

Strawberry response *cv.* Albion at increasing doses of zinc

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Abstract

The Zn is a micronutrient that has an important influence on the growth, yield and quality of crops. The objective of the present work was to evaluate the effect of increasing doses of Zn in the nutrient solution on the growth, yield and quality of the fruit in strawberry *cv.* Albion. For which strawberry plants were established in 2.5 L pots with tezontle, in an open hydroponic system. The treatments were: control (without application of Zn), 1, 2.5, 5, 7.5, 10, 15, 20, 30 and 40 $\mu\text{mol L}^{-1}$ of Zn ($\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$) in the nutrient solution. The yield was increased 151, 161 and 134% with the doses of 5, 7.5 and 10 $\mu\text{mol L}^{-1}$, respectively. With a dose of 7.5 $\mu\text{mol L}^{-1}$, the biomass of the plant increased 60%, 93% the fresh weight of the fruits and 24% its firmness. With the doses from 2.5 to 15 $\mu\text{mol L}^{-1}$ the total soluble solids (SST), titratable acidity (AT), the SST/AT ratio and the concentration of total soluble sugars were increased. The best foliar concentration of Zn (sufficiency) (22 mg kg^{-1} of dry matter) was recorded with 7.5 $\mu\text{mol L}^{-1}$. Therefore, it is suggested to use Zn doses between 5 and 15 $\mu\text{mol L}^{-1}$ in the nutrient solution to satisfactorily cover the requirement of this cultivar.

Keywords: fruit quality, hydroponics, leaf concentration of Zn, yield.

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Introduction

The strawberry (*Fragaria x ananassa* Duch.) is grown in agricultural regions located from the Arctic to the tropics and is one of the strawberries with greater worldwide acceptance, as it is an important source of antioxidants, vitamins and minerals. In addition, it is one of the fruits with greater uses, among which stand out the consumption in fresh and the elaboration of industrialized products like jams, sweets, cakes and yogurts. From the socioeconomic point of view, it is very important for our country; because it generates employment opportunities and currencies, as it is an export product for fresh and frozen consumption (SIAP, 2014).

At a national level, strawberries are produced in 12 states of the country, with Baja California, Michoacán and Guanajuato, which account for 95% of national production (SIAP, 2014). Worldwide, Mexico ranks second in production with a volume of 360 426 t (FAO, 2014).

The cultivars that are managed in Mexico have been developed mainly in California. Among the most used are Festival, Albion, Camarosa, Sweet Charlie, Galexia, Camino Real, Aromas, Ventana and Diamante; that through several growing cycles have shown their efficiency in the field. The cv. Albion is the second most important crop, it was originated by the University of California, it is precocious and the producers classify it as good. Its fruit is large and of very good quality, both for export and for the national market. Presents neutral day plants, moderately vigorous with marked tolerance to adverse weather conditions. It is not very susceptible to some soil pathogens (*Verticillium* and *Phytophthora*) and moderately tolerant to ashtray (*Sphaerotheca macularis*) and red spider (Anónimo, 2015).

On the other hand, zinc is after iron, the most abundant metal in living organisms. Even when crops require it in very small quantities, it has been reported that 40% of the cultivated soils throughout the world are deficient micronutrients (Castellanos *et al.*, 2014). In plants this micronutrient plays a very important role in the synthesis of proteins; in carbohydrate metabolism there are several Zn-dependent enzymes; likewise, Zn could have an influence on the synthesis of auxins (Alcántar *et al.*, 2016). At least 2 800 proteins are dependent on zinc, either because it is part of its structure or because it is an activator of its function (Andreini *et al.*, 2009). It affects the expression and regulation of genes and defense mechanisms (Cakmak *et al.*, 2000). Zinc is the only metal that is present in all types of enzymes including oxidoreductases, transferases, hydrolases, lyases, isomerases and ligases (Broadley *et al.*, 2012).

