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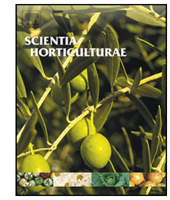

Effects of foliar applications of zinc and boron nano-fertilizers on pomegranate (*Punica granatum* cv. Ardestani) fruit yield and quality

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ABSTRACT

This study was carried out to assess the effects of the foliar application of nano-fertilizers of zinc (Zn) and boron (B) on pomegranate (*Punica granatum* cv. Ardestani) fruit yield and quality. A factorial experiment was conducted based on a completely randomized block design, with nine treatments and four replications per treatment. Foliar sprays of nano-Zn chelate fertiliser at three concentrations (0, 60 and 120 mg Zn L^{-1}) and nano-B chelate fertiliser (0, 3.25 and 6.5 mg B L^{-1}) were applied as a single spray before full bloom at a rate of 5.3 L $tree^{-1}$. The application of Zn and B increased the leaf concentrations of both microelements in August, reflecting the improvements in tree nutrient status. A single foliar spray with relatively low amounts of B or Zn nano-fertilizers (34 mg B $tree^{-1}$ or 636 mg Zn $tree^{-1}$, respectively) led to increases in pomegranate fruit yield, and this was mainly due to increases in the number of fruits per tree. The effect was not as large with Zn as with B. Fertilization with the highest of the two doses led to significant improvements in fruit quality, including 4.4–7.6% increases in TSS, 9.5–29.1% decreases in TA, 20.6–46.1% increases in maturity index and 0.28–0.62 pH unit increases in juice pH, whereas physical fruit characteristics were unaffected. Changes in total sugars and total phenolic compounds were only minor, whereas the antioxidant activity and total anthocyanins were unaffected.

1. Introduction

Pomegranate (*Punica granatum* L.) is a fruit tree of the *Punicaceae* family, which is mainly grown in subtropical and tropical regions (Adsule and Patil, 1995; Naik and Chand, 2011). Pomegranate fruits are consumed fresh or processed, and they are considered as a healthy food because of the high content of antioxidant compounds (Legua et al., 2012). In Iran, approximately 75% of pomegranate fruits are produced in five provinces, Fars, Khorasan, Markazi, Isfahan and Yazd (Varasteh et al., 2009), and pomegranate is consumed fresh as well as in processed forms for jams, jellies, syrups and juices (Kays, 1999; Alighourchi et al., 2008). Pomegranate is one of the most important commercial fruits in Iran: in 2015 the total area for pomegranate cultivation was 81,700 ha, and the total production was *ca.* 990,000 t (Anonymous, 2015).

Addition of fertilizers to supplement the natural soil fertility is essential for modern crop production, and precise management of nutrient elements is essential for a sustainable agriculture production (Barker and Pilbeam, 2006). In particular, microelements have important roles in fruit set and retention, as well as in fruit yield and quality (Singh and Ram, 1983; Khan et al., 1993). Zinc (Zn) is one of the essential micronutrients for plants, and Zn deficiency is common in many crops (Swietlik, 1999; Marschner, 2012; Ojeda-Barrios et al., 2014). Zinc is required for the activity of different enzymes, including dehydrogenases, aldolases, isomerases, transphosphorylases, RNA and DNA polymerases, and is also involved in the synthesis of tryptophan, cell division, maintenance of membrane structure and photosynthesis, and acts as a regulatory cofactor in protein synthesis (Marschner, 2012). Boron (B) deficiency is also a common micronutrient problem in agriculture, which results in yield reductions and impaired crop quality (Barker and Pilbeam, 2006). Boron roles in plants include effects on the germination of pollen grains, the elongation of pollen tube, fruit set and yield, and is also indirectly responsible for the activation of dehydrogenase enzymes, sugar translocation, nucleic acids and plant hormones (Brady and Weil, 1996; El-Sheikh et al., 2007; Marschner, 2012).

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Foliar sprays with fertilizers including microelements such as Zn, B, copper (Cu), manganese (Mn) and iron (Fe) have been shown to be convenient for field use, have a good effectiveness and very rapid plant response (Fernández et al., 2013). Also, foliar fertilizers help to avoid toxicity symptoms that may occur after soil application of the same microelements (Obreza et al., 2010). On the other hand, many problems in different fields of science and industry have been solved using nano-technologies (Scott and Chen, 2003), which are currently used for production, processing and application of nano-scale complexes. Materials that are smaller than 100 nm, at least in one dimension, are generally classified as nano-materials. Applications of this new technology have been found in agriculture, and nano-technologies are already applied to production, processing, storage, packing and transportation of agricultural products (Scott and Chen, 2003; Wiesner et al., 2006). The most important application of nanotechnology in agricultural crop production is the field of nano-fertilizers, which can feed plants gradually in a controlled manner, conversely to what occurs in the case of common fertilizers. These nano-fertilizers can be more efficient, decreasing soil pollution and other environmental risks that may occur when using chemical fertilizers (Naderi et al., 2011). One of the advantages of using nano-fertilizers is that application can be done in smaller amounts than when using common fertilizers (Selivanov and Zorin, 2001; Reynolds, 2002; Raikova et al., 2006; Batsmanova et al., 2013; Subramanian et al., 2015). Nano-materials can be used for designing new Zn fertilizers, with the solubility, diffusion and availability of Zn to plants being affected by the nano-particulate Zn characteristics (size, specific surface area and reactivity) (Subramanian and Sharmila Rahale, 2012; Mosanna and Khalilvand, 2015). For instance, positive effects of the application of nano-ZnO were reported on seed germination, seedling vigour, leaf chlorophyll content, stem and root growth in peanut (Prasad et al., 2012), and the positive effects of nano-ZnO contrasted with the negative effects on vegetable seed germination of a bulk form of ZnO (Singh et al., 2013).

The aim of this study was to test the effects of nano-Zn and nano-B fertilizers on the tree mineral nutrition status and fruit yield and quality in pomegranate trees grown in an important agricultural area in North Eastern Iran. In this area soils are sandy and with high pH, and therefore they are poor in nutrients. To the best of our knowledge, no studies have been conducted to investigate the effect of nano-Zn and nano-B fertilizers on pomegranate trees so far. The pomegranate cv. used, 'Ardestani', is of Iranian origin, late ripening and with rounded shape, red skin and aril, and of the sweet-sour type, and it is mostly used for fresh consumption.

