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Growth, yield, and calcium and boron uptake of tomato (*Lycopersicon esculentum* L.) and cucumber (*Cucumis sativus* L.) as affected by calcium and boron humate application in greenhouse conditions

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Abstract: The objective of this study was to examine the effect of calcium humate, boron humate, and humic acid solutions on growth, yield, quality, and calcium and boron uptake of tomato (*Lycopersicon esculentum* L.) and cucumber (*Cucumis sativus* L.), as well as changes in soil nutrient status after crop harvest. Four different concentrations (500, 1000, 3000, and 5000 mg kg^{-1}) of calcium humate (12% CaO, 15% humic and fulvic acid), boron humate (10% BOH_4, 15% humic and fulvic acid), and humic acid (15% humic and fulvic acid) were used in this research. The results revealed that Ca humate, B humate, and humic acid treatments applied at different rates positively affected the total marketable yield, average fruit weight, fruit diameter, fruit length, and leaf dry matter of tomato and cucumber plants. However, no statistically significant differences were observed among the treatments in terms of total soluble solids, fruit dry matter, chlorophyll, and plant length. The highest tomato yield (159,682 kg ha^{-1}) was obtained from 2936 mg kg^{-1} Ca humate application, while the highest cucumber yield (172,992 kg ha^{-1}) was obtained from 2910 mg kg^{-1} Ca humate application. In addition, the macro- and micronutrient concentration in leaves and fruits also increased significantly with these applications. Translocations of Ca and B from leaf to fruit were also affected positively by the Ca and B humate applications. Leaf Ca/fruit Ca ratio in tomato and cucumber decreased from 20.55 to 2.42 and from 10.70 to 1.37, respectively, while leaf B/fruit B ratio decreased from 15.80 to 1.82 in tomato and from 14.31 to 2.64 in cucumber. The applications also upgraded soil health by modifying the chemical characteristics of the soil and enhancing its nutrient status. The results showed that Ca and B humate nutrient sources should be used as an economical and simple application for tomato and cucumber plants without Ca and B deficiency during the plant growth period.

Key words: Calcium, boron, humate, tomato, cucumber, growth

1. Introduction

Vegetable crops grown under organic conditions play an important role in the global economy. Hence, some organic substances, such as humic acid (HA) and its salts calcium humate and boron humate, are used for plant cultivation around the world. Humic substances constituting 65%–70% organic matter are used in various areas of agriculture, namely plant physiology and environmental science, because of the multiple roles they play in these areas (Russo and Berlyn, 1990; Türkmen et al., 2004). HA-containing elements improve soil fertility and increase the availability of nutrient elements by holding them as mineral surfaces (El-Sharkawy and Abdel-Razzak, 2010; Paksoy et al., 2010). The important functional groups of HA include alcoholic hydroxyl, carboxyl, ketone, phenolic hydroxyl, and quinone (Russo and Berlyn, 1990). The mechanism of HA that promotes plant growth is not exactly known, but several explanations have been provided by researchers for increased oxygen uptake, respiration, photosynthesis, cell membrane permeability, phosphate uptake, and root cell elongation (Russo and Berlyn, 1990). Böhme and Lua (1997) stated that HA has important effects on nutrient uptake by plants and is also particularly important for the transport and availability of micronutrients.

Studies of the positive effects of HA on plant development have indicated the significance of optimum

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mineral supply, independently from nutrition (Chen and Aviad, 1990; Dursun et al., 2002). Many researchers have stated that humic substances lead to a remarkable increment in soil organic matter, improving plant growth and increasing crop production (Padem and Ocal, 1999; Abd El-Aal et al., 2005; Mahmoud and Hafez, 2010).

Calcium's functions in plants were widely reviewed by Hanson (1984) and Kirkby and Pilbeam (1984). Ca has an important effect on plant growth due to its function as a second messenger in the signal conduction among environmental factors and responses in plant growth and development. The important role of Ca in plant growth and nutrient uptake is emphasized in many ways. It can be indicated most easily by increased leakage of low-molecular-weight solutes from cells of Ca-deficient tissues in seriously deficient plants, by a general disintegration of membrane structures, and by a loss of cell segmentation (van Goer, 1996). The development of soil conditions and the establishment of equilibrium among mineral elements are also significant for soil productivity and plant production.

Boron is a minor element, and plants differ widely in their demands; the ranges of deficiency and toxicity levels are narrow. B management is challenging as its optimum application range is limited, and optimum B application rates can differ from one soil to another (Gupta, 1993; Marschner, 1995). The functions of B in plants have been related to cation and anion absorption, water relations, pollen viability, and the metabolism of phosphorus, nitrogen, fats, and carbohydrates (Shol'nik, 1965). Boron also affects cell division, cell-wall synthesis, membrane functioning, sugar transport, differentiation, regulation of plant hormone levels, root elongation, and growth of plants (Marschner, 1995).

Ca and B are considered to be vital elements in the primary walls, cell membranes, fruit growth, and development of plant cells (Pilbeam and Morely, 2007). Ca and B uptake and transportation from the soil to the plant shoots, leaves, and fruits are very limited and generally dependent on the loss of water through transpiration for uptake; thus, Ca and B are classified as immobile elements in plants (Clarkson, 1984). Because of these characteristics of Ca and B, shoot deficiency symptoms appear primarily in the upper leaves. Visual symptoms include deformed, strap-like leaves; chlorosis; and leaves that develop yellow-to-tan margins, eventually becoming necrotic (Nelson, 2003). Low levels of Ca in fruit tissues can also cause blossom end rot, which is a physiological disorder that reduces the yield of many vegetables such as tomato (Tonetto de Freitas et al., 2011).

B and Ca are important micro- and macronutrients required for plant growth and development. However, Ca and B uptake and their utilization in plants are very limited in agricultural practice. At present, there are several studies on increasing Ca and B uptake and plant growth. However, none of these studies have solved the problem. In this study, new substances were used, Ca humate and B humate, to increase plant Ca and B uptake and transport to the leaves and fruits of tomatoes and cucumbers. The objective of this research was to determine the effects of Ca humate, B humate, and HA applications on plant Ca and B uptake, transport to leaves and fruit, plant growth, and yield of tomato and cucumber.

2. Materials and methods

2.1. Experimental setup

The study was carried out under greenhouse conditions at Atatürk University, Erzurum (40°31'N, 40°54'E), Turkey. Kayra F1 tomato cultivar (*Lycopersicon esculentum* L.) and A21 F1 cucumber cultivar (*Cucumis sativus* L.) were grown as plant materials under natural light conditions, approximate day/night temperatures of 27/14 °C, and 75% relative humidity during the span of the experiment. Four different rates (500, 1000, 3000, and 5000 mg kg^{-1}) of Ca humate (Ca actosol containing 12% CaO, 15% humic and fulvic acid), boron humate (B actosol containing 10% BOH_4, 15% humic and fulvic acid), and humic acid (actosol containing 15% humic and fulvic acid) were applied as nutrient materials (Figures 1 and 2). The solutions (300 mL of corresponding source and concentration for each plant) were applied to the plant root zone with injection at 3 different growth stages (i.e. 20 days after planting, at the beginning of flowering, and in the middle of the harvest), except for the control plants, which were treated with water. The soil in the experimental area was analyzed before sowing and was identified as sandy loam with 35.6% sand, 48.2% silt, and 16.2% clay. The experiment was designed as completely randomized with 3 replicates and 10 plants in each replicate.