In strawberry, the foliar concentration of zinc less than 20 mg kg⁻¹ of dry matter, was associated with symptoms of deficiency, abortion of the fruit and consequently low yield; the best growth and production were associated with a zinc concentration of 7.5 to 10 µmol L⁻¹ in the nutrient solution (Lieten, 2003). For the cv. Albion is not known the adequate dose of zinc in the nutrient solution; as well as the optimum concentration in the plant, because it is a commercial cultivar of recent introduction; which prevents generating an adequate fertilization program. That is why the present investigation was raised with the objectives of evaluating the effect of different doses of zinc in the nutritive solution on the development and yield of strawberry cv. Albion plants; as well as to determine the optimum dose in the solution that is related to the best yield and quality of the fruit.

Materials and methods

The study was carried out in a glass greenhouse of the Autonomous University Chapingo, located between 19° 20' north latitude and 98° 53' west longitude at 2 240 m altitude. The temperature and relative humidity were recorded during the crop cycle (from April to October 2014) with an RHT10 Extech Instruments® datalogger (Table 1).

Table 1. Temperature and relative humidity registered monthly in the day and night during strawberry cv. Albion cultivation.

Month	Temperature (°C)		Relative humidity (%)	
	Day	Night	Day	Night
April	31 ± 0.5	17 ± 0.2	30 ± 0.9	59 ± 0.7
May	30 ± 0.4	17 ± 0.2	40 ± 0.9	71 ± 0.6
June	29 ± 0.4	18 ± 0.2	49 ± 0.9	78 ± 0.5
July	29 ± 0.4	17 ± 0.2	47 ± 0.9	77 ± 0.5
August	30 ± 0.4	17 ± 0.2	45 ± 1.0	75 ± 0.5
September	28 ± 0.4	17 ± 0.2	48 ± 0.9	76 ± 0.5
October	28 ± 0.4	15 ± 0.2	47 ± 1	76 ± 0.5

The data is the average of 1 668 readings.

Strawberry cv. Albion plants from Michoacán; which were established in pots (2.5 L capacity) with red tezontle (2 mm diameter) in an open hydroponic system. For the elaboration of the nutritive solutions, the analysis of the tap water that was used was taken into account, as well as the balance of anions and cations considered in the ionic relations of Steiner (1984). The treatments consisted of increasing doses of Zn: 1, 2.5, 5, 7.5, 10, 15, 20, 30 and 40 $\mu\text{mol L}^{-1}$ and a control treatment (without application of Zn) and the rest of the nutrients were kept constant: 0.063 mL L^{-1} H_3PO_4 , 0.9 g L^{-1} $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$, 0.05 g L^{-1} $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 0.708 g L^{-1} KNO_3 , 0.05 $\text{Mg}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$, 19.7 mg L^{-1} Fe EDTA, 0.393 mg L^{-1} $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, 0.57 mg L^{-1} H_3BO_3 , 0.64 mg L^{-1} $(\text{NH}_4)_6\text{M}_2\text{O}_7 \cdot 24\text{H}_2\text{O}$, 1.8 mg L^{-1} $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$. For the contribution of Zn, $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ was used. The pH of the solutions was between 6.4 and 6.5; the CE (dS m^{-1}) was 2.4. The daily irrigation volume of the nutrient solution was 200 mL, with an approximate drainage of 30%.

The experimental design was completely randomized, consisting of 10 treatments with 10 repetitions per treatment. As an experimental unit, a plant was considered. The variables were evaluated at 7 months after the transplant and were the ones listed below.

Accumulation of biomass

The plants of each treatment were sectioned into leaves, crown and root, the dry weight of each structure was recorded on an OHAUS® digital scale model Scout Pro, after drying them in a BINDER® forced air oven at a temperature of 65 °C for 72 h or until constant weight.

Zinc concentration (Zn)

Five repetitions of each organ were formed, for which the corresponding organs of two plants were joined by treatment; which were subjected to a wet digestion with a mixture of sulfuric (H_2SO_4) and perchloric (HClO_4) acids in a ratio of 2:1 (v/v) and hydrogen peroxide (H_2O_2) to 30%, the concentration was determined by spectrometry atomic emission of induction with coupled plasma ICP model 725-ES of Agilent® (Alcántar and Sandoval, 1999).

