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Effect of Paclobutrazol and Sulfate Zinc on Vegetative Growth, Yield and Fruit Quality of Strawberry (*Fragaria* × *ananassa* Duch. cv. Camarosa)

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ABSTRACT

This study aimed to improve vegetative growth, yield and fruit quality of strawberry (Fragaria × ananassa Duch. cv. Camarosa) using different concentrations of zinc sulfate ($ZnSO_4$) and pacloboyrazol (PBZ). Zinc (Zn) is a necessary element for plants. The growth inhibitor PBZ is a triazole and protects plants against several environmental stresses. Plants were treated with four levels of $ZnSO_4$ (0, 50, 100 and 150 mg L^{-1}) and four levels of PBZ (0, 30, 60 and 90 mg L^{-1}). Signification difference was seen among the four levels of $ZnSO_4$ and four levels of PBZ on the growth, yield and quality of strawberry. The results showed that the foliar application of $ZnSO_4$ prior to flowering stage was recommended to increase fruit quality and yield of strawberry. The highest total soluble solid (T.S.S.) (8.30%) was obtained from interaction of 30 mg L^{-1} PBZ and 150 mg L^{-1} $ZnSO_4$. Leaf area can be maximized (42.20 cm) by the application of 90 mg L^{-1} PBZ and 100 mg L^{-1} $ZnSO_4$. The 60 mg L^{-1} PBZ produced the maximum leaf number (22.1). Also, 100 mg L^{-1} $ZnSO_4$ proved longest for length of petiole (8.80 cm). The treatment of 90 mg L^{-1} PBZ along with 150 mg L^{-1} $ZnSO_4$ had the greatest effect on the fruit set (87.10%). Maximum yield per plant (87.80 g), the number of fruit per plant (10.10) and the number of flower per plant (12.00) were obtained by treatment of 90 mg L^{-1} PBZ without $ZnSO_4$. Maximum and minimum fruit weight (9.50 and 8.20 g) was observed in treatments of 90 mg L^{-1} PBZ along with 150 mg L^{-1} $ZnSO_4$ and control, respectively. All yield and fruit quality traits in plants treated with different concentrations of PBZ and $ZnSO_4$ had higher value than those of the control plants.

Key words: fruit set, flower number, leaf area, T.S.S., yield
Abbreviations: PBZ-pacloboyrazol; $ZnSO_4$- zinc sulfate

INTRODUCTION

Strawberry (*Fragaria* x *ananassa* Duch. cv. Camarosa) is the most important cultivated berry in the world, its annual yield being 12-15 million kilograms and about 80% of the total yield of cultivated berries. The major part of the crop is sold on the fresh fruit market [28]. Camarosa is a vigorous, early, short-day cultivar [20]. Camarosa has a high yielding capacity and high quality of fruits [3]. The plants grown one year are never kept for a second fruiting season. On the other hand, strawberry is an annual crop, being planted every year after the plants are cultured for some months in the high elevation nurseries.

Triazole compounds are new synthetic plant growth regulators that act as anti gibberellins and known to inhibit shoot growth in plants [12]. Include the increased antioxidant potentials, inhibition of plant growth, decreased inter-node elongation, increased chlorophyll levels, thicker leaf tissue and increased root to shoot ratio [22]. Triazoles affect the isoprenoid pathway and alter the levels of certain plant hormones by inhibiting gibberellins synthesis, increasing cytokinin levels and reducing ethylene evolution [15]. Paclobutrazol (PBZ) or (2RS,3RS)-1-(4-dichlorophenyl)-4-dimethyl-2-(1,2,4-triazole-1-yl)pentan-3-ol), is a potent inhibitor of gibberellins biosynthesis [14]. It has also been reported that PBZ inhibits gibberellins biosynthesis [12, 23]. PBZ can be applied as an overall spray, as a soil drench or by way of trunk painting; better results have been achieved when used as a soil drench, reportedly effective in inducing flowering in apple and pear [35] and reducing stem elongation in apple [32] and citrus [2]. PBZ has been reported to protect plants against several environmental stresses, i.e. drought, low and high temperature, etc.[12, 23]. PBZ has biochemical effects on plants, such as detoxification of active oxygen [17] and increased levels of proline [21], antioxidants [29] and chlorophyll contents [11].