Tomato and cucumber seeds were sown in 45-celled seedling trays filled with peat, and 35-day-old seedlings were transplanted to the soil with 70 × 50 cm row spacing distances in the second week of May 2013. Before planting, N, P, and K were banded and applied to each plot at the rates of 350 N kg ha^{-1} (as ammonium sulfate), 110 kg P ha^{-1} (as triple super phosphate), and 180 kg K ha^{-1} (as potassium sulfate) as a basal fertilizer. During the growing period, plants were drip-irrigated to maintain 60% soil moisture. Cucumber fruits were initially harvested in the third week of June while the tomato fruits were harvested

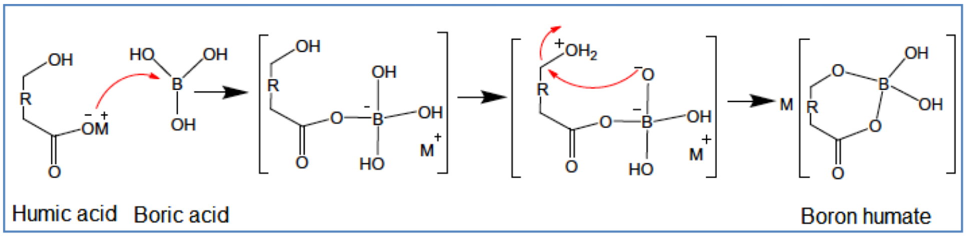

Figure 1. The symbolic structure of B humate.

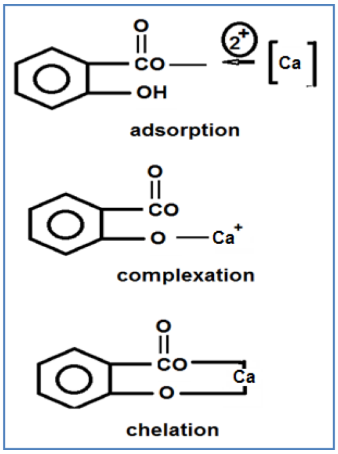

Figure 2. The symbolic structure of Ca humate.

in the second week of August, and fruit picking was done regularly at 2-day intervals. The studies were terminated in the second week of October of the same year.

2.2. Growth and yield parameters

The effect of the applications on total marketable yield, plant length, and fruit and leaf dry matter was evaluated. Furthermore, 10 fruits (i.e. tomato and cucumber) from each replicate were analyzed for fruit diameter, average fruit weight, fruit length, and total soluble solids (TSS). The plant samples for dry weight were kept in an oven at 65 °C for 48 h. The TSS levels in fruits were determined with a portable refractometer. Leaf greenness was determined with a portable chlorophyll meter (SPAD-502).

2.3. Mineral analysis

Fruit and leaf tissue was taken during the harvest after the last applications and then oven-dried at 68 °C for 48 h and ground to pass through a 1-mm mesh screen. The Kjeldahl method and a Vapodest 10 Rapid Kjeldahl distillation unit (Gerhardt, Königswinter, Germany) were used to determine total N (Bremner, 1996). Macro- [Ca, K, Mg, Na, and P] and microelements (B, Cu, Fe, Mn, and Zn) were determined after the wet digestion of dried and ground subsamples using a HNO_3-H_2O_2 acid mixture (2:3 v/v) in 3 steps [first step: 145 °C, 75% radio-frequency power (RF), 5 min; second step: 180 °C, 90% RF, 10 min; and third step: 100 °C, 40% RF, 10 min] in microwave digestion (Bergof Speedwave Microwave Digestion Equipment MWS-2, Eningen, Germany) (Mertens, 2005a). B, Ca, Cu, Fe, K, Mg, Mn, Na, P, and Zn were determined using an inductively coupled plasma spectrophotometer (Optima 2100 DV, ICP/OES; Perkin-Elmer, Shelton, CT,

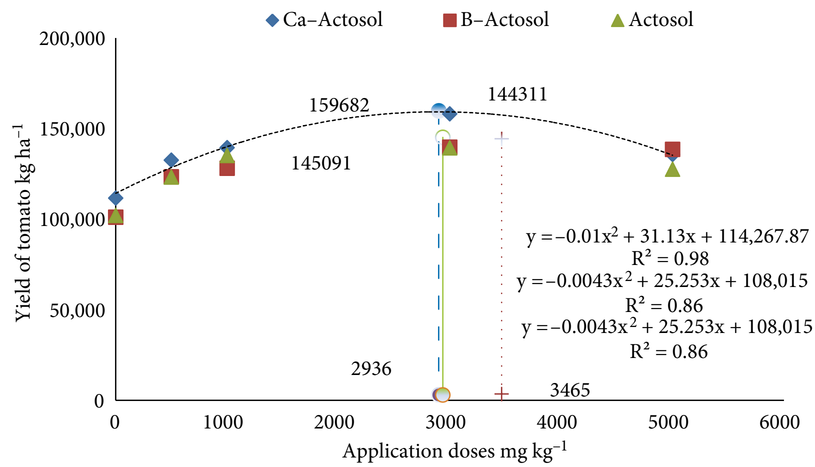

Figure 3. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on tomato yield.

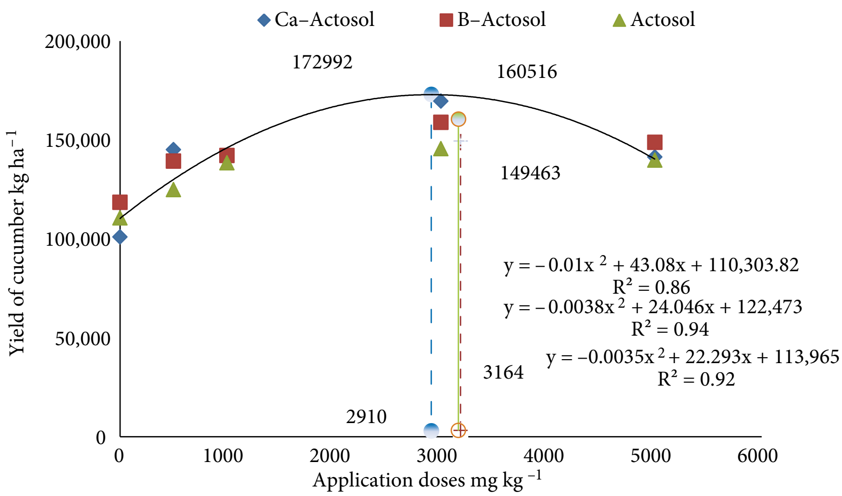

Figure 4. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on cucumber yield.

USA) (Mertens, 2005b). A postharvest soil sample was also taken and sieved with a 2-mm mesh screen to analyze the different chemical properties and soil nutrient statuses.

2.4. Data analysis

Studies were conducted based on a completely randomized design with 4 treatments and 3 replicates, each with 10 plants. All data were subjected to analysis of variance using SPSS 18.0 (SPSS Inc., 2010). Means were separated by Duncan's multiple range test (DMRT).

3. Results

3.1. Growth and yield parameters

Ca humate (Ca actosol), B humate (B actosol), and humic acid (actosol) treatments applied at different rates positively affected the total marketable yield, average fruit weight, fruit diameter, fruit length (for cucumber), and leaf dry matter of tomato (Figure 3; Table 1) and cucumber plants (Figure 4; Table 2). However, no statistically significant differences were observed among the treatments in terms of fruit length (for tomato), TSS, fruit dry matter, chlorophyll, and plant length (Figures 5 and 6).

Statistical analysis showed significant differences among Ca actosol, B actosol, and actosol treatments for average fruit weight (Tables 1 and 2). When comparing the treatments, the highest average tomato fruit weight (162.80 g) was attained with the application of 3000 mg Ca actosol kg^{-1} while the application of B actosol at 1000 mg kg^{-1} showed the highest average fruit weight of cucumber (190.00 g). Control plants had the lowest value of average fruit weight in both crops.

Fruit diameter also exhibited a similar trend to that observed in fruit weight. There was a significant difference among treatments regarding fruit diameter (Tables 1 and 2). The highest fruit diameter (69.64 mm) for tomato and cucumber (38.93 mm) was recorded in 3000 mg kg^{-1} Ca and 1000 mg kg^{-1} B actosol application, respectively, while the lowest fruit diameter was obtained from the control (Tables 1 and 2). The effects of the applications on tomato fruit length were not statistically significant. However, the highest fruit length of the cucumber plant (18.13 cm) was noted with 1000 mg kg^{-1} B application, and the effects of the applications were statistically significant (Tables 1 and 2).