Performance per plant

The total of fruits was harvested and weighed for each treatment and repetition. The fruits were harvested when they reached a bright red color.

The following fruit variables were evaluated in 15 fruits per treatment.

Fresh weight of the fruits

The weight was recorded on an Ohaus® digital scale model Scout Pro.

Roundness index

The length and equatorial diameter of 15 fruits per treatment were measured, using a General® digital vernier No. 143. The results of the relationship, which were obtained by dividing the length/diameter, were used to determine the shape of the fruits; where values less than one were considered oval, greater than one as elongated and values of one as round fruits (Martínez-Bolaños *et al.*, 2008).

Firmness of the fruit

Fifteen fruits used for the roundness index were evaluated in the same with a Qa Supplies® penetrometer model FT O2 with 2.3 mm thick tip at the height of the equatorial diameter. The units were transformed and the results were expressed in Newtons (N).

Total soluble solids (SST)

It was registered with a Atago® digital refractometer model PAL-1.

Titration acidity (AT)

The technique described by the AOAC was used (Horwitz, 1980). The percentage of acidity was calculated based on citric acid (meq= 0.064), which is the highest amount in strawberry fruits. With the SST and AT data the SST/AT ratio was obtained.

pH of the fruit

It was measured in the same extract used for the determination of titration acidity with a Conductronic® potentiometer model PC45.

Total soluble sugars (AST)

It was determined with the anthrone method described by Witham *et al.* (1971). The readings were made at 600 nm in a Thermo Spectronic® spectrophotometer model Genesys 10 UV. The concentration of sugars was estimated from a standard curve containing 0 to 250 µg of glucose mL⁻¹.

Statistic analysis

An analysis of variance and Tukey's means test ($p \leq 0.05$) were performed with the statistical program SAS version 9 (SAS Institute Inc., 2006). The graphics were made with SigmaPlot version 11.

Results and discussion

When comparing with the control (without Zn) it was observed that with the doses of 1 and 2.5 µmol L⁻¹ an increase in biomass accumulation of 40% was obtained, with 5 to 30 µmol L⁻¹ was increased between 50 and 60% and with the application of 40 µmol L⁻¹ the increase was 30% (Figure 1). With all the doses of Zn an increase of the biomass was obtained, although the greater accumulation was obtained with 7.5 and 10 µmol L⁻¹, whereas with higher doses it was decreasing. Lieten (2003) found similar results in strawberry cv. The santa cultivated in peat, where the best growth was obtained with doses of 7.5 to 10 µmol L⁻¹ of Zn in the nutrient solution; while doses above 30 µmol L⁻¹ were toxic, which led to symptoms of Fe deficiency, reduction in yield and increase in the number of deformed fruits. In our case, in the cv. Albion, with doses of 30 and 40 µmol L⁻¹ (Figure 1), no symptoms of Zn toxicity or Fe deficiency were observed; which indicates that the needs of Zn vary among cultivars.

The leaf was the organ that presented the highest accumulation of biomass, followed by the fruit, root and crown (Figure 1), due to its number, size and morphology in the plant. This trend of accumulation in the aerial part of the strawberry plant as Zn is supplemented in the nutrient solution, has also been observed in other crops such as wheat (Erenoglu *et al.*, 2011). The above is related to the role that Zn plays in growth; in such a way that small leaves are formed under deficient conditions, due to a reduction in cell expansion (Marschner, 2012).