2. Materials and methods

2.1. Experimental site, plant materials and treatments

The experiment was carried out in two seasons, 2014 and 2015, in a commercial pomegranate (*Punica granatum* L., cv. 'Ardestani') orchard. The orchard was located in the central part of the Razavi Khorasan province in North Eastern Iran (Tous Dasht; lat. 35°1′ 24.33″N, long. 58°50′19.61″E, altitude 967 m). The soil was a coarse-loamy over fragmental, mixed, thermic xeric Torriorthents (64% sand, 12% clay and 24% silt), with a pH of 8.08 in water and an EC of 9.4 dS m^{-1}. The region is arid, with 248 mm of annual mean precipitation and a mean annual temperature of 14.8 °C. Trees were eight-year-old, with three trunks and approximately 2.5–3 m in height. Trees were planted in regular rows, with a 3 × 5 m frame (667 trees ha^{-1}), irrigated by a drip irrigation system and treated in winter with NPK fertilizers at 150:150:150 g $tree^{-1}$. A factorial experiment was carried out based on completely randomized block design with nine fertilisation treatments and four replications per treatment. The two products used were "*Nano Zinc Chelate Fertiliser*" (120 g Zn L^{-1}) and "*Nano chelated fertiliser Boron*" (6.5 g B L^{-1}) (Sepher Parmis, Teheran, Iran), and both contain nanoparticles (composition patent-protected, average size 50 nm, range from 23 to 80 nm; http://www.sepehrparmis.com/fa_IR/Pages/Page/نانو). Fertilizers were used in combined spray applications at concentrations 0, 60 and 120 mg Zn L^{-1} (Zn0, Zn1, and Zn2, respectively; equivalent to 0.0, 0.9 and 1.8 mM Zn) and 0, 3.25 and 6.5 mg B L^{-1} (B0, B1, and B2, respectively; equivalent to 0.0, 0.3 and 0.6 mM B). The nine treatments were named Zn0 + B0 (control), Zn1 + B0, Zn2 + B0, Zn0 + B1, Zn1 + B1, Zn2 + B1, Zn0 + B2, Zn1 + B2 and Zn2 + B2. The fertiliser solution was prepared by diluting the commercial liquid product with well water available in the orchard. Trees were sprayed only once per season, one week before the first full bloom, on May 5th 2014, and April 19th 2015, with 5.3 L per tree (until full foliage wetting; total doses were 318 or 636 mg Zn $tree^{-1}$ and 17 or 34 mg B $tree^{-1}$). In pomegranate, flowering occurs in three waves (Holland et al., 2009), with the first one contributing most to fruit yield. Leaves were sampled from the middle part of fruiting shoots (100 leaves from the three trunks all around the canopy in each tree) on August 11th in the first season and on August 6th in the second season (approximately 90 days after full bloom). Fruits were harvested on October 22nd in the first season and on October 14th in the second season, and harvest date was based on general fruit appearance and fruit chemical properties (see below).

2.2. Plant measurements

2.2.1. Concentration of macro and micro-elements in leaves

The concentrations of macro and micronutrients in pomegranate leaves were measured in the Iranian laboratory in the first season and in the Spanish laboratory in the second season. In Iran, samples were prepared as in Chapman and Pratt (1961), and total N, P, K and Ca were determined using Kjeldhal method, spectrophotometry, flame emission photometry and complexometry, respectively; B was measured using azomethine H, and Fe, Zn and Mn concentrations were measured using atomic absorption spectrophotometry (AAS). In the Spanish laboratory samples were digested using a microwave device (Carrasco-Gil et al., 2016) and analysed for Ca, Mg using flame emission spectroscopy, for Fe, Mn, Cu and Zn using AAS and for B using ICP-OES. Since results from the two years were similar in spite of the different methodologies used, data for the two seasons were averaged.

2.2.2. Physical properties

In order to determine fruit physical properties, four fruits were selected randomly from each tree replication, and fruit weight was measured using an electronic balance. Fruit diameter, length, fruit calyx diameter (the widest part of the calyx) and peel thickness were measured using a digital Vernier gauge. In order to determine peel weight and aril percentage, fruits were manually peeled and the weight of total arils and peel were measured. The weight of 100 arils was measured and the juice volume of 100 g of arils, extracted by a manual extractor, was expressed in ml per 100 g of arils. All measurements were made according established methods (Tehranifar et al., 2010a,b; Hasani et al., 2012, 2016). For all physical parameters four replications per treatment and year were carried out.

2.2.3. Chemical properties

2.2.3.1. Titratable acidity, total soluble solids and juice pH. Titratable acidity (TA) was determined by the titration method (to pH 8.2 with 0.1 N NaOH), and results were expressed as percentage of citric acid. Total soluble solids (TSS, as%) and juice pH were measured at room temperature using a digital refractometer and a digital pH meter,

Table 1
Effects of nano-Zn and -B foliar fertilizers on leaf mineral composition (n = 3). Data shown are means of the two seasons, except for N.

Treatment	N[a] (%)	P (%)	K (%)	Ca (%)	Mg (%)	Fe (mg/kg)	Zn (mg/kg)	B (mg/kg)	Mn (mg/kg)	Cu (mg/kg)
Zn0 + B0	1.84 a	0.10 a	0.85 e	2.31 a	0.358 a	112.0 a	13.3 e	21.1 b	71.3 a	7.1 a
Zn1 + B0	1.87 a	0.10 a	0.89 cde	2.47 a	0.344 abcd	114.7 a	15.7 cde	21.3 b	70.2 a	7.0 a
Zn2 + B0	1.86 a	0.10 a	0.98 ab	2.44 a	0.323 cde	115.2 a	17.6 bc	21.7 b	66.2 a	6.5 a
Zn0 + B1	1.85 a	0.10 a	0.87 de	2.42 a	0.350 ab	116.8 a	14.7 de	22.3 b	70.1 a	7.0 a
Zn1 + B1	1.91 a	0.11 a	0.94 abc	2.49 a	0.346 abc	113.8 a	18.2 bc	23.0 b	66.6 a	6.7 a
Zn2 + B1	1.95 a	0.11 a	1.00 a	2.38 a	0.340 abcd	114.7 a	21.4 a	22.9 b	66.8 a	6.5 a
Zn0 + B2	1.85 a	0.11 a	0.91 bcd	2.39 a	0.320 de	110.0 a	16.4 cd	25.3 a	69.2 a	6.4 a
Zn1 + B2	1.88 a	0.11 a	0.96 ab	2.46 a	0.330 bcde	111.0 a	17.9 bc	25.0 a	68.0 a	6.9 a
Zn2 + B2	1.90 a	0.11 a	0.98 ab	2.40 a	0.311 e	106.8 a	19.6 ab	25.1 a	65.8 a	7.0 a

Zn0, Zn1 and Zn2 are 0, 60 and 120 mg Zn L^{-1}, and B0, B1 and B2 are 0, 3.25 and 6.5 mg B L^{-1}, respectively. Means with the same letter in each column were not significantly different using Duncan's multiple range test at $p < 0.05$.
[a] Data for N are only for the season 2014.

respectively. The TSS/TA ratio was used as a maturity index. Four replications per treatment and year were carried out.

2.2.3.2. Total phenols and antioxidant activity. To determine total phenols contents in juice, the Folin–Ciocalteu (FC) reagent method was used (Singleton and Rossi, 1965). Anti-oxidant activity was determined using 1,1-diphenyl-2-picrylhydrazyl (DPPH) (Brand-Williams et al., 1995). First, 100 μL of juice was diluted with 10 mL of methanol:water (6:4, v/v) (final ratio juice/diluted methanol 1:100). Then, the diluted juice was mixed with 2 mL of 0.1 mM DPPH in methanol, and the mixture was shaken and left in darkness and at room temperature for 15 min. The absorbance of the solution was measured at 515 nm using a spectrophotometer (Cecil Bio Quest, CE 2502, Cambridge, UK). The reaction mixture without DPPH was used for background correction. The antioxidant activity was calculated as: antioxidant activity (%) = $[1-(Abs_{sample}/Abs_{control})] \times 100$. Four replications per treatment and year were carried out.