Zinc (Zn) is an important essential micronutrient for plant. Zn is an essential metal for normal plant growth and development, since it is a constituent of many enzymes and proteins organisms. Elevated concentrations of Zn exist in many agricultural soils from management practices including application of sewage sludge or animal manure and from mining activities, and this may represent a risk to environmental quality and sustainable food production [19]. Zn is the second most abundant transition metal after iron (Fe) and is involved in various biological processes in organisms [5]. The effects of Zn on plants have been reviewed [6]. Excess of Zn can also affect the uptake of other nutrient elements [4, 33]. Final production (quantity and quality) is one of the main characteristics that should be evaluated in studies concerning plant crops [25]. The present study was conducted to determine the effect of PBZ and $ZnSO_4$ on vegetative growth, yield and fruit quality of strawberry (*Fragaria* × *ananassa* Duch. cv. Camarosa)

MATERIALS AND METHODS

Plant Materials
The experiment was conducted during 2011 and 2012 on strawberry plants at a greenhouse located in Gorgan city, Golestan province, in the northern part of Iran (latitude 36°50´N, longitude 24°44´E). This work was carried out with strawberry cv. 'Camarosa'. Camarosa which well-adapted to the Golestan area, is a vigorous, early and short-day cultivar obtained from the Spanish public strawberry breeding program with a high yielding capacity and high quality of fruits [3]. This cultivar was prepared from the University of Golestan.

Cultivation Media Conditions
The plants were fertilized in early spring with McGeary organics general purpose fertilizer (N–P–K), which was formulated to meet the National Organic Program standards (containing steamed bone meal, feather meal, soybean meal, and compost). The water management was done by drip irrigation. Weed control was performed by hand weeding and tilling. Wood chips and dried straws were applied as mulch to conserve moisture and help control weeds. The conventionally grown strawberries were fertilized with 84 kg of actual nitrogen/hectare, 56 kg of actual phosphorus/hectare, and 56 kg of actual potassium/hectare in early April. An additional 28 kg of nitrogen/hectare were applied later.

Treatments
The interaction effect of PBZ and Zn on vegetative and sexual growth, yield and fruit quality in strawberry (*Fragaria* x *ananassa* Duch. cv. Camarosa) was evaluated. Plants were treated with four levels of $ZnSO_4$ (0, 50, 100 and 150 mg L^{-1}) and four levels of PBZ (0, 30, 60 and 90 mg L^{-1}). A complete nutrient solution (Zn and PBZ) was applied twice each 14 days. Control plants were watered without PBZ and Zn.

Measurements
At the end of the experiment and after application of treatments, five plants were randomly sampled for the measurement of growth parameters. The leaf area, the number of leaf and length of petiole were measured in each plant treatment. The areas of primary leaves were determined by an area meter (Crump Scientific Products, UK) [16]. Following flowering, the number of flower was counted in each plant treatment. Fruits were harvested early in the morning and rapidly transferred to the laboratory. Strawberries were sorted on the basis of size, color (70% full red color) and absence of physical damage, and were randomly divided into lots of ten fruit. Fruits were selected based on module of weight and computation, toward the get yield per plant. The numbers and weight of fruits per

plant computation were measured in each of the plants per treatment. Total fruit weight of each plant was separately measured and considered as yield. To evaluate fruit quality, total soluble solid (T.S.S.) were measured using a refractometer [10]. To obtain total fruit set, mean of fruit number were divided on flower number per plant.

Analytical Procedures
The study was carried out in a factorial experiment based on completely randomized block design in three replications. The experiment with 12 treatments was included with 5 shrubs allocated to block with three replications. Thus, the 192 shrubs were used.

Statistical Analysis
Data were subjected to analysis of variance in SPSS statistical software and means were compared by the F test at the 0.05 and 0.01 of probability level.

RESULTS

Effect of PBZ and Zn on Vegetative Growth
ANOVA analysis showed that the leaf number, length of petiole and leaf area were significantly affected by PBZ ($p<0.01$), Zn ($p<0.01$) and interaction of PBZ and Zn ($p<0.01$). The data obtained from the effect of PBZ and Zn on leaf number, length of petiole and leaf area is summarized in Table 2. The largest number of leaf (22.10) was found when the plantlets were treated with 60 mg L^{-1} PBZ without Zn (Table 2). This result was comparatively better than the leaf number of control (Table 2). The smallest number of leaf (16.90) was obtained in control plantlets (without PBZ and Zn) (Table 2). The use of PBZ was more effective as compared to Zn. PBZ alone was able to induce more leaf number. The number of leaf could be increased when PBZ was used in combination with Zn. In all mixture treatments of PBZ and Zn, least number of leaves was observed in treatments without PBZ (Table 2).