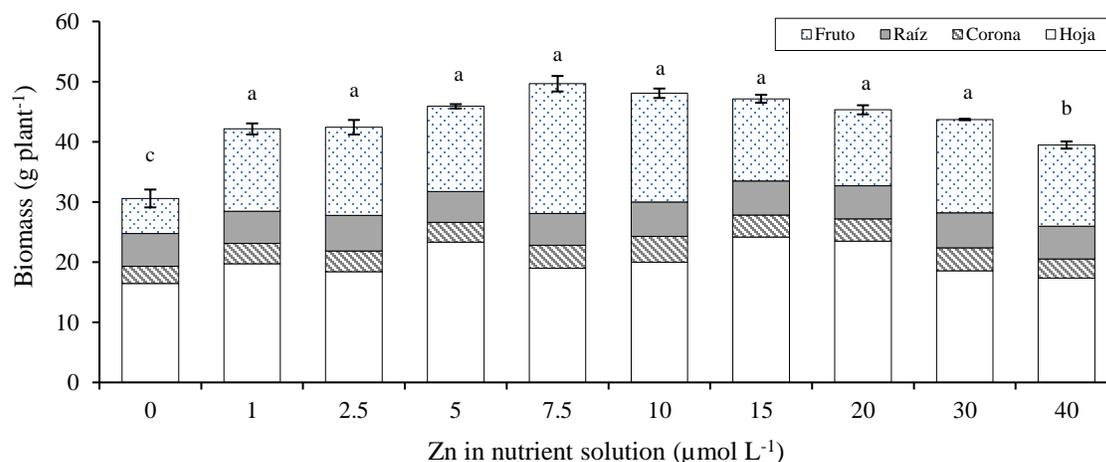


Figure 1. Effect of the dose of Zn in the nutritive solution on the biomass accumulated in strawberry cv. Albion. Means with the same letters are not statistically different (Tukey, $p \leq 0.05$).

The concentration of Zn in the leaves, crown and root increased as the Zn increased in the nutrient solution. In the leaves, it was observed that without the application of Zn its concentration was 10 mg kg⁻¹ of dry matter; whose origin comes from the impurities present in the fertilizers used for the elaboration of the nutritive solution. With the dose of 1 µmol L⁻¹ of Zn the concentration was 12 mg kg⁻¹ of dry matter and with 2.5 and 5 µmol was 13 mg kg⁻¹ of dry matter so the plant was in mild deficiency of the element, since the concentrations of sufficiency according to Hancock (1999) are between 20 and 50 mg kg⁻¹ of dry matter. With the rest of the doses of Zn in the nutrient solution, the concentration in the leaf was 15 to 22 mg kg⁻¹ of dry matter (Figure 2); with 7.5 µmol L⁻¹ the sufficiency (22 mg kg⁻¹ of dry matter) of Zn in the plant was had. However, the plants of the various treatments showed no symptoms of deficiency, which means that the cv. Albion has less demand for this micronutrient.

In the crown, without application and with the dose of 1 µmol L⁻¹, the values obtained were 10 and 12 mg kg⁻¹, respectively; while with the rest of the doses of Zn (2.5 to 40 µmol L⁻¹) the value ranged between 13 and 26 mg kg⁻¹ of dry matter. For the root, without application of Zn in the nutrient solution the value obtained was 14 mg kg⁻¹ and for the rest of the doses (1 to 40 µmol L⁻¹) the value was found between 20 and 116 mg kg⁻¹ of Zn (Figure 2). Of the three organs of the plant, the root presented the highest concentration of Zn.