Results indicated that leaf dry matter of both tomato and cucumber exhibited significant ($P < 0.05$) responses to the application of Ca actosol, B actosol, and actosol treatments (Tables 1 and 2). The highest leaf dry matter of tomato (16.63%) and cucumber (13.59%) was obtained from 500 mg kg^{-1} B actosol and 5000 mg kg^{-1} actosol application, respectively.

Total marketable yield of both tomato and cucumber was significantly affected by the application of Ca actosol, B actosol, and actosol treatments (Tables 1 and 2). The highest yield per hectare (143,085 kg ha^{-1}) of tomato was obtained from 3000 mg kg^{-1} Ca actosol application, and the lowest yield was determined as 111,603 kg ha^{-1} in the control. Furthermore, the highest yield (158792 kg ha^{-1}) of cucumber was obtained from 5000 mg kg^{-1} Ca actosol application, and the lowest yield was determined as 118,503 kg ha^{-1} in the control. According to the regression analysis, the highest marketable yield per hectare (159,682 kg) of tomato could be obtained from 2936 mg kg^{-1} Ca humate application, whereas the highest marketable yield of cucumber (172,992 kg ha^{-1}) could be obtained from 2910 mg kg^{-1} Ca humate application (Figures 3 and 4).

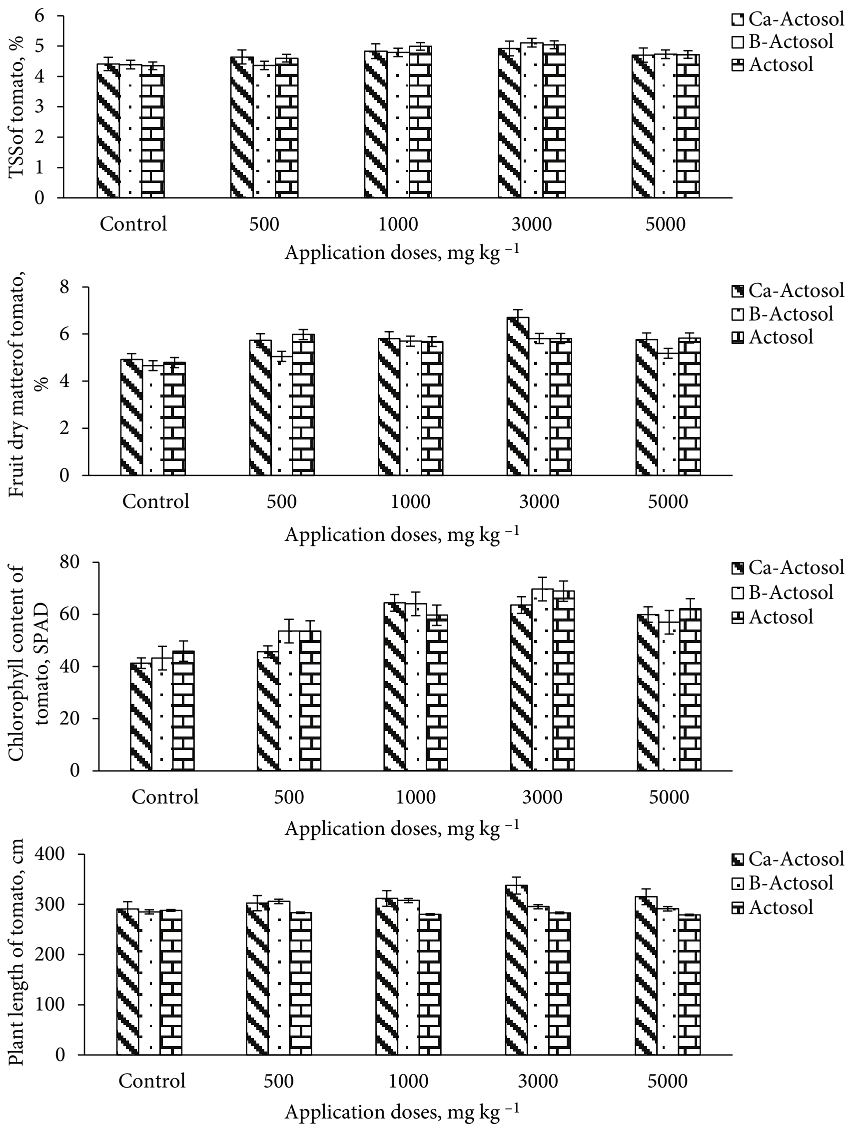

Figure 5. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on TSS, fruit dry matter, chlorophyll, and plant length of tomato.

3.2. Nutrient analysis of tomato leaf and fruit

The effects of applications on nutrient content of fruit and leaf were found to be statistically significant. In tomato fruits, compared to the other soil amendments, B actosol showed the highest N, P, Fe, Mn, Cu, and B content as 3.51%, 0.70%, 125.87 mg kg^{-1}, 49.55 mg kg^{-1}, 11.28 mg kg^{-1}, and 15.76 mg kg^{-1}, respectively. However, the highest content of Mg, Ca, and Zn (0.28%, 0.99%, and 59.30 mg kg^{-1}, respectively) was observed in Ca actosol applications, and of K (2.72%) in actosol applications (Table 3).

On the other hand, data regarding leaf nutrient contents illustrated the highest values of N (3.28%), K (3.89%), Mg (0.45%), Mn (20.97 mg kg^{-1}), and B (89.17 mg kg^{-1}) in B actosol, and of P and Cu in actosol applications (0.56% and 7.00 mg kg^{-1}, respectively). Ca actosol exhibited the highest values in the cases of Ca, Fe, and Zn contents (4.70%, 51.04 mg kg^{-1}, and 58.80 mg kg^{-1}, respectively) (Table 4).

Ca actosol and B actosol significantly translocated the nutrients from leaves to fruits in tomato plants. As regards the Ca actosol application, the ratio between leaf Ca (1.85%)/fruit Ca (0.09%) was 20.55 in control treatments; however, the leaf Ca (4.82%)/fruit Ca (1.99%) ratio was 2.42 in 3000 mg kg^{-1} Ca actosol treatments. On the other hand, leaf B (4.9 mg kg^{-1})/fruit (0.31 mg kg^{-1}) ratio was 15.80 in control treatments, but leaf B (83.72 mg kg^{-1})/fruit B (45.76 mg kg^{-1}) ratio was 1.82 in 3000 mg kg^{-1} B actosol treatments (Figure 7).

Table 1. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on total marketable yield, average fruit weight (FW), fruit diameter, fruit length, and leaf dry matter of tomato.

Applications	Concentration (mg kg^{-1})	Total marketable yield (kg ha^{-1})	Average fruit weight (g)	Fruit diameter (mm)	Fruit length (mm)	Leaf dry matter (%)
Control	0	111,603 c*	129.20 c*	62.78 c**	52.34 ^{ns}	14.80 bc*
Calcium actosol	500	132,448 ab	143.87 bc	64.52 abc	54.48	14.68 bc
	1000	130,440 ab	154.27 ab	67.78 a	56.26	14.42 c
	3000	143,085 a	162.80 a	69.64 a	56.75	14.77 bc
	5000	135,655 ab	147.20 ab	66.68 ab	55.65	14.98 bc
Boron actosol	500	132,505 ab	149.87 ab	67.25 a	55.37	16.63 a
	1000	139,615 ab	156.27 ab	67.76 a	55.50	15.46 abc
	3000	128,085 abc	145.47 bc	65.31 abc	55.25	15.64 abc
	5000	123,352 bc	147.20 ab	66.14 ab	55.27	15.78 ab
Actosol	500	123,215 bc	146.80 ab	66.55 ab	54.85	15.52 abc
	1000	124,250 bc	145.20 bc	65.18 abc	54.62	15.82 ab
	3000	125,070 bc	139.73 bc	63.75 bc	54.29	15.75 ab
	5000	127,473 abc	151.60 ab	67.29 a	55.58	14.82 bc

Means within column not followed by the same letter differ significantly by DMRT. *: $P < 0.05$; **: $P < 0.01$.