Table 2 shows that incubation of plantlets with 100 mg L^{-1} Zn without PBZ resulted in longest petiole (8.80 cm) (Table 2). Other concentrations of Zn caused increasing the length of petiole (8.60 cm for 50 mg L^{-1} Zn and 8.70 for 150 mg L^{-1} Zn) (Table 2). The shortest petiole (7.00 cm) was obtained in plantlets treated with 90 mg L^{-1} PBZ without Zn. The length of petioles incubated with different concentrations of Zn was longer than the petioles formed on plantlets incubated with different concentrations of PBZ (Table 2). Longer length of petiole (8.80, 8.70 and 8.60 cm) was obtained in plantlets grown on media without PBZ. Shortest length of petioles was calculated in plantlets treated with 90 mg L^{-1} PBZ, single and in combination to different concentration of Zn (Table 2). In all treatments of PBZ, with or without Zn, the length of petioles decreased as the concentration of PBZ increased (Table 2). Thus, the maximum and minimum length of petioles in each series of PBZ concentrations (0, 30, 60 and 90 mg L^{-1}) was seen in 0 and 90 mg L^{-1} PBZ, respectively.

Data presented in Table 2 shows that the highest and lowest leaf areas (42.20 and 37.80 cm, respectively) were obtained from plants grown on medium enriched with 90 mg L^{-1} PBZ along with 100 mg L^{-1} Zn and control, respectively. In most treatments, the combination of PBZ and Zn improved leaf area. The combination of 60 mg L^{-1} PBZ along with 150, 100 and 50 mg L^{-1} Zn with 41.60, 41.60 and 41.20 cm leaf areas were found to be the most suitable compounds (Table 2). In most treatments of PBZ, with or without Zn, the leaf area increased as the concentration of PBZ increased (Table 2).

Effect of PBZ and Zn on Yield and Fruit Quality
ANOVA analysis showed that the number of fruit per plant, average fruit weight, yield per plant, number of flower per plant, fruit set and T.S.S. were significantly affected by PBZ ($p<0.01$), Zn ($p<0.01$) and interaction of PBZ and Zn ($p<0.01$). Results obtained from the effect of PBZ and Zn on the number of fruit per plant, average fruit weight, yield per plant, number of flower per plant and fruit set is summarized in Table 3. The largest number of fruit per plant (10.10) was obtained in treatment of 90 mg L^{-1} PBZ without Zn (Table 3). This result was comparatively better than the number of fruit per control plants (Table 3). The use of PBZ was more effective as compared to Zn. PBZ alone was able to induce most fruit number. Thus, the least number of fruit per plant (6.60) was obtained in control plants (without PBZ and Zn) (Table 3). Current study showed that in all treatments of PBZ, with or without Zn, the number of fruit per plant increased as the concentration of PBZ increased (Table 3). The most suitable concentration of PBZ and Zn are 90 and 50 mg L^{-1}, respectively (Table 3). In most treatments, the combination of PBZ and Zn improved the number of fruit per plant. In all mixture treatments, minimum number of fruit was shown in plants grown on media without PBZ (Table 3).

The highest value of fruit weight (9.50 g) was obtained by using the 90 mg L^{-1} PBZ along with 150 mg L^{-1} Zn (maximum levels of PBZ and Zn) (Table 3). The lowest value of fruit weight (8.20 g) was obtained in control plantlets (without PBZ and Zn) (Table 3). Table 3 shows that the 60 mg L^{-1} PBZ is also a proper concentration for increasing fruit weight. Different concentrations of Zn had least positive effect on enhancing the fruit weight (Table 3).

The maximum yield per plant (87.80 g) was obtained in treatment of 90 mg L^{-1} PBZ without Zn (Table 3). This result was comparatively better than yield per control plants (Table 3). The use of PBZ was more effective as compared to Zn. PBZ alone was able to induce highest yield per plant. Thus, the minimum yield per plant (54.10 g) was obtained in control plants (without PBZ and Zn) (Table 3). Current study showed that in all treatments of PBZ, with or without Zn, plant yield increased as the concentration of PBZ increased (Table 3). The most suitable concentration of PBZ and Zn are 90 and 150 mg L^{-1}, respectively (Table 3). In most treatments, the combination of PBZ and Zn improved the number of fruit per plant. In all mixture treatments, minimum number of fruit was shown in plants grown on media without PBZ (Table 3).