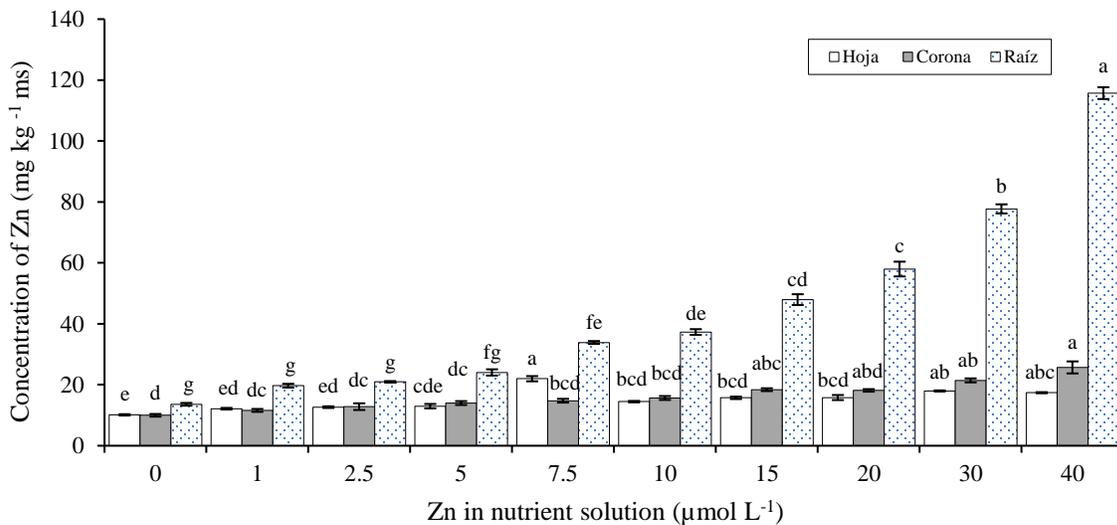


Figure 2. Effect of the dose of Zn in the nutrient solution on the concentration of this element in strawberry cv. Albion organs. Means with the same letters are not statistically different (Tukey, $p \leq 0.05$). ms= dry matter.

With the doses of Zn from 5 to 15 µmol L⁻¹ the highest yields were obtained, reaching a maximum of 86 g plant⁻¹ with the application of 7.5 µmol L⁻¹, which surpassed the control 61.73% (Figure 3). The application of Zn in the nutrient solution increased the yield, as observed by Lieten (2003) in strawberry cv. Elsanta plants, who obtained the best yields with doses of 5 to 15 µmol L⁻¹ of Zn, while with 30 µmol L⁻¹ the yield was significantly reduced. This same trend was observed in strawberry cv. Selva, where with the dose of 100 mg L⁻¹ of Zn, the yield was 54.17 g plant⁻¹, 4% higher than that of the control Abdollahi *et al.* (2010).

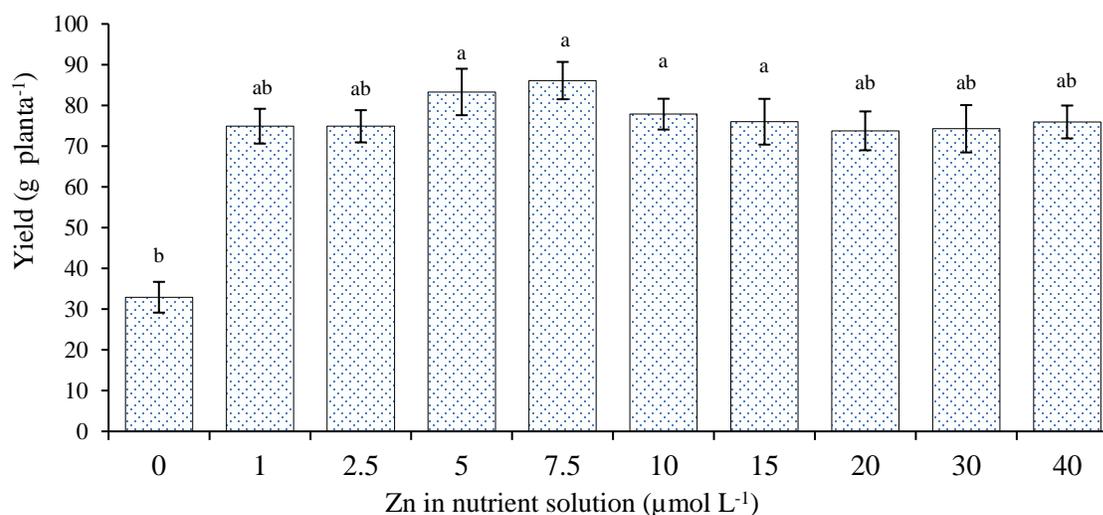


Figure 3. Effect of the dose of Zn in the nutritive solution on strawberry yield cv. Albion. Means with the same letters are not statistically different (Tukey, $p \leq 0.05$).