3.3. Nutrient analysis of cucumber leaf and fruit

Cucumber fruit also exhibited the same trend as tomato for nutrient contents, i.e. the highest content of N (4.33%), P (1.01%), Mg (0.32%), Fe (177.38 mg kg^{-1}), Mn (70.48 mg kg^{-1}), Cu (11.46 mg kg^{-1}), and B (50.90 mg kg^{-1}) was attained with B actosol applications. However, the highest content of Ca and Zn (1.99% and 62.48 mg kg^{-1}, respectively) in Ca actosol and K (4.43%) was attained with actosol applications (Table 5). Moreover, in cucumber leaves, B actosol applications showed the highest content of N, K, Mg, Zn, and B as 3.29%, 5.53%, 0.69%, 76.72 mg kg^{-1}, and 99.17 mg kg^{-1}, respectively, while Ca actosol showed highest values for P (0.75%), Ca (4.82%), Fe (90.75 mg kg^{-1}), and Mn (55.08 mg kg^{-1}). The highest content of Cu (8.32 mg kg^{-1}) was noted in actosol applications (Table 6). Calcium actosol and B actosol significantly translocated the nutrients from leaves to fruits in cucumber plants. In Ca actosol application, leaf Ca (0.76%)/fruit Ca (0.076%) ratio was 10.70 in control treatments, and leaf Ca (2.66%)/ fruit Ca (1.94%) ratio was 1.37 in 3000 mg kg^{-1} Ca actosol treatments. On the other hand, leaf B (8.59 mg kg^{-1})/fruit (0.60 mg kg^{-1}) was 14.31 in control treatments, but leaf B (88.72 mg kg^{-1})/fruit B (33.59 mg kg^{-1}) ratio was 2.64 in 3000 mg kg^{-1} B actosol treatments. These results show that Ca and B actosol application increased the calcium and boron translocation from the leaves to the fruit (Figure 8).

3.4. Soil chemical properties and nutrient status

Soil analysis after crop harvest showed that the highest pH value (7.57) was found in the treatments where Ca actosol and B actosol were applied at the rate of 5000 mg kg^{-1}, increased by 5% compared to the control (Table 7). The organic matter content was highest after the treatment, with 5000 mg kg^{-1} B actosol showing 50% increase compared to the control. Soil $CaCO_3$ content decreased with increasing concentration of applications, except for Ca actosol. The highest soil $CaCO_3$ content was determined in 500 mg kg^{-1} Ca actosol and B actosol applications (1.12% and 1.10%, respectively). Conversely, the content of NH_4, NO_3, and cation exchange capacity (CEC) increased with increasing concentration of applications. The application of 5000 mg kg^{-1} Ca actosol exhibited the highest CEC (45.20 cmolc kg^{-1}) and soil N-NO_3 content (158.00 mg kg^{-1}) (Table 7).

Generally, soil nutrient status increased with the increasing rate of applications. The concentration of K, Ca, and Zn in the soil was found as highest after the application of 5000 mg kg^{-1} Ca actosol (3.19 cmolc kg^{-1}, 42.20 cmolc kg^{-1}, and 4.02 mg kg^{-1}, respectively), while the application of both Ca and B actosol at the rate of 5000 mg kg^{-1} showed the highest contents of Na, Fe, Cu, and Mn (0.77 cmolc kg^{-1}, 3.10 mg kg^{-1}, 4.60 mg kg^{-1}, and 18.00 mg kg^{-1}, respectively). A significant reduction in soil Mg content was recorded with these applications, and the

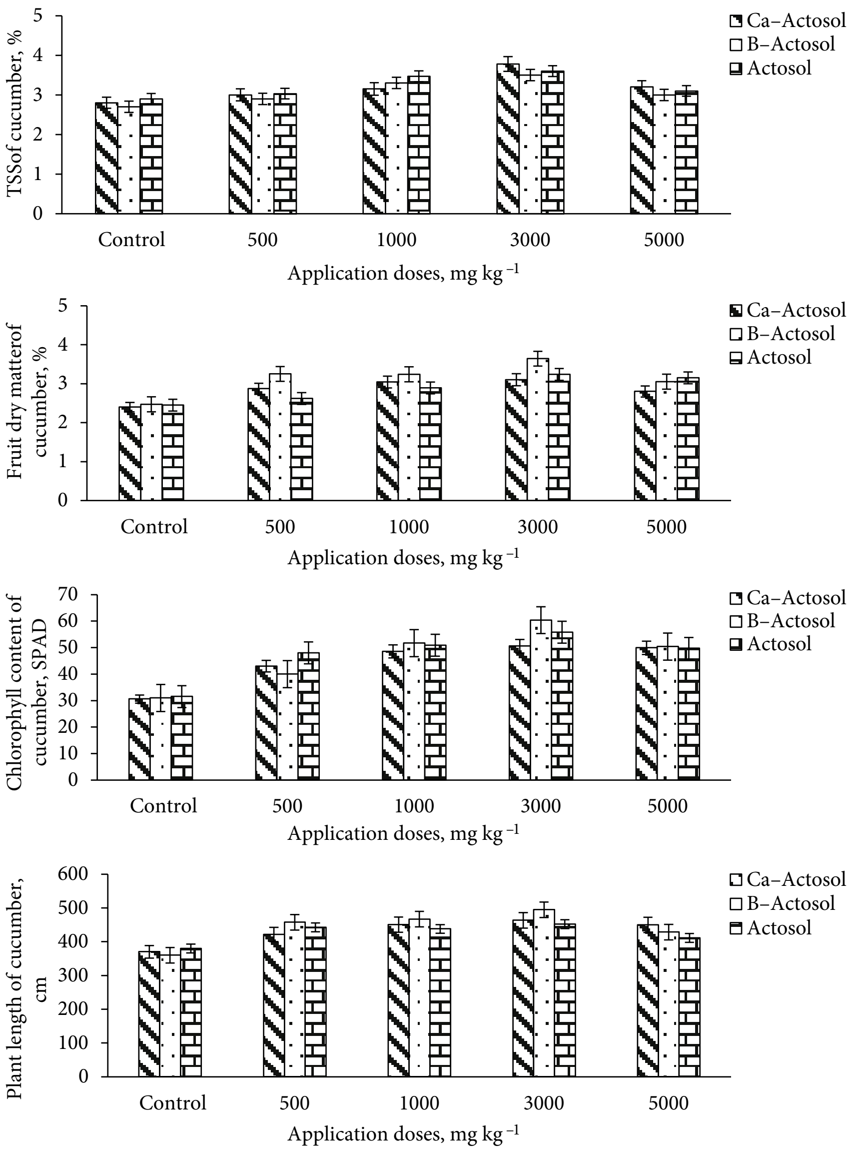

Figure 6. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on TSS, fruit dry matter, chlorophyll, and plant length of cucumber.

highest content of Mg (4.94 cmolc kg^{-1}) was found in the control. Treatment with B actosol increased the B content of the soil, and the highest value of B (4.10 mg kg^{-1}) was determined in 5000 mg kg^{-1} B actosol application (Table 8).