2.2.3.3. Total anthocyanins and total soluble sugars. Total anthocyanins were estimated by the pH differential method using two buffer systems: 25 mM K chloride pH 1.0, and 0.4 M Na acetate pH 4.5 (Giusti and Wrolstad, 2001). Samples were diluted with K chloride buffer until the absorbance of the sample at 510 nm was within the linear range of the spectrophotometer (Cecil Bio Quest, CE 2502). The same dilution factor was later used to dilute the sample with the Na acetate buffer. Readings were performed at 510 nm and 700 nm in the two different buffers after 15 min of incubation, four times per sample. Total anthocyanin contents was calculated as follows: total anthocyanins = $[(A \times MW \times DF \times 100)/MA]$, where A = (A510–A700) pH4.5; MW: molecular weight (449.2); DF: dilution factor; MA: molar absorptive coefficient of cyanidin-3-glucoside (26.900). Results were expressed as mg cyanidin-3-glucoside 100 g^{-1} of juice. Four replications per treatment and year were carried out.

Total soluble sugar contents in pomegranate juice were determined using the anthrone reagent method (Dubois et al., 1951). A certain volume of pomegranate juice was diluted with distilled water, and then 0.1 mL of the mixture was added to four mL of anthrone (150 mg pure anthrone in 100 mL of H_2SO_4 72%). The sample was heated for 10 min in boiling water at 70 °C, and after cooling at room temperature absorbance was determined at 625 nm using a spectrophotometer. The total sugar contents was calculated using a glucose standard curve. Four replications per treatment and year were carried out.

2.3. Statistical analysis

The experimental design used was a randomized complete block design with four replications. Data were statistically evaluated by analysis of variance (ANOVA) to assess the significance of the main factors and the significance of interactions. Combined analysis of variance was carried out assuming environment (years and blocks) as random and treatments as fixed factors. Data were analysed using SAS (statistical analysis system; SAS Institute Inc., Cary, NC, U.S.A.) base 9 software, and means were compared using Duncan's multiple range test at the $p < 0.05$ level.

3. Results and discussion

The leaf concentrations of Zn and B in summer in untreated trees were 13.3 and 21.1 mg kg^{-1} DW, respectively (Table 1). Leaf concentrations below 15 mg Zn kg^{-1} DW are usually found in Zn deficient trees (Benton Jones et al., 1991; Ojeda-Barrios et al., 2014), and in pomegranate trees values of 12–20 mg Zn kg^{-1} DW have been reported as Zn-deficient in Iran (Khorsandi et al., 2009). Regarding B, concentrations in the range 28–37 mg kg^{-1} DW have been found in pomegranate trees grown in controlled conditions without B in the nutrient solution (Sarafi et al., 2013, 2014), whereas in B-deficient pomegranate trees leaf B concentrations in August were in the range 28–32 mg B kg^{-1} DW (Korkmaz and Askin, 2015). Therefore, the Zn and B leaf concentration values found in August in the studied orchard point out to the presence of a mixed deficiency of Zn and B. On the other hand, the leaf concentrations of N, P, K, Ca and Mg were 1.84, 0.10, 0.85, 2.31 and 0.36%, respectively, whereas those of Fe, Mn and Cu were 112.0, 71.3 and 7.1 mg kg^{-1} DW, respectively (Table 1). These concentrations are similar to those reported in pomegranate leaves in August by Korkmaz and Askin (2015), with the exception of Mg concentrations, which were somewhat lower.

3.1. Changes in leaf mineral concentrations with foliar fertilisation

A single Zn foliar spray increased the leaf concentration of Zn, from the untreated values of 13.3 Zn kg^{-1} DW to 15.7–18.2 mg Zn kg^{-1} DW (18.0–36.8%) with the lowest dose used (0.9 mM Zn), and to 17.6–21.4 mg Zn kg^{-1} DW (32.3–60.9%) with the highest dose (1.8 mM Zn) (Table 1). A single B foliar spray did not increase significantly the leaf concentration of B at the lowest dose used (0.3 mM B), with B concentrations remaining at 22.3–23.0 mg kg^{-1} DW, whereas with the highest dose used (0.6 mM B) the final concentrations of B were 25.0–25.3 mg kg^{-1} DW (a 18.4–19.9% increase). On the other hand, Zn and B fertilizers, alone or combined, had not significant effects on the leaf concentrations of N, P, Ca, Fe and Mn, whereas the concentration of K increased slightly in all treatments except in Zn1 + B0 and Zn0 + B1 (Table 1).

Previous studies have shown that B and Zn sprays generally increase the B and Zn concentrations in leaves, although in many of these studies several fertiliser applications and/or higher Zn/B concentrations have been used. For instance, B sprays increased the leaf

Table 2
Effects of nano-Zn and −B foliar fertilizers on pomegranate fruit yield, number of fruits per tree, fruit cracking, fruit diameter and length, fruit calyx diameter and fruit average weight. Data shown are means of the two years.

Treatment	Yield (kg per tree)	Number of fruits per tree	Fruit cracking	Fruit diameter (mm)	Fruit length (mm)	Fruit calyx diameter (mm)	Fruit average weight (g)
Zn0 + B0	13.8 e	50.6 d	3.1 a	75.5 abc	79.9 a	20.0 a	272.8 a
Zn1 + B0	14.3 de	52.7 cd	2.8 a	76.5 abc	77.8 a	20.9 a	272.2 a
Zn2 + B0	15.8 bc	57.6 bc	2.8 a	77.7 ab	77.9 a	20.2 a	274.5 a
Zn0 + B1	14.4 de	52.2 cd	2.9 a	73.2 c	80.1 a	20.4 a	274.9 a
Zn1 + B1	15.0 cd	51.3 d	2.6 a	78.2 ab	81.7 a	20.7 a	292.8 a
Zn2 + B1	16.2 b	58.7 b	2.5 a	76.6 abc	79.0 a	20.9 a	276.6 a
Zn0 + B2	18.0 a	64.4 a	2.6 a	74.2 bc	80.5 a	20.4 a	279.7 a
Zn1 + B2	18.5 a	65.9 a	2.5 a	74.9 abc	80.2 a	19.6 a	281.1 a
Zn2 + B2	18.4 a	63.0 ab	2.8 a	78.8 a	81.6 a	21.2 a	291.9 a
Significance							
Zn	**	*	NS	*	NS	NS	NS
B	**	**	NS	NS	NS	NS	NS
Zn*B	*	*	NS	NS	NS	NS	NS
year	**	NS	NS	**	**	**	**

Zn0, Zn1 and Zn2 are 0, 60 and 120 mg Zn L^{-1}, and B0, B1 and B2 are 0, 3.25 and 6.5 mg B L^{-1}, respectively. Means with the same letter in each column were not significantly different using Duncan's multiple range test at $p < 0.05$. *, ** and NS are significant at $p \leq 0.05$, at $p \leq 0.01$ and not significant, respectively.

B concentration in navel orange (Maurer and Taylor, 1999), olive (Perica et al., 2001) and sweet orange trees (Tariq et al., 2007), and Zn sprays increased the leaf concentration of Zn in pomegranate (Khorsandi et al., 2009; Hasani et al., 2012) and pecan nut trees (Ojeda-Barrios et al., 2014). In some cases, fertiliser applications have been reported to cause changes in the concentrations of other nutrients, possibly due to interactions in ion uptake and/or transport (Loneragan et al., 1982). For instance, in pomegranate, foliar applications of $ZnSO_4$ decreased the concentrations of Mn and P (Hasani et al., 2012), whereas in mandarin tree leaves the application of $ZnSO_4$, either alone or combined with H_3BO_3, increased leaf concentrations of Zn and K (Khan et al., 2012). In pistachio seedlings an antagonistic effect of Zn with Fe, Mn and Cu was observed, since the concentration of Fe was significantly larger in Zn-deficient seedlings, and soil application of Zn decreased Fe concentrations by 11–13% (Shahriaripour and Tajabadipour, 2010). The lack of interactions found in our study is likely due to the low doses of Zn and B applied.