The application of Zn, in all the evaluated doses improved the weight of the fruit, the weight with the treatment without application of Zn in the nutritive solution, was reduced in approximately 50%, compared with the greater weight registered with the dose of $7.5 \mu\text{mol L}^{-1}$ (Table 2). These results coincide with Lieten (2003), who mentions that strawberry cv. Elsanta plants that received $0.5 \mu\text{mol L}^{-1}$ of Zn in the nutrient solution produced fruits of smaller weight and size.

The highest values recorded in the length of the fruit were observed with the doses of 2.5 to $40 \mu\text{mol L}^{-1}$ of Zn, while for the equatorial diameter there were no differences between the doses, but with respect to the control. The roundness index was between 1 and 1.22, which indicates that it is a round to slightly elongated fruit (Table 2). The length of the fruit was affected by the different doses of Zn in the nutrient solution, most likely due to the role of Zn in the synthesis of auxins, which control the growth of the receptacle and are synthesized by achenes (Abdollahi *et al.*, 2010).

According to the patent published by Shaw and Larson (2006) and works by Ornelas-Paz *et al.* (2013) for this cultivar, the equatorial length and diameter reported in this study are smaller, most likely because the phenotype of this cultivar can vary according to environmental and crop conditions. Some environmental factors that can affect fruit growth are temperature, light intensity and photoperiod (Hancock, 1999).

It is known that temperatures higher than 35°C in strawberry plants, cause the fruit and the plant to stop growing (Rowley *et al.*, 2011). Wang and Camp (2000) indicate that ideal day/night temperatures for optimal growth of strawberry cvs. Earliglow fruits and Kent should be $18/12^\circ\text{C}$. In the present experiment, average maximum temperatures of 39°C and day/night temperatures of up to $31/17^\circ\text{C}$ were recorded, respectively. So, the high temperatures recorded could affect the growth of the fruits.

Table 2. Effect of the dose of Zn in the nutritive solution on the variables of fruit quality in strawberry cv. Albion.

Zinc ($\mu\text{mol L}^{-1}$)	Fresh weight (g)	Length (mm)	Equatorial diameter (mm)	Roundness index (shape)	Firmness (N)
0	4.86 b ^z	20.5 d	20.5 b	1 d	1.35 b
1	7.07 ab	25.1 c	23.1 a	1.07 dc	1.41 ab
2.5	7.51 a	26.2 bc	24.1 a	1.09 bc	1.41 ab
5	8.37 a	26.7 bc	23.3 a	1.14 a	1.5 ab
7.5	9.36 a	29.6 ab	24.8 a	1.2 a	1.67 ab
10	8.45 a	27.4 abc	23.2 a	1.19 a	1.74 a
15	8.99 a	30.8 a	25.2 a	1.22 a	1.55 ab
20	7.58 a	26.8 bc	23.1 a	1.17 ab	1.49 ab
30	7.71 a	28.3 abc	23.5 a	1.2 a	1.4 ab
40	8 a	28.3 abc	23.9 a	1.18 a	1.35 b
DSH	2.55	3.45	2.42	0.08	0.36

^z= Means with the same letters are not statistically different (Tukey, $p \leq 0.05$). DSH= honest significant difference.

The Zn improved the firmness of the fruits; however, with the dose of $10 \mu\text{mol L}^{-1}$ the firmer fruits were obtained, while in the control and with $40 \mu\text{mol L}^{-1}$ the fruits were softer, with the same firmness value (Table 2).