4. Discussion

4.1. Growth and yield parameters

The results of the present investigation revealed the significant effects of Ca actosol, B actosol, and actosol treatment on yield and yield attributes of tomato and cucumber when compared to the control. Among the treatments, the effect of Ca actosol and B actosol was more prominent for most of the traits over actosol application. As regards the total marketable yield, Ca actosol and B actosol resulted in increased yield of tomato (rates of 28.21%, 25.10%, and 14.22%, respectively) and cucumber (rates of 34.15%, 22.45%, and 22.22%, respectively). According to the results of the study, total tomato and cucumber marketable yield did not increase with increased Ca actosol and B actosol doses; the maximum values were determined at 3000 mg Ca actosol kg^{-1}, while the application of B actosol was determined at 1000 mg kg^{-1} (Figures 3 and 4). Humic substance has auxin, gibberellin, and cytokine, as well as organic acid and amino acid. According to the regression analysis, the results of this study show that 3000

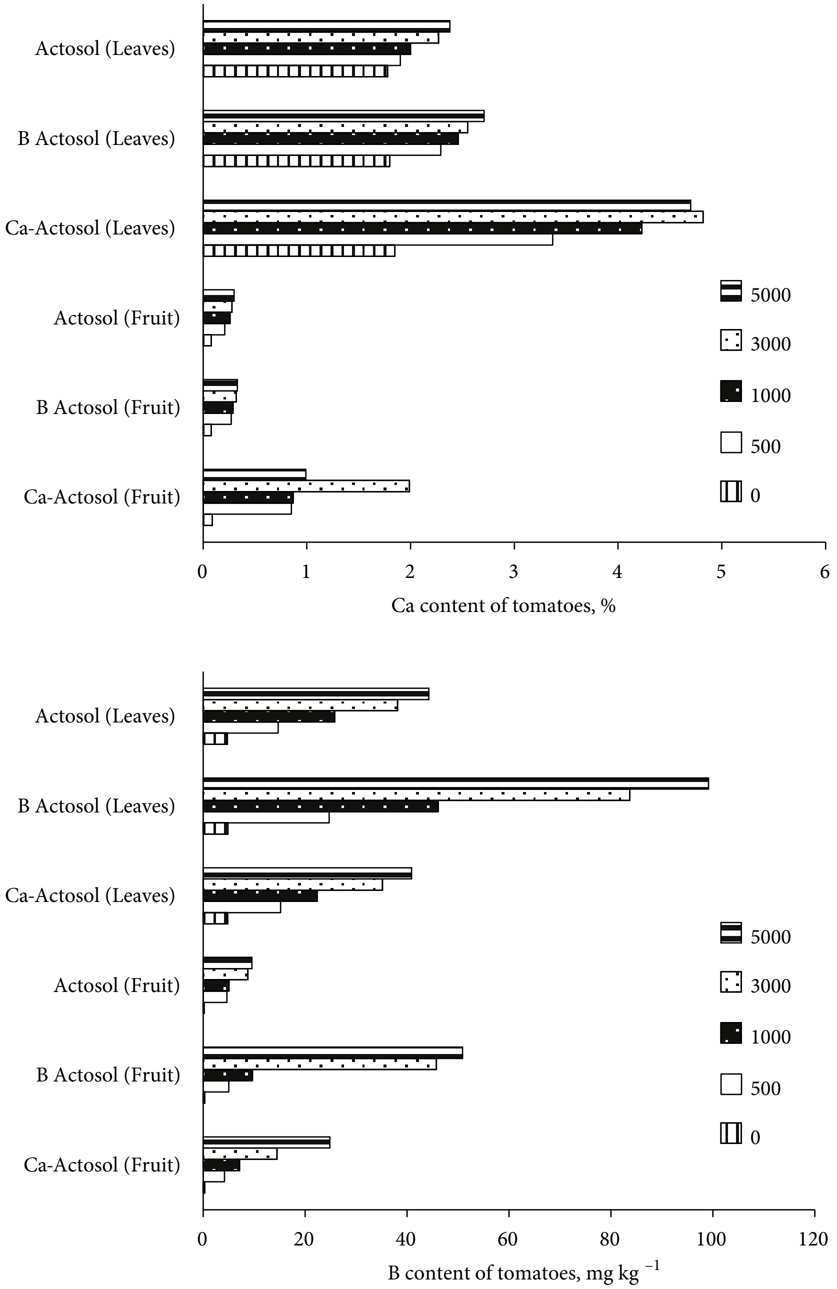

Figure 7. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on calcium and boron translocation from leaves to fruit of tomato.

mg kg^{-1} is a sufficient level for optimum yield of tomato and cucumber plant. Doses above this level can cause a decrease in yield because of the inhibition effects of plant phytohormone content; in addition, an increase in the osmotic potential of soil and plants needs extra irrigation.

Our results are in accordance with the findings of Çelik et al. (2008) and Karakurt et al. (2009), who documented a significant increase in growth and yield with the application of Ca and HA. Similarly, Karaman et al. (2012) documented that B humate as soil + foliar application on tomato crop increased fruit diameter, number of branches, and plant B content by 37%, 50%, and 84%, respectively, while Ca humate application increased root weight, plant weight, and plant Ca content by 62%, 29%, and 70%, respectively, when compared to the control.

Yildirim (2007) determined that HA treatments (doses of 10 mL/L and 20 mL/L; soil and foliar applications) significantly increased fruit number, diameter, weight,

Table 2. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on total marketable yield, average fruit weight (FW), fruit diameter, fruit length, and leaf dry matter of cucumber.

Applications	Concentration (mg kg^{-1})	Total marketable yield (kg ha^{-1})	Average fruit weight (g)	Fruit diameter (mm)	Fruit length (cm)	Leaf dry matter (%)
Control	0	118,503 c**	127.67 f***	34.24 e**	16.30 c*	9.16 h***
Calcium actosol	500	139,312 b	164.00 bc	37.24 abcd	17.13 abc	11.91 f
	1000	142,080 ab	168.33 bc	36.23 cde	17.83 ab	12.11 e
	3000	148,870 ab	161.00 bcd	36.21 cde	17.37 abc	11.55 g
	5000	158,792 a	163.33 bcd	37.22 abcd	17.20 abc	12.46 d
Boron actosol	500	145,103 ab	176.00 ab	38.18 abc	17.32 abc	11.99 ef
	1000	142,178 ab	190.00 a	38.93 a	18.13 a	12.71 c
	3000	139,637 b	142.33 def	35.07 de	16.57 c	13.47 a
	5000	141,400 ab	139.00 ef	35.09 de	16.47 c	13.14 b
Actosol	500	144,832 ab	153.33 cde	36.73 abcd	16.70 bc	12.43 d
	1000	138,410 b	172.00 abc	38.75 ab	16.60 bc	13.52 a
	3000	135,580 b	155.00 bcde	36.45 bcde	16.50 c	12.54 cd
	5000	139,713 b	156.33 bcde	36.20 cde	17.37 abc	13.59 a

Means within column not followed by the same letter differ significantly by DMRT. *: $P < 0.05$,**: $P < 0.01$,***: $P < 0.001$.

yield (with foliar 20 mL/L HA), TSS, and dry matter of tomato fruits. El-Nemer et al. (2012) found a significant effect of HA on fruit length, fruit diameter, and fruit yield of cucumber, showing an increase of 89%, 44% and 85%, respectively, over the control. The results of our study are also supported by the findings of Hao and Papadopoulos (2003), who reported increased fruit yield and growth of tomato with HA and Ca sprays.

The increase in yield and yield components of both tomato and cucumber recorded in this study could be due to the fact that HA stimulates root growth and affects root morphology by organic acid exudation, which in turn leads to increased nutrient uptake and thereby improves the growth and yield of crops (Canellas et al., 2008). Humic substances with auxin-like activities could promote cell elongation, apical dominance, and rooting, resulting in high crop yield (Atiyeh et al., 2002; Nardi et al., 2002).