3.2. Changes in fruit physical properties with fertilisation

3.2.1. Tree yield, number of fruits per tree and fruit cracking

Foliar spraying of Zn and B fertilizers, alone or combined, increased significantly fruit yield (by 8.6-34%, depending on the treatments), with the exception of the more diluted treatments Zn1 + B0 and Zn0 + B1 (Table 2). Both B and Zn fertilisation seem to have an effect on yield, but with B the effect was more intense. The highest yields (18.0-18.5 kg $tree^{-1}$) were obtained with the Zn0 + B2, Zn1 + B2 and Zn2 + B2 treatments, which led to 30.4–34.0% increases when compared with the control one (13.8 kg $tree^{-1}$). The application of Zn and B led to significant increases in the number of fruits per tree (by 13.8–30.2%, depending on the treatments), except in the diluted treatments Zn1 + B0, Zn0 + B1 and Zn1 + B1 (Table 2). Again, both B and Zn seem to have an effect on the number of fruits per tree, but with B the effect was more intense. The highest increases were found in the treatments Zn0 + B2, Zn1 + B2 and Zn2 + B2 treatments, which led to 24.5-30.2% increase in the number of fruits per tree (63.0–65.9 fruits $tree^{-1}$ compared with the control value of 50.6 fruits $tree^{-1}$). On the other hand, fruit cracking was not affected by the treatments.

Increases in fruit yield with B fertilisation have been reported in different fruit crops, including almond (Nyomora et al., 1997), apple (Wojcik et al., 1999), pear (Wojcik and Wojcik, 2003), persimmon (Khayyat et al., 2007) and peach (Yadav et al., 2013). The combined application of Zn and B increased walnut yield 4-fold (Keshavarz et al., 2011). Zinc sprays increased yield in apple (Amiri et al., 2008) and pistachio (Kizilgoz et al., 2010; Soliemanzadeh et al., 2013), and a positive correlation between Zn leaf concentration and pistachio yield (Kizilgoz et al., 2010) and a negative one between Zn leaf concentration and orange fruit drop (Garcia et al., 1984) have been reported. Increases in yield and number of fruits with Zn and B have been reported in almond (Pandit et al., 2011). Also, Zn and B fertilisation increased fruit set and the number of fruits per tree in different fruit tree species, including papaya (Jeyakumar et al., 2001), olive (Perica et al., 2001), cherry (Usenik and Stampar, 2007), palm (Sarrwy et al., 2012), and peach (Ali et al., 2014). The application of $ZnSO_4$, either alone or accompanied with BO_3H_3, increased the number of fruits in mandarin trees by 17 and 21%, respectively (Khan et al., 2012). Concerning the use of nano-fertilizers, foliar sprays with ZnO nano-particles led to 26–30% increases in peanut yield when compared with $ZnSO_4$ used at much higher doses (15-fold) in two different seasons (Prasad et al., 2012), and nano-chelate Zn has been shown to increase maize seed yield per plant almost 2-fold (Mosanna and Khalilvand, 2015).

The increased yield and number of fruits in pomegranate trees as the result of B foliar sprays are likely to be due to the known essential roles of B in fruit set, pollen grain germination and pollen tube elongation (Thompson and Batjer, 1950; Talaie et al., 2001; Wojcik and Wojcik, 2003; Baldi et al., 2004). Boron is rapidly absorbed by flowers (Sarrwy et al., 2012) and the B fertilisation effects can be attributed to its effect on fruit set and development or other metabolic processes such as carbohydrate transport, which are enhanced by its application (Peres and Reyes, 1983; Mengel and Kirkby, 2001; Marschner, 2012). The increased yield and number of fruits in pomegranate trees as the result of Zn foliar sprays are also in line with previous reports. Zinc acts in fruit bud formation and flowering (Swietlik, 1999; Usenik and Stampar, 2002), playing roles in the synthesis of tryptophan, an auxin precursor, and in the translocation of metabolites to the site of bud development or to the bud itself (Ryugo, 1988; Day, 1994).

3.2.2. Fruit size, peel thickness, arils and peel percentages, aril/peel ratio, weight of 100 arils and juice content of arils

Regarding fruit size physical parameters, fruit diameter, fruit length, fruit calyx diameter and fruit average weight were not affected significantly with the treatments when compared to the untreated controls (Table 2). However, the largest fruit diameter was obtained with the high concentrations of Zn and B (Zn2 + B2), with fruits being significantly larger (6.1–7.6%) than those obtained with the treatments Zn0 + B1 and Zn0 + B2. On the other hand, the

Table 3
Effects of nano-Zn and –B foliar fertilizers on pomegranate fruit aril and peel percentages, aril/peel ratio, weight of 100 arils, juice content of 100 g arils and peel thickness. Data shown are means of the two years.

Treatment	Total aril (%)	Total peel (%)	Aril/peel ratio	Weight of 100 arils (g)	Juice content of 100 g arils (ml)	Peel thickness (mm)
Zn0 + B0	57.6 a	42.4 a	1.36 a	36.6 a	62.1 a	2.44 a
Zn1 + B0	57.0 a	43.0 a	1.32 a	36.7 a	62.3 a	2.51 a
Zn2 + B0	56.0 a	44.0 a	1.27 a	37.6 a	63.1 a	2.56 a
Zn0 + B1	57.7a	42.3 a	1.36 a	36.6 a	62.6 a	2.41 a
Zn1 + B1	56.0 a	44.0 a	1.27 a	37.2 a	63.3 a	2.57 a
Zn2 + B1	55.9 a	44.1 a	1.26 a	38.8 a	62.6 a	2.57 a
Zn0 + B2	56.2 a	43.8 a	1.28 a	36.6 a	62.1 a	2.51 a
Zn1 + B2	55.9 a	44.1 a	1.26 a	37.3 a	62.9 a	2.61 a
Zn2 + B2	55.5 a	44.5 a	1.24 a	37.7 a	62.9 a	2.64 a
Significance						
Zn	NS	NS	NS	NS	NS	NS
B	NS	NS	NS	NS	NS	NS
Zn*B	NS	NS	NS	NS	NS	NS
year	NS	NS	NS	NS	NS	NS

Zn0, Zn1 and Zn2 are 0, 60 and 120 mg Zn L^{-1}, and B0, B1 and B2 are 0, 3.25 and 6.5 mg B L^{-1}, respectively. Means with the same letter in each column were not significantly different using Duncan's multiple range test at $p < 0.05$. *, ** and NS are significant at $p \leq 0.05$, at $p \leq 0.01$ and not significant, respectively.

application of Zn and B did not affect significantly the aril and peel percentages, aril/peel ratio, weight of 100 arils, juice content of arils and peel thickness (Table 3).

In some cases, effects on fruit size have been observed with Zn and B applications. The size of olive fruits has been reported to increase significantly when trees were treated by foliar applications of Zn alone or with GA3 (Ramezani and Shekafandeh, 2009), and foliar sprays of $ZnSO_4$ and BO_3H_3 increased average fruit weight and juice weight percentage in mandarin (Khan et al., 2012). Increases in fruit diameter have been attributed to the role of Zn on the synthesis of tryptophan (Sahota and Arora, 1981) and auxin (Alloway, 2008; Boettcher et al., 2010), therefore affecting cell elongation and division.