The highest concentration of SST resulted with the application of $10 \mu\text{mol L}^{-1}$ and the lowest in the control (Table 3), this trend was also observed by Abdollahi *et al.* (2010) for strawberry cv. Selva plants, in which it was found that with the dose of 100mg L^{-1} of Zn the SST increased from 5 to 5.7% with respect to the control. Probably because Zn plays a very important role in the activity of enzymes that participate in photosynthesis and in the metabolism of carbohydrates; as well as in the transport of sugars, towards the demand points as the fruit (Marschner, 2012). The values of SST registered in the different treatments were within the optimum range, between 6 and 9%, reported by Hancock (1999) for red fruits.

With the applications of 7.5 to $40 \mu\text{mol L}^{-1}$, the AT oscillated between 0.8 and 0.95%, while in the rest of the applications it varied from 1 to 1.2% (Table 3); these data agree with the range of AT in the strawberry fruits proposed by Dale and Luby (1991), which is between 0.42 and 1.24%. The pH values found in the fruit with the doses of 5 to $40 \mu\text{mol L}^{-1}$ of Zn ranged between 3.03 and 3.45 (Table 3), values very close to the pH reported by Hancock (1999) as the average value, which is of 3.5.

Table 3. Effect of the dose of Zn in the nutritive solution on the chemical variables of the fruit in strawberry cv. Albion.

Zinc ($\mu\text{mol L}^{-1}$)	SST ($^{\circ}\text{Brix}$)	AT (%)	pH	SST/AT	AST (%)
0	5.94 b ^z	1.04 ab	2.76 f	5.84 c	4.19 d
1	6.88 ab	1 b	2.91 ef	6.86 bc	5.65 abc
2.5	6.92 ab	1.2 a	2.93 def	5.78 c	5.98 abc
5	7.4 ab	1.06 ab	3.03 de	7.05 bc	6.48 ab
7.5	7.66 ab	0.93 bc	3.05 cde	8.25 abc	6.59 a
10	8.3 a	0.95 bc	3.11 bcd	8.76 ab	6.5 ab
15	7.72 ab	0.8 c	3.25 b	9.65 a	6.29 abc
20	7.6 ab	0.94 bc	3.27 b	8.18 abc	5.63 abc
30	7.58 ab	0.94 bc	3.21 bc	8.05 abc	5.34 bcd
40	7.84 ab	0.93 bc	3.45 a	8.51 ab	5.26 cd
DSH	2.06	0.16	0.18	2.55	12.05

^z= Means with the same letters are not statistically different (Tukey, $p \leq 0.05$). SST: total soluble solids; AT= titratable acidity; SST/AT= total soluble solids/titratable acidity ratio; AST= total soluble sugars; DSH= honest significant difference.

With respect to the SST/AT ratio, from 5 $\mu\text{mol L}^{-1}$ of Zn the fruits were sweeter; while with low doses of this element the fruits were more acidic (Table 3). Keutgen and Pawelzik (2007) mention that a relation between 8.5 and 14 is considered an appropriate balance for strawberry flavor, and although in the present study some treatments were found outside this range, it is possible to observe that the Zn in the solution nutritive had a significant effect on the increase of this relationship; this element has also been shown to have an important role in the increase of sugars and decrease in acidity in the fruits (Abdollahi *et al.*, 2010).

The highest values of total soluble sugars (AST) were registered with doses of 5 to 15 $\mu\text{mol L}^{-1}$ of Zn, corresponding in part, with the treatments in which the highest concentration of SST was found. In the control and with higher doses of Zn (30 and 40 $\mu\text{mol L}^{-1}$), AST decreased (Table 3). The AST values in strawberry fruits were found in the range proposed by Dale and Luby (1991), which is from 4.1 to 6.6%.

Conclusions

Under the conditions in which the work was carried out, it was observed that all doses of Zn improved the characteristics of the crop; however, with the doses of Zn between 7.5 and 15 $\mu\text{mol L}^{-1}$ in the nutrient solution, the best accumulated biomass, yield and fruit quality were obtained; so, it is proposed to use this range of concentrations to cover the requirements of this element in strawberry cv. Albion plants, and thus improve its competitiveness and profitability.

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