphorus and iron, production of siderophores and enzymes and induction of systemic resistance, while indirect stimulation is basically related to biocontrol, including antibiotic production, chelation of available Fe in the rhizosphere, synthesis of extracellular enzymes to hydrolyze the fungal cell wall and competition for niches within the rhizosphere [25-28].

The plant hormones regulate multiple physiological processes, including root initiation, root elongation, and root hair formation. Soil enzymes are involved in the catalysis of a large number of reactions necessary for life processes of microorganisms in soils, decomposition of organic residues, cycling of nutrients, and formation of organic matter and soil structure [29- 31].

The synergic effect of Fulzyme Plus SP contents might have helped in N-fixation and its quick release for plants absorption, which resulted in better root proliferation. The increasing of shoot dry weight might be due to increase leaf area and total chlorophyll content or may be attributed to a higher nutritional uptake mainly by greater expansion of root system due to increased supply of photosynthetic productions in the leaves , also, the reason for increased vegetative growth might be due to the production of plant growth regulators in the rhizosphere which are absorbed by the roots, these results were similar to studies on strawberry conducted by [32] and [33].

Data obtained from table (1) also indicate that the interaction effects of (Rubegem x Fulzyme SP) and (Rubygem x Fulzyme Plus SP) caused significant increase in leaf area and root dry weight, and the highest significant value for total chlorophyll content was recorded with the treatments of (Rubygem x Fulzyme Plus SP) and (Florida Fortuna x Fulzyme Plus SP), however the interaction effects of (Rubygem x Fulzyme SP, Fulzym Plus SP x Seamino, (Florida Fortuna x Fulzyme SP and Fulzyme Plus SP) were superior in shoot dry weight as compared with other interactions.

Table-1: Influence of microbial inoculants, seaweed extracts and their interactions on vegetative growth characteristics and root dry weight of strawberry cultivars ‘Rubygem’ and ‘Florida Fortuna’

	Leaf area (cm <sup>2</sup> )	Total chlorophyll content (SPAD)	Shoot dry weight (g)	Root dry weight (g)
<b><u>Cultivars</u></b>				
Rubygem	68.17 a	35.94 a	31.63 a	13.78 a
Florida Fortuna	58.91 b	35.94 a	27.85 b	12.29 a
<b><u>Biostimulants</u></b>				
Fulzyme SP	71.10 a	39.71 b	31.82 ab	14.76 a
Fulzyme Plus SP	71.55 a	42.11 a	32.73 a	15.34 a
Seamino	63.81 b	38.92 b	29.94 b	10.92 b
Control	49.68 c	23.00 c	24.46 b	11.13 b
<b><u>Interactions</u></b>				
Rubygem x Fulzyme SP	78.17 a	39.71 a	33.26 a	15.73 a
Rubygem x Fulzyme Plus SP	77.31 a	42.11 a	32.93 a	17.08 a
Rubygem x Seamino	68.72 b	38.92 a	33.53 a	9.71 b
Rubygem x Control	52.47 d	23.00 b	26.78 b	12.60 ab
Florida Fortuna x Fulzyme SP	64.04 bc	39.71 a	30.39 a	13.78 ab
Florida Fortuna x Fulzyme Plus SP	65.75 b	42.11 a	32.52 a	13.60 ab
Florida Fortuna x Seamino	58.86 c	38.92 a	26.35 b	12.13 ab
Florida Fortuna x Control	46.89 d	23.00 b	22.15 c	9.66 b

Means of each factor and their interactions followed with the same letters are not significantly different from each other according to Duncan's Multiple Range test (P≤0.05).