3.3. Fruit chemical properties

3.3.1. Juice pH, total soluble solids (TSS), titratable acidity (TA) and maturity index (TSS/TA ratio)

Pomegranate juice pH increased significantly (by 0.28–0.62 pH units, depending on the treatment) upon Zn and B fertilisation, except in the case of the more diluted treatments Zn1 + B0 and Zn0 + B1 (Table 4). Also, the more concentrated B and Zn treatments increased TSS in juice (4.4–7.6%), with the highest and lowest TSS values (17.06 and 15.85%, respectively) being observed in trees treated by the highest concentrations of Zn and B (Zn2 + B2) and the untreated controls, respectively (Table 4). Regarding TA, all treatments with the exception of Zn1 + B0 showed values lower than the controls (9.5–29.1% decreases, depending on the treatments), with the lowest one being for the treatment Zn1 + B2 (Table 4). As a result, B and Zn fertilisation markedly increased the maturity index (TSS/TA ratio) (by 20.6–46.1%, depending on the treatment), due to the increases in TSS and decreases in TA (Table 4). The highest increase in the maturity index was obtained in the trees sprayed with the treatment Zn2 + B2, followed by the treatments Zn2 + B1 and Zn1 + B2.

Results found are in line with the increased TSS and decreased TA with foliar application of $ZnSO_4$ in pomegranate trees (Hasani et al., 2012), and with the increases in maturity index in date palm with BO_3H_3 and $Ca(NO_3)_2$ applications, especially when both products were applied together (Sarrwy et al., 2012). However, Zn foliar application has been reported to increase fruit TA in pomegranate (El-Khawaga, 2007), and to have no effects on the TSS/TA ratio when applied alone or combination with N in sweet orange (Sahota and Arora, 1981). Increases in TSS in apple fruits with Zn fertilisation have been previously attributed to Zn effects on carbohydrate synthesis and translocation (Yogeratnam and Greenham, 1982).

3.3.2. Total phenolic compounds and antioxidant activity

Results show that only the more concentrated B foliar treatments (Zn0 + B2, Zn1 + B2 and Zn2 + B2) caused small (1%) but statistically significant changes in the amount of total phenolic compounds in pomegranate juice, whereas the antioxidant activity was not affected with any of the treatments (Table 4). The highest concentration of total phenolic compounds (409.92 mg 100 g^{-1} fresh weight) was found with the Zn2 + B2 treatment, whereas the lowest one (406.64 mg 100 g^{-1} fresh weight) occurred in fruits of the control treatment.

The total phenolic compounds parameter is considered as one of the most important nutritional value parameters in pomegranate fruits (Tehranifar et al., 2010a, 2010b). Similar high phenolic compounds concentrations have been found in Iranian pomegranate varieties (300–1000 mg 100 g^{-1} fresh weight; Tehranifar et al., 2010b), whereas Moroccan varieties contained lower phenolic concentrations (<200 mg 100 g^{-1} fresh weight; Legua et al., 2012). Many investigators have observed a positive effect of Zn and B on total phenolic contents in fruits. For instance, foliar sprays of B and Zn increased total phenolic compounds in olive (Saadati et al., 2013), foliar applications of Zn and B increased the total phenolics in pungent pepper (Manas et al., 2014), and Zn foliar sprays increased phenolics contents in grape berries (Song et al., 2015). The latter authors indicate that the increase was due to an increased expression of genes responsible to phenolic compounds biosynthesis. Also, the known role of B in the metabolism of phenolic compounds (Dear and Aronoff, 1965) has been confirmed in olive trees (Liakopoulos and Karabourniotis, 2005).

3.3.3. Total sugars and anthocyanins

The amount of total sugars in pomegranate fruit juice increased significantly (by 1.9–4.6%) only in the case of the combined Zn and B treatments (Zn1 + B1, Zn2 + B1, Zn1 + B2 and Zn2 + B2), whereas there was no significant effect in total anthocyanins with any of the treatments (Table 4). The highest content of total sugars (14.93 g 100 g^{-1} fresh weight) was obtained in trees treated with the highest concentration of Zn and B (Zn2 + B2), whereas the lowest one (14.26 g 100 g^{-1} fresh weight) was observed in the control untreated trees.

Increases in total sugars were found in papaya, mandarin and mango fruits after foliar applications of Zn and B (Sing et al., 2002; Babu and Yadav, 2005; Anees et al., 2011). The effect of Zn on total sugars can be due to its role in the starch and nucleic acid metabolism, and also to the activities of various enzymes involved in these biochemical reactions (Alloway, 2008), whereas the effects of B on total sugars can be ascribed to the roles of B in transport of

Table 4
Effects of nano-Zn and -B foliar fertilizers on pomegranate fruit juice pH, TSS, TA, maturity index, total phenols, antioxidant activity, total sugars and total anthocyanins. Data shown are means of the two years.

Treatment	Juice pH	TSS (%)	TA (%)	Maturity index (TSS/TA ratio)	Total phenols (mg 100 g^{-1} FW)	Antioxidant activity (%)	Total sugars (g 100 g^{-1} FW)	Total anthocyanins (mg 100 g^{-1} FW)
Zn0 + B0	3.42 e	15.85 d	1.89 a	8.49 c	406.64 e	23.88 a	14.26 d	7.69 a
Zn1 + B0	3.55 de	15.97 d	1.81 ab	8.85 c	406.92 de	24.17 a	14.28 d	7.76 a
Zn2 + B0	3.70 cd	16.30 cd	1.59 c	10.24 b	408.09 bcde	25.72 a	14.43 bcd	8.20 a
Zn0 + B1	3.53 de	15.96 d	1.71 bc	9.43 bc	407.74 cde	24.3 a	14.37 cd	8.01 a
Zn1 + B1	3.73 c	16.26 cd	1.43 d	11.51 a	407.56 cde	24.98 a	14.54 bc	7.86 a
Zn2 + B1	4.04 a	16.96 ab	1.37 d	12.37 a	408.60 bcde	26.41 a	14.63 b	8.66 a
Zn0 + B2	3.83 bc	16.14 cd	1.39 d	11.71 a	408.77 abc	26.11 a	14.43 bcd	8.51 a
Zn1 + B2	3.99 ab	16.56 bc	1.34 d	12.34 a	409.48 ab	26.72 a	14.60 bc	8.72 a
Zn2 + B2	3.98 ab	17.06 a	1.37 d	12.41 a	409.92 a	29.48 a	14.93 a	8.68 a
Significance								
Zn	**	**	**	**	*	NS	**	NS
B	**	**	**	**	**	NS	**	NS
Zn*B	*	NS	*	*	NS	NS	NS	NS
year	NS	NS	NS	NS	NS	NS	NS	NS

Zn0, Zn1 and Zn2 are 0, 60 and 120 mg Zn L^{-1}, and B0, B1 and B2 are 0, 3.25 and 6.5 mg B L^{-1}, respectively. Means with the same letter in each column were not significantly different using Duncan's multiple range test at $p < 0.05$. *, ** and NS are significant at $p \leq 0.05$, at $p \leq 0.01$ and not significant, respectively.
FW: fresh weight.

sugars and carbohydrate metabolism (Hansch and Mendel, 2009). When the effect of foliar sprays of some growth regulators and mineral nutrients were studied in pomegranate, it was found that Zn decreased total anthocyanins in the first season, with the lowest values being obtained with 0.3% Zn (Khalil and Aly, 2013).