The highest number of flower per plant (12.00) was measured in treatment of 90 mg L^{-1} PBZ without Zn (Table 3). This result was comparatively better than the number of flower per plant (Table 3). The use of PBZ was more effective as compared to Zn. PBZ alone was able to induce highest the number of flower per plant. The lowest number of flower per plant (8.20) was obtained in control plants (without PBZ and Zn) (Table 3). This study showed that in most treatments of PBZ, with or without Zn, the number of flower per plant increased as the concentration of PBZ increased (Table 3). In most cases, suitable concentration of PBZ for increasing the number of flower per plant is 90 mg L^{-1}, while the lack of Zn in media is more suitable (Table 3). In most treatments, the combination of PBZ and Zn improved the number of flower per plant. In more mixture treatments, minimum number of flower was shown in plants grown on media without PBZ (Table 3).

The present investigation demonstrated that all the tested concentrations of PBZ and Zn enhanced the fruit set (Table 3). However, the differences between these treatments were not noticeable, but between these treatments and control were noticeable. In most cases, PBZ produced more fruit set than the control (Table 3). Maximum 87.10% and minimum 76.20% fruit set were recorded in plants treated with 90 mg L^{-1} PBZ along with 150 mg L^{-1} Zn and control, respectively (Table 3).

The 150 mg L^{-1} Zn without PBZ and 90 mg L^{-1} PBZ without Zn was found maximum and minimum to induce 8.30 and 6.70% T.S.S. (Table 3). Among different concentrations of PBZ, 30 mg L^{-1} was better than the others concentrations. In most treatments, T.S.S. was decreased as PBZ concentration increased. Thus, the treatments without PBZ induced higher T.S.S. than the treatments containing PBZ (Table 3). Among different concentrations of Zn, 150 mg L^{-1} was better than the others concentrations. The role of Zn on increasing T.S.S. was more important than that of PBZ. Table 3 shows that in most treatments, concentrations of 0 and 30 mg L^{-1} PBZ may not induce different rate of T.S.S. (Table 3).

DISCUSSION

PBZ is widely applied as a plant retardant in plants, provides quantity and quality controls in several crops. The efficacy of PBZ on growth, flowering, fruiting and fruit quality has been reported earlier [30]. Our study showed that the application of 60 and 90 mg L^{-1} of PBZ increased the number and area of leaf, but decreased petiole length. Contrary to our findings, Shakeri et al. [31] has been reported that PBZ reduces the growth and the number of leaf in strawberry. PBZ also reduced vegetative growth of apple [18]. Vegetative growth components were reduced after annually application of PBZ in apples [18]. The reducing effect of PBZ on plant height has been observed in ornamental plants. Studies showed that PBZ could well be used to control the growth of *Dianthus caryophyllus* cv. Mondriaan and improve its commercial quality. These studies demonstrated that PBZ reduced plant height by 9.2-64.3%. PBZ significantly reduced foliage height compared with the control. Studies have shown that PBZ is needed to be applied annually to increase the number of flower and fruit yields in strawberry [31]. Yield efficiency values were consistent with the fruit yield values. Our studies revealed that PBZ (especially the 90 mg L^{-1}) strongly increased yield and sexual growth of strawberry. Our findings are in consistent with some other studies. Total yield of 'Golden Delicious' apples were increased after application of PBZ [34]. Studies of Yadava and Singh [36] showed that PBZ is needed to use annually to increase the yield of mango fruit. PBZ had highly significant effect on the number of flowers per plant. Contrary to our findings, fruit-size reduction in apples has been previously reported by many researchers [13]. PBZ used for growth has been reported to reduce fruit size [27]. Also, the heavy fruit load may have depleted carbohydrates reserves necessary for the following year's flower initiation, fruit set and/or initial growth of the tree. PBZ increased achenes per fruit of strawberry but decreased its yield [24]. Fruit weight increased as fruit number increased [31].

Current study revealed that the application of 100 mg L^{-1} of $ZnSO_4$ increased the length of petiole and leaf area. $ZnSO_4$ did not change the number of leaf, and the maximum number of leaf was observed in treatment without $ZnSO_4$. Contrary to our finding, some studies showed that $ZnSO_4$ increased the number of leaf in strawberry cv. Camarosa and Armore [7, 26]. Our study showed that none of $ZnSO_4$ concentrations increased quality traits of strawberry, alone except for T.S.S. 150 mg L^{-1} of $ZnSO_4$ increases fruit weight and fruit set providing the presence of PBZ. However, Abdollahi et al. [1] showed that $ZnSO_4$ increased fruit number, size and quality of strawberry. Dixi and Gamdagin [8] reported that a foliar spray of $ZnSO_4$ on March and April increased the size, T.S.S. and juice of oranges. Dobroluybsikii et al. [9] reported that the application of $ZnSO_4$ can increase T.S.S. in fruit of guava. This result confirms our finding.