The hormone-like activity of HA, which is indicated as concentration-specific (Zandonadi et al., 2007), and improved absorption of mineral nutrients because of increases in cell permeability (Dursun et al., 2002; Zandonadi et al., 2007) could be responsible for the stimulatory effect of HA on plant yield and quality. Furthermore, the formation of a complex between HA and mineral ions, the catalysis of HA by the enzymes in the plant, the influence of HA on respiration and photosynthesis, the stimulation of nucleic acid metabolism, and the hormonal activity of HA are among the factors that have been used to describe the effect of HA on plant growth parameters (Türkmen et al., 2004).

Another possible factor might be Ca, which along with cell division and cell elongation is also involved in cell membrane stability and permeability, thus strengthening the plants (Ashraf, 2004; Hirschi, 2004). Calcium deficiency results in reduced leaf size, necrosis of young leaves, and reduction in fruit quality and yield (Hao and Papadopoulos, 2003). Increasing Ca^{2+} concentration in the nutrient solution increased plant length, dry matter yield, leaf area, fruit fresh weight, number of fruits, fruit diameter, total yields, and marketable fruit yields (Hao and Papadopoulos, 2003; Rubio et al., 2009; Shafeek et al., 2013).

B fertilization is frequently used in agricultural production. The positive effect of B on different crops was also previously reported by many researchers, i.e. on tomato (Salam et al., 2009), wheat (Güneş et al., 2003), sunflower (Oyinlola, 2007), pepper and cucumber (Dursun et al., 2010), celery (Dambrauskienė et al., 2007), and soybean (Ross et al., 2006). As a result of these investigations, it was reported that different B applications increased the nutrient content, plant growth, and yield of different plant species.

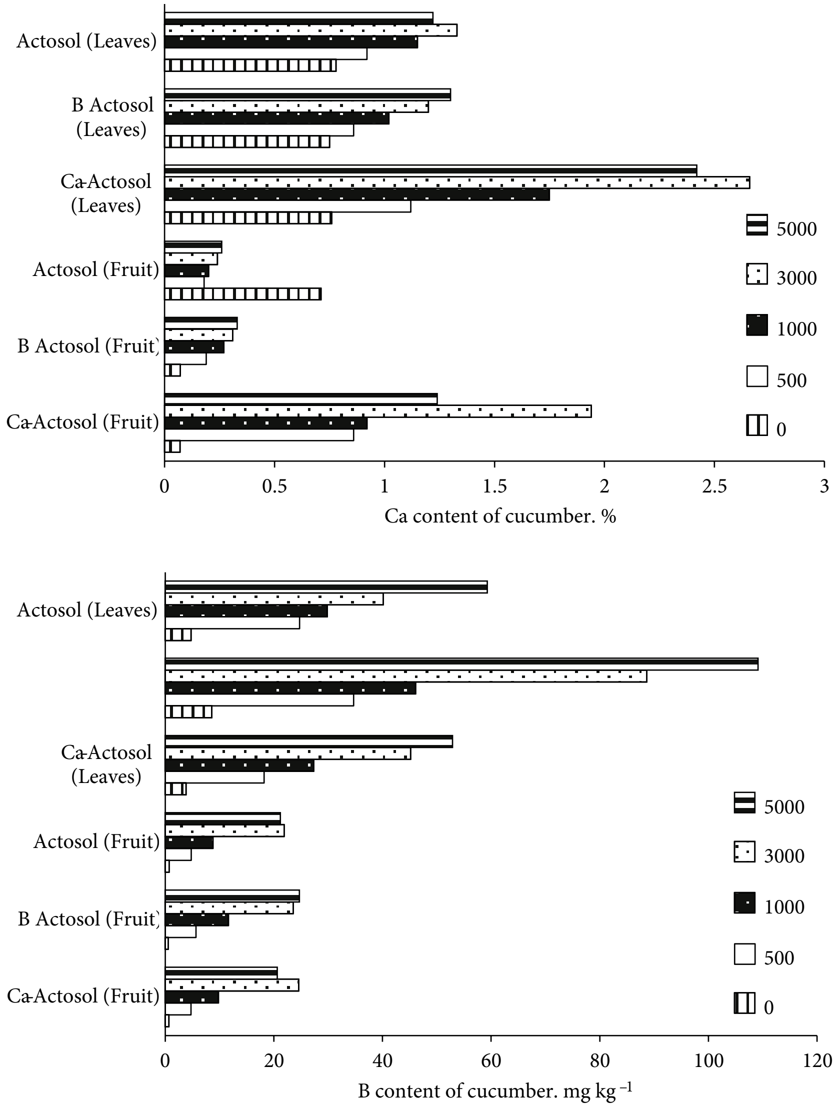

Figure 8. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on Ca and B translocation from leaves to fruit of cucumber.

4.2. Accumulation and translocation of nutrients

Application of Ca actosol, B actosol, and actosol treatments significantly increased fruit and leaf nutrient element contents of both tomato and cucumber over the control. Humic substance has chelator properties. If HA is applied to soil, the application increases fertilizer use efficiency and plant uptake, especially for micronutrient elements such as Fe, Mn, Zn, Cu, and B, in addition to N, P, and Ca. A benefit of HA in agricultural systems is its ability to complex metal ions (Stevenson, 1982). HA can form aqueous complexes with micronutrients, although not to the same extent as many synthetic chelating agents (Aiken et al., 1985). It increases cation exchange capacity and enhances soil fertility, converting the mineral elements into forms available to plants (Stevenson, 1994). Some reports explain these positive effects in terms of the ability of HA to hold the nutrients in the rhizosphere. HA enhances the absorption capacity of the nutrients of the roots by having carboxylic and phenolic groups and increasing H^+-ATPase activity in the root cells (Canellas et al., 2002)

The available studies have revealed correlations between root growth and development and the uptake of some nutrients. For instance, HA caused an increase in the length and dry weight of maize plant roots and enhanced the uptake of nitrogen, phosphorus, K^+, Ca^{2+}, Cu^{2+}, Mn^{2+}, Zn^{2+}, and Fe^{3+} (Eyheraguibel et al., 2008). Humic substances increased root length in sunflower (Kolsarıcı et al., 2005), as well as root dry weight in tomato and cucumber (Atiyeh et al., 2002). Similarly, humic substances stimulated root development and enhanced nitrogen, K^+, Cu^{2+}, and Mn^{2+}

Table 3. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on the nutrient content of tomato fruits.

Applications	Concentration	N	P	K	Mg	Ca	Fe	Zn	Mn	Cu	B
	mg kg^{-1}	%					mg kg^{-1}				
Control	0	1.73 h ***	0.16 d***	0.38 g***	0.18 ns	0.19 d***	8.95 m***	16.84 i***	6.07 m***	0.58 l***	2.31 d***
	500	2.28 e	0.42 c	0.38 g	0.28	0.85 b	36.50 k	41.79 d	26.97 h	4.53 k	2.20 d
Calcium	1000	2.37 d	0.49 bc	0.36 g	0.23	0.87 b	58.67 g	48.45 c	34.70 g	4.86 j	3.18 c
Actosol	3000	2.32 de	0.50 bc	0.34 g	0.24	0.99 a	60.65 f	59.30 a	37.10 f	6.10 h	3.52 c
	5000	2.26 e	0.53 b	0.25 h	0.24	0.99 a	68.51 e	56.67 b	38.72 d	6.84 e	3.90 c
	500	2.06 f	0.49 bc	0.61 f	0.23	0.25 d	125.02 b	15.67 j	38.37 e	11.28 a	4.10 c
Boron	1000	2.58 b	0.57 b	0.81 d	0.26	0.27 c	125.87 a	12.40 k	47.23 c	10.67 b	5.02 c
Actosol	3000	3.51 a	0.68 a	0.81 d	0.24	0.29 c	118.54 c	9.49 l	47.90 b	9.36 c	9.71 b
	5000	2.48 c	0.70 a	0.70 e	0.24	0.32 c	105.34 d	7.78 m	49.55 a	7.44 d	15.76 a
	500	1.92 g	0.14 d	1.92 c	0.19	0.22 d	19.94 l	20.34 f	10.20 l	6.04 h	2.09 d
	1000	2.12 f	0.17 d	2.43 b	0.17	0.18 d	37.85 j	20.56 e	11.72 k	6.24 g	2.22 d
Actosol	3000	2.46 c	0.17 d	2.72 a	0.14	0.17 d	47.21 i	19.46 g	12.82 i	5.85 i	4.69 c
	5000	2.27 e	0.21 d	2.41 b	0.11	0.16 d	49.25 h	19.31 h	12.16 j	6.62 f	5.10 c

Means within column not followed by the same letter differ significantly by DMRT ($P < 0.001$).