4. Conclusion

Results obtained indicate that a single foliar spray with relatively low amounts of B or Zn nano-fertilizers (34 mg B tree^{-1} or 636 mg Zn tree^{-1}, respectively) led to increases in pomegranate fruit yield, mainly due to increases in the number of fruits per tree. The effect was not as large with Zn as with B, and the lowest doses of Zn and B (17 mg B tree^{-1} or 318 mg Zn tree^{-1}, respectively) led to only minor effects in yield. Fertilization with the highest of the two doses also led to significant improvements in fruit quality at harvest, including 4.4–7.6% increases in TSS, 9.5–29.1% decreases in TA, 20.6–46.1% increases in maturity index and 0.28–0.62 pH unit increases in juice pH. Physical fruit characteristics (including fruit cracking, peel thickness, fruit length, fruit calyx diameter, fruit average weight, aril and peel percentages, the aril/peel ratio, weight of 100 arils and juice content of arils were unaffected. On the other hand, changes in total sugars and total phenolic compounds were only minor, in the ranges 1.9–4.6 and 1%, respectively, whereas the antioxidant activity and total anthocyanins were unaffected. The application of Zn and B also increased the leaf concentrations of both microelements in August, reflecting the improvements in tree nutrient status.

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References

Adsule, R.N., Patil, N.B., 1995. Handbook of fruit science and technology. In: Salunkhe, D.K., Kadam, S.S. (Eds.), Pomegranate. Marcel Dekker Publishers, New York, pp. 455–464.
Ali, A., Perveen, S., Muhammad Shah, S.N., Zhang, Z., Wahid, F., Shah, M., Bibi, S., Majid, A., 2014. Effect of foliar application of micronutrients on fruit quality of peach. Am. J. Plant Sci. 5, 1258–1264.
Alighourchi, H., Barzegar, M., Abbasi, S., 2008. Effect of gamma irradiation on the stability of anthocyanins and shelf-life of various pomegranate juices. Food Chem. 110, 1036–1040.
Alloway, B.J., 2008. Zinc in Soils and Crop Nutrition. International Zinc Fertilizer Industry Association, Brussels Belgium, pp. 30–35.
Amiri, M.E., Fallahi, E., Golchin, A., 2008. Influence of foliar and ground fertilization on yield, fruit quality, and soil, leaf and fruit mineral nutrition in apple. J. Plant Nutr. 31, 515–525.
Anees, M., Tahir, F.M., Shahzad, J., Mahmood, N., 2011. Effect of foliar application of micronutrients on the quality of mango (*Mangifera indica* L.) cv. Dusehri fruit. Mycopath 9, 25–28.
Anonymous, 2015. Iran Statistical Year Book. http://amar.maj.ir/Portal/File/ShowFile.aspx?ID=d017b964-cbc7-4c70-84e6-1c928aca0b2f.
Babu, K.D., Yadav, D.S., 2005. Foliar spray of micronutrients for yield and quality improvement in Khasi mandarin (*Citrus reticulata* Blanco). Int. J. Hortic. 62, 280–281.
Baldi, E., Toselli, M., Deudellar, D., Tagliavini, M., Marangoni, B., 2004. Foliar feeding of stone fruit trees. Informatore-Agrario 60, 43–46.
Barker, A.V., Pilbeam, D.J., 2006. Handbook of Plant Nutrition. CRC Press, ISBN 9780824759049.
Batsmanova, L.M., Gonchar, L.M., Taran, N.Y., Okanenko, A.A., 2013. Using a colloidal solution of metal nanoparticles as micronutrient fertiliser for cereals. Proceedings of the International Conference Nanomaterials: Applications and Properties.
Benton Jones Jr, J., Wolf, B., Mills, H.A., 1991. Plant Analysis Handbook: A Practical Sampling, Preparation, Analysis, and Interpretation Guide. Micro-Macro-Publishing, Athens, GA, U.S.A.
Boettcher, C., Keyzers, R.A., Boss, P.K., Davies, C., 2010. Sequestration of auxin by the indole-3-acetic acid-amido synthetase GH3–1 in grape berry (*Vitis vinifera* L.) and the proposed role of auxin conjugation during ripening. J. Exp. Bot. 61, 3615–3625.
Brady, N.C., Weil, R.R., 1996. The Nature and Properties of Soils. Prentice-Hall, New Jersey, U.S.A.
Brand-Williams, W., Cuvelier, M.E., Berset, C., 1995. Use of a free radical method to evaluate antioxidant activity. LWT-Food Sci. Technol. 28, 25–30.
Carrasco-Gil, S., Rios, J.J., Álvarez-Fernández, A., Abadía, A., García-Mina, J.M., Abadía, J., 2016. Effects of individual and combined metal foliar fertilization on iron- and manganese-deficient *Solanum lycopersicum* plants. Plant Soil 402, 27–45, Erratum Plant Soil. 402, 409–410.
Chapman, H.D., Pratt, P.F., 1961. Methods of Analysis for Soils, Plants and Water. University of California, Berkeley, CA, U.S.A.
Day, K., 1994. Correcting zinc deficiency in stone fruit orchard. Calif. Grower 18, 14–15.
Dear, J., Aronoff, S., 1965. Kinetics of chlorogenic and caffeic acids during the onset of boron deficiency in sunflower. Plant Physiol. 40, 458–459.
Dubois, M., Gilles, K., Hamilton, J.K., Rebers, P.A., Smith, F., 1951. A colorimetric method for the determination of sugars. Nature 168, 167.

El-Khawaga, A.S., 2007. Reduction in fruit cracking in 'Manfaluty' pomegranate following a foliar application with paclobutrazol and zinc sulphate. J. Appl. Sci. Res. 3, 837–840.

El-Sheikh, M.H., Khafgy, S.A.A., Zaied, S.S., 2007. Effect of foliar application with some micronutrients on leaf mineral content, yield and fruit quality of Florida prince desert red peach trees. J. Agric. Biol. Sci. 3, 309–315.

Fernández, V., Sotiropoulos, T., Brown, P.H., 2013. Foliar fertilization. In: Scientific Principles and Field Practices. International Fertilizer Industry Association, Paris.

Garcia, A., Haydar, N.E., Ferrer, C., 1984. Influence of Zn and Mn on the physiological behavior and yields of valencia oranges. Centro agrícola instituto superior agrícola ciego de avila. Cuba 10, 57–58.

Giusti, M.M., Wrolstad, R.E., 2001. Characterization and measurement of anthocyanins by UV–visible spectroscopy. In: Wrolstad, R.E., Schwartz, S.J. (Eds.), Current Protocols in Food Analytical Chemistry. John Wiley and Sons, New York, pp. 1–13.

Hansch, R., Mendel, R., 2009. Physiological functions of mineral micronutrients (Cu, Zn, Mn, Fe, Ni, Mo, B, Cl). Curr. Opin. Plant Biol. 12, 259–266.

Hasani, M., Zamani, Z., Savaghebi, G., Fatahi, R., 2012. Effects of zinc and manganese as foliar spray on pomegranate yield, fruit quality and leaf minerals. J. Soil. Sci. Plant Nutr. 12, 471–480.