Table 1: The main chemical properties of the growing medium

Type of soil	Potassium (ppm)	Phosphorus (ppm)	Nitrogen (ppm)	EC (×10)	pH	Sand (%)	Clay (%)	Lay (%)
Silt-loam	254	20	0.17	3.0	7.2	24	60	16

Table 2: Effects of PBZ and Zinc Sulfate on vegetative growth and quality of fruit ratios for strawberry

Treatments (mg L^{-1})	Leaf number	Length of petiole (cm)	Leaf area (cm)
Control	16.90ef	8.50ab	37.80f
(PBZ 30 + Zn 0)	18.20d	8.00b	39.20d
(PBZ 60 + Zn 0)	22.10a	7.40cd	41.10b
(PBZ 90 + Zn 0)	20.50bc	7.00d	40.60c
(PBZ 0 + Zn 50)	17.50e	8.60ab	38.60e
(PBZ 30 + Zn 50)	18.30d	8.20b	40.70c
(PBZ 60 + Zn 50)	18.70d	8.10b	41.20b
(PBZ 90 + Zn 50)	19.10c	7.80c	41.30b
(PBZ 0 + Zn 100)	18.70d	8.80a	38.80e
(PBZ 30 + Zn 100)	18.80d	8.30b	40.70c
(PBZ 60 + Zn 100)	19.30c	8.50ab	41.60b
(PBZ 90 + Zn 100)	20.10bc	8.10b	42.20a
(PBZ 0 + Zn 150)	19.60c	8.70a	39.40d
(PBZ 30 + Zn 150)	19.70c	8.50ab	40.20c
(PBZ 60 + Zn 150)	20.70bc	8.20b	41.60b
(PBZ 90 + Zn 150)	21.00b	8.10b	41.30b

In each column, means with the similar letters are not significantly different at 5% level of probability using F test

Table 3: Effects of PBZ and Zinc Sulfate on sexual growth and yield of strawberry

Treatments	Number of fruit/plant	Average fruit weight (g)	Yield/plant (g)	Number of flower/plant	Fruit set (%)	T.S.S. (%)
Control	6.60d	8.20c	54.10e	8.20d	76.20e	7.60c
(PBZ 30 + Zn 0)	7.90c	8.40c	68.00d	9.70c	82.40b	7.30d
(PBZ 60 + Zn 0)	8.90b	9.10a	80.90b	10.50bc	84.70b	7.10e
(PBZ 90 + Zn 0)	10.10a	8.70b	87.80a	12.00a	84.10b	6.70f
(PBZ 0 + Zn 50)	6.80d	8.30c	56.40e	8.30d	82.90b	7.80b
(PBZ 30 + Zn 50)	8.20bc	8.70b	71.30c	9.80c	83.60b	7.90b
(PBZ 60 + Zn 50)	9.00b	9.00b	80.10b	10.80b	84.20b	7.50c
(PBZ 90 + Zn 50)	9.10b	8.90b	81.90b	11.10b	81.00c	7.10e
(PBZ 0 + Zn 100)	7.40c	8.60b	63.60d	9.70c	79.50d	8.10ab
(PBZ 30 + Zn 100)	7.50c	8.90b	66.70d	9.10c	82.40b	8.10ab
(PBZ 60 + Zn 100)	7.80c	9.10b	70.20d	9.50c	83.10b	7.90b
(PBZ 90 + Zn 100)	7.90c	9.00b	71.80d	9.30c	83.80b	7.80b
(PBZ 0 + Zn 150)	7.30c	8.70b	63.50d	8.90cd	82.10b	8.30a
(PBZ 30 + Zn 150)	7.90c	8.80b	69.50d	9.30c	84.90b	8.20a
(PBZ 60 + Zn 150)	8.50b	9.10b	77.30c	10.10bc	83.30b	8.00ab
(PBZ 90 + Zn 150)	8.70b	9.50a	82.60b	10.20bc	87.10a	7.90b

In each column, means with the similar letters are not significantly different at 5% level of probability using F test

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