#### Yield and fruit characteristics

The results from table (2) clearly show that ‘Rubygem’ cultivar recorded highest significant value in number of fruits per plant (18.52), fruit fresh weight (9.75 g) and plant yield (180.57 g.plant<sup>-1</sup>), however no significant difference was found in the fruit size between the two cultivars. The significant differences observed may be due to the genetic variability between the two cultivars. There is substantial evidence that genetic factors control the growth, yield and fruit quality of strawberry [34]. These results are in conformity with the findings of [35] and [36]. The same table also illustrates that the application of Fulzyme Plus SP caused significant increase in number of fruits per plant (17.41), fruit fresh weight (10.18 g), fruit size (10.13

cm<sup>3</sup>) and plant yield (177.23 g.plant<sup>-1</sup>). The increasing of leaf area and total chlorophyll content (Table 1) might be helped to improve photosynthetic efficiency and to synthesis of total carbohydrates and proteins which are necessary in cell division and enlargement leading to improving the vegetative growth and nutritional status and maintaining a good balance between total carbohydrates and nitrogen which led to an increase fruit number per plant, translocation of assimilates to the fruits and increasing both cell division and enlargement which cause an increase of fruit fresh weight and fruit size which finally resulting in increasing the yield of strawberry plant, the similar results indicated that the number of fruits and plant yield for strawberry increased as a result of application of PGPR [37].

The results from table (2) show that no significant difference were obtained between interaction effects of (Rubygem x Seamino) and ( Rubygem x Fulzyme Plus SP) in number of fruits per plant which were (23.46 and 21.93) respectively, and the two treatments were superior as compared with other interactions, whereas the highest value in fruit fresh weight was recorded in the interaction of (Rubygem x Fulzyme SP) which was (11.12 g) and highest value in fruit size was recorded as effect of interaction of (Florida Fortuna x Fulzyme Plus SP) which was (10.80 cm<sup>3</sup>) while the interaction effect of (Rubygem x Fulzyme Plus SP) caused significant increase in the strawberry yield which was (227.63 g.plant<sup>-1</sup>). All the interaction effects of Cultivars, Fulzyme SP, Fulzyme Plus SP and Seamino significantly affected in growth and yield. The highest means of these parameters were found at the interactions between the highest levels of each factor, and this may be attributed to the synergism effect of cultivars, Fulzyme SP, Fulzyme Plus SP and Seamino as

mentioned above.

Table-2: Influence of microbial inoculants, seaweed extracts and their interactions on yield and fruit characteristics of strawberry cultivars ‘Rubygem’ and ‘Florida Fortuna’

	No. of fruits per plant	fruit fresh weight (g)	fruit size (cm <sup>3</sup> )	Plant yield (g)
<b><u>Cultivars</u></b>				
Rubygem	18.52 a	9.75 a	8.97 a	180.57 a
Florida Fortuna	10.61 b	8.86 b	9.07 a	94.00 b
<b><u>Biostimulants</u></b>				
Fulzyme SP	14.61 b	9.96 ab	9.39 b	145.52 ab
Fulzyme Plus SP	17.41 a	10.18 a	10.13 a	177.23 a
Seamino	11.59 c	9.14 d	8.90 b	105.93 b
Control	6.92 d	7.94 c	7.66 c	54.94 c
<b><u>Interactions</u></b>				
Rubygem x Fulzyme SP	18.06 ab	11.12 a	9.28 b	200.83 b
Rubygem x Fulzyme Plus SP	21.93 a	10.38 ab	9.46 b	227.63 a
Rubygem x Seamino	23.46 a	9.30 bcd	9.02 bc	218.21 ab
Rubygem x Control	11.35 bc	8.19 de	8.09 cd	92.96 d
Florida Fortuna x Fulzyme SP	11.19 de	8.81 cde	9.50 b	98.58 cd
Florida Fortuna x Fulzyme Plus SP	12.90 cd	9.98 abc	10.80 a	128.74 c
Florida Fortuna x Seamino	11.63 de	8.98 bcde	8.77 bc	104.44 cd
Florida Fortuna x Control	7.24 e	7.68 e	7.22 d	55.60 e

Means of each factor and their interactions followed with the same letters are not significantly different from each other according to Duncan's Multiple Range Test (P≤0.05).

## Conclusion

Based on the obtained results from this study, the ‘Rubygem’ cultivar was superior in leaf area and shoot dry weight characteristics, number of fruits per plants, fruit weight and plant yield as compared with ‘Florida Fortuna’ cultivar, also the result from this study was found that Fulzyme Plus SP caused significant increase of most of the vegetative growth characteristics and fruit characteristics as compared with Fulzyme SP and seaweed extracts as Seamino.

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