Hasani, M., Zamani, Z., Savaghebi, G., Sheikh Sofla, H., 2016. Effect of foliar and soil application of urea on leaf nutrients concentrations, yield and fruit quality of pomegranate. J. Plant Nutr. 39, 749–755.

Holland, D., Hatib, K., Bar-Ya'akov, I., 2009. Pomegranate botany, horticulture, breeding. Hortic. Rev. 35, 127–191.

Jeyakumar, P., Durgadev, D., Kumar, N., 2001. Effects of zinc and boron fertilisation on improving fruit yields in papaya (*Carica papaya* L.) cv. Co5. Plant nutrition: food security and sustainability of agro ecosystems through basic and applied research. Fourteenth International Plant Nutrition Colloquium, Hannover, Germany, 356–357.

Kays, S.J., 1999. Preharvest factors affecting appearance. Postharvest Biol. Technol. 15, 233–247.

Keshavarz, K., Vahdati, K., Samar, M., Azadegan, B., Brown, P.H., 2011. Foliar application of zinc and boron improves walnut vegetative and reproductive growth. HortTechnology 21, 181–186.

Khalil, H.A., Aly, H.S.H., 2013. Cracking and fruit quality of pomegranate (*Punica granatum* L.) as affected by pre-harvest sprays of some growth regulators and mineral nutrients. J. Hortic. Sci. Ornament. Plants 5, 71–76.

Khan, N., Malik, A.B., Makbdoom, M.I., Hag, A., 1993. Investigations on the efficiency of exogenous synthetic growth regulators on fruit drop in mango (*Mangifera indica* L.). Egypt J. Hortic. 20, 1–14.

Khan, A.S., Ullah, W., Malik, A.U., Ahmad, R., Saleem, B.A., Rajwana, I.A., 2012. Exogenous applications of boron and zinc influence leaf nutrient status, tree growth and fruit quality of Feutrell's early (*Citrus reticulata* Blanco). Pak. J. Agric. Sci. 49, 113–119.

Khayyat, M., Tafazoli, E., Eshghi, S., Rajaee, S., 2007. Effect of nitrogen, boron, potassium and zinc sprays on yield and fruit quality of date palm. Am.-Eurasian J. Agric. Environ. Sci. 2, 289–296.

Khorsandi, F., Yazdi, F.A., Vazifehshenas, M.R., 2009. Foliar zinc fertilization improves marketable fruit yield and quality attributes of pomegranate. Int. J. Agric. Biol. 11, 766–770.

Kizilgoz, I., Sakin, E., Aslant, N., 2010. The effect of zinc fertilization on the yield of Pistachio (*Pistacia vera* L.) grown under rainfed condition. Afr. J. Agric. Res. 5, 3427–3430.

Korkmaz, N., Askin, M.A., 2015. Effects of calcium and boron foliar application on pomegranate (*Punica granatum* L.) fruit quality yield, and seasonal changes of leaf mineral nutrition. Acta Hortic. 1089, 413–422.

Legua, P., Melgarejo, P., Abdelmajid, H., Martínez, J.J., Martínez, R., Ilham, H., Hafida, H., Hernandez, F., 2012. Total phenols and antioxidant capacity in 10 Moroccan pomegranate varieties. J. Food Sci. 71, 115–120.

Liakopoulos, G., Karabourniotis, G., 2005. Boron deficiency and concentrations and composition of phenolic compounds in *Olea europaea* leaves: a combined growth chamber and field study. Tree Physiol. 25, 307–315.

Loneragan, J.F., Grunes, D.L., Welch, R.M., Advayi, E.A., Tengah, A., Lazer, V.A., Cary, E.E., 1982. Phosphorous accumulation and toxicity in leaves in relation to zinc supply. Soil Sci. Soc. Am. J. 46, 345–352.

Manas, D., Bandopadhyay, P.K., Chakravarty, A., Pal, S., Bhattacharya, A., 2014. Effect of foliar application of humic acid, zinc and boron on biochemical changes related to productivity of pungent pepper (*Capsicum annuum* L.). Afr. J. Plant Sci. 8, 320–335.

Marschner, H., 2012. Mineral Nutrition of Higher Plants. Academic Press Limited Harcourt Brace and Company, Publishers, London, pp. 347–364, ISBN: 978-0-12-384905-2.

Maurer, M.A., Taylor, K.C., 1999. Effect of foliar boron sprays on yield and fruit quality of navel oranges. University of Arizona College of Agriculture 1999 Citrus Research Report. http://ag.arizona.edu/pubs/crops/az1138.

Mengel, K., Kirkby, E.A., 2001. Principles of Plant Nutrition. Kluwer Academic Publisher, ISBN: 978-94-010-1009-2.

Mosanna, R., Khalilvand, B.E., 2015. Morpho-physiological response of maize (*Zea mays* L.) to zinc nano-chelate foliar and soil application at different growth stages. J. New Biol. Rep. 4, 46–50.

Naderi, M., Danesh Shahraki, A.A., Naderi, R., 2011. Application of nanotechnology in the optimization of formulation of chemical fertilizers. Iran J. Nanotech. 12, 16–23.

Naik, S.K., Chand, P.K., 2011. Tissue culture-mediated biotechnological intervention in pomegranate: a Review. Plant Cell Rep. 30, 707–721.

Nyomora, A.M.S., Brown, P.H., Freeman, M., 1997. Foliar applied boron increases tissue boron concentration and nut set of almond. J. Am. Soc. Hortic. Sci. 23, 159–165.

Obreza, T.A., Zekri, M., Hanlon, E.A., Morgan, K., Schumann, A., Rouse, R., 2010. Soil and Leaf Tissue Testing for Commercial Citrus Production. University of Florida Extension Service SL, pp. 253.04.

Ojeda-Barrios, D.L., Perea-Portillo, E., Hernández-Rodríguez, O.A., Martínez-Téllez, J., Abadía, J., Lombardini, L., 2014. Foliar fertilization with zinc in pecan trees. HortScience 49, 562–566.

Pandit, A.H., Wani, M.S., Mir, M.A., Bhat, K.M., Wani, S.M., Malik, A.R., 2011. Effect of foliar application of boron and zinc on fruit set and productivity of almond. Acta Hortic. 903, 1007–1010.

Peres, L.A., Reyes, R.D., 1983. Effect of nitrogen and boron application on *Carica papaya* L. 1. Growth and yield. J. Agric. Univ. Puerto Rico 67, 181–187.

Perica, S., Brown, P.H., Connell, J.H., Nyomora, A.M.S., Dordas, C., Hu, H.N., Stangoulis, J., 2001. Foliar boron application improves flower fertility and fruit set of olive. HortScience 36, 714–716.

Prasad, T.N.V.K.V., Sudhakar, P., Sreenivasulu, Y., Latha, P., Munaswamya, V., Raja Reddy, K., Sreeprasad, T.S., Sajanlal, P.R., Pradeep, T., 2012. Effect of nanoscale zinc oxide particles on the germination, growth and yield of peanut. J. Plant Nutr. 35, 905–927.

Raikova, O.P., Panichkin, L.A., Raikova, N.N., 2006. Studies on the Effect of Ultrafine Metal Powders Produced by Different Methods on Plant Growth and Development. Nanotechnologies and Information Technologies in the 21 st Century. Proceedings of the International Scientific and Practical Conference, 108–111.