Table 4. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on the nutrient content of tomato leaves.

Applications	Concentration	N	P	K	Mg	Ca	Fe	Zn	Mn	Cu	B
	mg kg^{-1}	%					mg kg^{-1}				
Control	0	2.27 e***	0.17 d***	1.25 k***	0.12 e***	1.85 d***	15.25 k***	21.34 l***	3.59 l ***	2.82 i***	4.85 e***
Calcium actosol	500	2.45 d	0.34 c	2.36 g	0.21 de	3.37 b	21.56 h	40.29 h	6.75 j	4.35 g	15.22 d
	1000	2.66 c	0.37 bc	3.29 d	0.36 ab	3.83 b	27.28 f	49.25 e	9.72 g	6.05 c	22.38 d
	3000	3.17 b	0.34 c	3.34 d	0.37 ab	4.20 a	51.04 a	58.80 a	18.05 c	6.53 b	35.22 c
	5000	3.16 b	0.33 c	2.82 e	0.34 bc	4.70 a	50.11 b	47.67 f	18.13 c	6.06 c	40.92 c
Boron actosol	500	2.39 d	0.32 c	2.66 f	0.25 cd	2.29 c	13.20 l	38.45 i	6.15 k	4.20 h	24.70 d
	1000	2.70 c	0.35 c	3.77 b	0.37 ab	2.26 c	16.43 j	47.60 f	9.35 h	4.77 e	36.15 c
	3000	3.28 a	0.37 bc	3.89 a	0.45 a	2.25 c	29.04 e	51.27 d	20.97 a	6.53 b	63.72 b
	5000	3.15 b	0.36 bc	3.48 c	0.33 bc	2.10 c	29.10 e	55.00 c	20.18 b	6.07 c	89.17 a
Actosol	500	2.25 e	0.45 b	1.71 j	0.19 de	2.17 c	19.36 i	30.08 k	7.05 i	4.65 f	14.73 e
	1000	2.46 d	0.55 a	2.17 h	0.28 bcd	2.19 c	23.47 g	37.23 j	9.90 f	5.87 d	25.85 d
	3000	3.12 b	0.56 a	2.42 g	0.35 b	2.27 c	42.47 c	46.30 g	13.77 d	7.00 a	38.15 c
	5000	3.11 b	0.56 a	1.88 i	0.12 e	2.88 c	42.24 d	57.24 b	12.78 e	6.50 b	44.32 c

Means within column not followed by the same letter differ significantly by DMRT ($P < 0.001$).

Table 5. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on the nutrient content of cucumber fruits.

Applications	Concentration	N	P	K	Mg	Ca	Fe	Zn	Mn	Cu	B
	mg kg^{-1}	%					mg kg^{-1}				
Control	0	1.51 j ***	0.15 e***	0.54 g ***	0.23 abc ***	0.09 d***	10.95 m***	14.69 j ***	5.76 m***	3.83 l***	0.31 f***
Calcium actosol	500	2.26 h	0.54 d	0.56 g	0.27 ab	0.85 c	41.27 k	42.46 d	39.35 h	4.60 k	4.20 e
	1000	2.44 g	0.62 cd	0.52 g	0.29 a	0.87 b	70.55 g	47.04 c	49.35 g	4.72 j	7.18 d
	3000	2.33 h	0.82 b	0.48 g	0.30 a	1.99 a	90.75 e	62.48 a	52.76 f	6.42 i	14.52 c
	5000	2.15 i	0.66 c	0.36 h	0.29 a	0.99 b	80.61 f	61.30 b	55.08 e	7.40 h	24.90 b
Boron actosol	500	2.60 ef	0.56 d	0.90 f	0.29 a	0.27 c	141.37 c	15.92 i	55.97 d	11.46 a	5.02 e
	1000	3.24 c	0.68 c	1.16 d	0.32 a	0.29 c	151.33 b	12.04 k	67.18 c	10.36 b	9.71 d
	3000	4.33 a	1.01 a	1.15 d	0.29 a	0.32 c	177.38 a	10.00 l	68.13 b	9.86 c	45.76 a
	5000	3.03 d	0.83 b	1.00 e	0.29 a	0.33 c	123.95 d	8.42 m	70.48 a	8.05 g	50.90 a
Actosol	500	2.18 i	0.14 e	2.47 c	0.19 bcd	0.21 c	25.07 l	23.00 g	10.37 l	8.81 e	4.69 e
	1000	2.55 f	0.17 e	3.05 b	0.17 cd	0.26 c	47.46 j	24.72 f	11.38 k	8.87 e	5.10 e
	3000	3.69 b	0.18 e	4.43 a	0.15 cd	0.28 c	58.30 i	29.12 e	13.50 i	8.32 f	8.82 d
	5000	2.67 e	0.23 e	3.02 b	0.12 d	0.30 c	60.21 h	22.73 h	13.15 j	9.42 d	9.61 d

Means within column not followed by the same letter differ significantly by DMRT ($P < 0.001$).

Table 6. The effects of Ca actosol (humate), B actosol (humate) and actosol (humic acid) applications on the nutrient content of cucumber leaves.

Applications	Concentration	N	P	K	Mg	Ca	Fe	Zn	Mn	Cu	B
	mg kg^{-1}	%					mg kg^{-1}				
Control	0	1.86 h***	0.10 f***	0.54 i***	0.10 g***	1.85 e***	11.25 l***	11.34 m***	2.75 m***	3.13 l***	4.85 e***
Calcium actosol	500	2.87 cd	0.47 c	0.56 i	0.27 cde	3.37 c	41.27 d	42.46 h	39.35 d	4.60 j	15.22 d
	1000	2.97 b	0.59 b	0.52 i	0.29 cd	4.23 b	70.54 c	47.04 f	49.35 c	4.72 i	22.38 d
	3000	2.87 cd	0.75 a	0.48 i	0.30 c	4.82 a	90.75 a	62.48 c	52.76 b	6.42 e	35.22 c
	5000	2.77 e	0.62 b	0.36 j	0.29 cd	4.70 a	80.61 b	61.30 d	55.08 a	7.40 c	40.92 c
Boron actosol	500	2.37 f	0.41 cd	3.88 d	0.28 cd	2.29 c	13.41 k	43.48 g	6.25 l	4.17 k	24.70 d
	1000	2.78 de	0.44 cd	5.36 b	0.44 b	2.46 c	15.95 j	57.23 e	9.10 j	4.92 h	46.15 c
	3000	3.29 a	0.60 b	5.53 a	0.67 a	2.55 c	30.60 i	76.72 a	22.09 e	6.54 d	83.72 b
	5000	2.99 b	0.45 c	4.95 c	0.69 a	2.71 c	31.48 h	64.72 b	21.83 f	5.76 g	99.17 a
Actosol	500	1.92 h	0.25 e	2.75 h	0.15 fg	1.90 e	33.05 g	18.08 l	8.16 k	4.88 h	14.73 e
	1000	2.16 g	0.35 d	2.95 g	0.18 efg	2.00 d	35.57 f	33.42 k	12.10 i	6.25 f	25.85 d
	3000	2.82 de	0.36 d	3.55 e	0.22 cdef	2.27 c	40.47 e	38.20 i	15.18 h	8.32 a	38.15 c
	5000	2.95 bc	0.35 d	3.08 f	0.20 def	2.38 c	41.24 d	35.54 j	18.14 g	7.66 b	44.32 c

Means within column not followed by the same letter differ significantly by DMRT ($P < 0.001$).