Ramezani, S., Shekafandeh, A., 2009. Roles of gibberellic acid and zinc sulphate in increasing size and weight of olive fruit. Afr. J. Biotechnol. 8, 6791–6794.

Reynolds, G.H., 2002. Forward to the future nanotechnology and regulatory policy. Pac. Res. Inst. 24, 1–23.

Ryugo, K., 1988. Fruit Culture. John Wiley and Sons, New York, pp. 259–261.

Saadati, S., Moallemi, N., Mortazavi, S.M.H., Seyyednejad, S.M., 2013. Effects of zinc and boron foliar application on soluble carbohydrate and oil contents of three olive cultivars during fruit ripening. Sci Hortic. 164, 30–34.

Sahota, G.S., Arora, J.S., 1981. Effect of N and Zn on Hamlin sweet orange. J. Jap. Soc. Hortic. Sci. 50, 281–286.

Sarafi, E., Chatzissavvidis, C., Therios, I., 2013. Response of Two Pomegranate (*Punica Granatum* L.) Cultivars to Five B Concentrations: Growth Performance, B, Chlorophyll and Proline Concentration. Boron satellite meeting, Plant Nutrition Colloquium.

Sarafi, E., Chatzissavvidis, C., Therios, I., 2014. Effect of calcium and boron on the ion status, carbohydrate and proline content, gas exchange parameters and growth performance of pomegranate cv. Wonderful plants grown under NaCl stress. Turk. J. Agric. Nat. Sci. 2, 1606–1617, Special Issue.

Sarrwy, S.M.A., Gadalla, E.G., Mostafa, E.A.M., 2012. Effect of calcium nitrate and boric acid sprays on fruit set, yield and fruit quality of cv. Amhat date palm. World J. Agric. Sci. 8, 506–515.

Scott, N., Chen, H., 2003. Nanoscale Science and Engineering for Agriculture and Food Systems. A Report Submitted to Cooperative State Research, Education and Extension Service. USDA. National Planing Workshop, Washington.

Selivanov, V.N., Zorin, E.V., 2001. Sustained Action of ultrafine metal powders on seeds of grain crops. Perspekt. Materialy 4, 66–69.

Shahriaripour, R., Tajabadipour, A., 2010. Zinc nutrition of pistachio: interaction of zinc with other trace elements. Commun. Soil Sci. Plant Anal. 41, 1885–1888.

Sing, D.K., Paul, P.K., Ghosh, S.K., 2002. Response of papaya to foliar application of boron, zinc and their combinations. Department of Pomology and Postharvest Technol. Uttar Banga Krishi Viswavidyalaya, Pundibari-736 165, Cooch Behar (West Bengal), India.

Singh, R.S., Ram, S., 1983. Studies on the use of palm growth substances for fruit retention in mango cv. Dashehair. Indian J. Hortic. 40, 188–194.

Singh, N.B., Amist, N., Yadav, K., Singh, D., Pandey, J.K., Singh, S.C., 2013. Zinc oxide nanoparticles as fertilizer for the germination, growth and metabolism of vegetable crops. J. Nanoeng. Nanomanuf. 3, 353–364.

Singleton, V.L., Rossi, J.A., 1965. Colourimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. Am. J. Enol. Vitic. 16, 144–158.

Soliemanzadeh, A., Mozafari, V., Tajabadi pour, A., Akhgar, A., 2013. Effect of Zn, Cu and Fe foliar application on fruit set and some quality and quantity characteristics of pistachio trees. South West J. Hortic. Biol. Environ. 4, 19–34.

Song, C.Z., Liu, M.Y., Meng, J.F., Chi, M., Xi, Z.M., Zhang, Z.W., 2015. Promoting effect of foliage sprayed zinc sulfate on accumulation of sugar and phenolics in berries of *Vitis vinifera* cv. merlot growing on zinc deficient soil. Molecules 20, 2536–2554.

Subramanian, K.S., Sharmila Rahale, C., 2012. Ball milled nanosized zeolite loaded with zinc sulfate: a putative slow release Zn fertilizer. Int. J. Innov. Hortic. 1, 33–40.

Subramanian, K.S., Manikandan, A., Thirunavukkarasu, M., Sharmila Rahale, C., 2015. Nano-fertilizers for balanced crop nutrition. In: Rai, M., Ribeiro, C., Mattoso, L., Duran, N. (Eds.), Nanotechnologies in Food and Agriculture. Springer International Publishing, Switzerland, pp. 69–80.

Swietlik, D., 1999. Zinc nutrition in horticultural crops. Hortic. Rev. 23, 109–180.

Talaie, A., Badmahmoud, A.T., Malakout, M.G., 2001. The effect of foliar application of N, B and Zn on quantitative and qualitative characteristics of olive fruit. Iran J. Agric. Sci. 32, 727–736.

Tariq, M., Sharif, M., Shah, Z., Khan, R., 2007. Effect of foliar application of micronutrients on the yield and quality of sweet orange (*Citrus sinensis* L.). Pak. J. Biol. Sci. 10, 1823–1828.
Tehranifar, A., Zarei, M., Esfandiyari, B., Nemati, Z., 2010a. Physicochemical properties and antioxidant activities of pomegranate fruit (*Punica granatum*) of different cultivars grown in Iran. Hortic. Environ. Biotech. 51, 573–579.
Tehranifar, A., Zarei, M., Nemati, Z., Esfandiyari, B., Vazifeshenas, M.R., 2010b. Investigation of physico-chemical properties and antioxidant activity of twenty Iranian pomegranate *(Punica granatum* L.) cultivars. Sci. Hortic 126, 180–185.
Thompson, A.H., Batjer, L.P., 1950. The effect of boron in germinating medium on pollen germination and pollen tube growth for several deciduous fruit trees. Proc. Am. Soc. Hortic. Sci. 56, 227–230.
Usenik, V., Stampar, F., 2002. Effect of foliar application of zinc plus boron on sweet cherry fruit set and yield. Acta Hortic. 594, 245–249.
Usenik, V., Stampar, F., 2007. Effect of late season boron spray on boron accumulation and fruit set of 'Summit and Hedelfinger sweet cherry (*Prunus avium* L.). Acta Agric. Slov. 89, 51–58.
Varasteh, F., Arzani, K., Zamani, Z., Mohseni, A., 2009. Evaluation of the most important fruit characteristics of some commercial pomegranate (*Punica granatum* L.) cultivars grown in Iran. Acta Hortic. 818, 103–108.
Wiesner, M.R., Lowry, G.V., Alvarez, P., Dionysion, D., Biswas, P., 2006. Assessing the risks of manufactured nanomaterials. Environ. Sci. Technol. 40, 4336–4345.
Wojcik, P., Wojcik, M., 2003. Effect of boron fertilization on 'Conference' pear tree vigor, nutrition, and fruit yield and storability. Plant Soil 256, 413–421.
Wojcik, P., Cieslinski, G., Mika, A., 1999. Apple yield and fruit quality as influenced by boron applications. J. Plant Nutr. 9, 1365–1378.
Yadav, V., Singh, P.N., Yadav, P., 2013. Effect of foliar fertilization of boron, zinc and iron on fruit growth and yield of low-chill peach cv. Sharbati. Int. J. Sci. Res. Public 3, 1–6.
Yogeratnam, N., Greenham, D.W.P., 1982. The application of foliar sprays containing N, Mg, Zn and B to apple trees. I. Effect on fruit set and cropping. J. Hortic. Sci. 57, 151–154.