Table 7. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on the chemical content of soil.

Applications	Concentration (mg kg^{-1})	pH	CaCO3 (%)	Organic matter (%)	Total N (%)	NH4-N (mg kg^{-1})	NO3-N (mg kg^{-1})	CEC (cmolc kg^{-1})
Control	0	7.20 e***	0.95 b***	1.47 f***	0.01 b***	55.00 m***	41.64 m***	18.57 m***
Calcium actosol	500	7.40 bc	1.12 a	1.77 d	0.01 b	76.00 j	72.00 j	24.10 j
	1000	7.33 cd	0.88 b	1.62 e	0.01 b	90.00 h	88.00 g	35.28 f
	3000	7.45 b	0.94 b	1.80 d	0.02 ab	102.00 d	140.00 c	40.70 d
	5000	7.57 a	1.04 a	2.00 b	0.03 a	121.00 c	158.00 a	45.20 a
Boron actosol	500	7.30 d	1.10 a	1.74 d	0.01 b	78.00 i	77.00 i	24.66 i
	1000	7.33 cd	0.95 b	1.77 d	0.01 b	96.00 f	98.00 f	36.88 e
	3000	7.35 cd	0.88 b	1.90 c	0.02 ab	125.00 b	135.00 d	42.40 c
	5000	7.35 cd	0.74 c	2.20 a	0.03 a	129.00 a	144.00 b	44.66 b
Actosol	500	7.40 bc	0.89 b	1.47 f	0.01 b	69.16 l	51.12 k	19.06 l
	1000	7.33 cd	0.60 d	1.11 h	0.01 b	74.62 k	44.88 l	23.96 k
	3000	7.45 b	0.74 c	1.50 f	0.02 ab	92.82 g	99.40 e	32.19 g
	5000	7.57 a	0.71 c	1.37 g	0.02 ab	100.32 e	80.58 h	30.70 h

Means within column not followed by the same letter differ significantly by DMRT ($P < 0.001$).

Table 8. The effects of Ca actosol (humate), B actosol (humate), and actosol (humic acid) applications on the nutrient content of soil.

Applications	Concentration	K	Ca	Mg	Na	P	Fe	Cu	Mn	Zn	B
	mg kg^{-1}		cmolc kg^{-1}					mg kg^{-1}			
Control	0	2.23 g***	14.14 m***	4.94 a***	0.40 ef***	6.94 l***	1.07 h***	2.80 h***	8.00 j***	1.19 j***	0.24 j***
Calcium actosol	500	2.52 e	28.00 e	3.29 e	0.60 c	13.00 j	2.50 d	3.22 f	11.00 h	2.36 f	0.45 i
	1000	2.42 f	33.00 c	3.45 d	0.63 bc	20.00 g	2.74 c	3.78 cd	16.00 c	2.76 d	1.24 h
	3000	2.94 bc	40.00 b	3.58 c	0.72 ab	28.00 c	2.90 b	4.20 b	17.00 b	3.95 ab	1.99 e
	5000	3.19 a	42.20 a	3.74 b	0.77 a	33.00 b	3.10 a	4.60 a	18.00 a	4.02 a	2.31 d
Boron actosol	500	2.44 ef	18.66 j	3.19 f	0.65 bc	14.00 h	2.66 c	3.20 f	12.00 g	2.66 e	1.35 fg
	1000	2.66 d	22.50 i	3.25 ef	0.68 abc	25.00 d	2.74 c	3.80 c	18.00 a	2.98 c	3.20 c
	3000	2.88 c	27.63 f	3.58 d	0.72 ab	33.00 b	2.90 b	4.20 b	17.00 b	3.87 b	3.90 b
	5000	2.97 b	30.62 d	3.74 b	0.77 a	38.00 a	3.10 a	4.60 a	18.00 a	3.92 b	4.10 a
Actosol	500	2.09 h	15.38 l	2.99 h	0.43 e	10.28 k	2.08 f	2.84 h	10.01 i	1.68 h	1.31 gh
	1000	1.65 i	17.97 k	2.86 i	0.32 f	13.58 i	1.87 g	3.02 g	13.27 f	1.41 i	1.43 f
	3000	2.44 ef	24.34 h	3.26 ef	0.51 d	22.15 f	2.41 e	3.70 de	15.47 d	2.80 d	2.02 e
	5000	2.18 g	24.46 g	3.10 g	0.39 ef	22.41 e	2.12 f	3.67 e	14.93 e	2.05 g	1.97 e

Means within column not followed by the same letter differ significantly by DMRT ($P < 0.001$).

content in ryegrass (Bidegain et al., 2000) and increased root fresh and dry weight of tomato and eggplant seedlings (Dursun et al., 1999).

An increased mineral nutrient uptake with HA and Ca in different plant parts is also reported by other researchers (Çelik et al., 2008; El-Nemer et al., 2012; Kazemi, 2013). Our results are in confirmation of those of Tattini et al. (1990), who stated that the uptake of macro- and microelements can be accelerated by humates. Fernández-Escobar et al. (1999) found stimulated accumulation of K, B, Ca, and Fe in leaves with the application of HA. Kazemi (2013) reported highest leaf N and K content (2.61% and 3.4%, respectively) with 20 ppm HA + 10 mM Ca. Similarly, Smolen and Sady (2009) reported that Ca application caused an increase in the concentration of total N in carrot compared with the control plants. Unlu et al. (2011) reported a significant effect of foliar- and soil-applied HA on the fruit quality and yield of cucumber in a greenhouse experiment. In addition, some other researchers reported that foliar sprays of humic substances also increase growth, yield, and quality in a number of crop species via increased nutrient uptake (Yildirim, 2007; Karakurt et al., 2009), serving as a source of mineral plant nutrients and regulating their release throughout the growth period (Chen and Aviad, 1990; Atiyeh et al., 2002).

The increase in nutrient concentration in response to HA is probably due to the ability of humic substances to stimulate microbiological activity (Mayhew, 2004), increasing cell membrane permeability and enhancing water and nutrient uptake (Sibanda and Young, 1986; Valdrighi et al., 1996). Moreover, some researchers attributed the positive effects of HA to its influence on plant roots (Atiyeh et al., 2002; Türkmen et al., 2004) and on the metabolism of soil microbial population and soil physical conditions (Chen and Aviad, 1990; Muscolo et al., 1999; Zandonadi et al., 2007). High concentrations of HA improve root growth in hydroponic systems and cause an increase in root volume, which may be due to easier absorption and more efficient nutrients. It is likely that an increase in nutrient uptake by plants can be particularly associated with an increase in the root growth.

Studies of B showed that it is significant in the production and quality of tomato and is required by crop plants for cell division, nucleic acid synthesis, uptake of calcium, and transport of carbohydrates (Bose and Tripathi, 1996). B plays a vital role in flowering and fruit formation (Nonnecke, 1989) and its deficiency affects the translocation of sugar, starches, nitrogen, and phosphorus and the synthesis of amino acids and proteins (Stanley et al., 1995). In a different study, it was reported that B application resulted in increased Brussels sprout yield and tissue Fe, Cu, K, P, Mn, and Zn contents, indicating B deficiency. Earlier studies determined that B addition of 6 kg ha^{-1} was enough to elevate soil B levels (Turan et al., 2009).

Tomato and cucumber are considered to be the most important vegetable crops and their global consumption is very high. Therefore, there must be an adequate amount of Ca and B in these fruits. Ca and B are immobile elements in plants, yet Ca actosol and B actosol significantly translocated the nutrients from leaves to fruits both in tomato and cucumber when compared to the control (Figures 7 and 8). This could be due to the fact that humic substances are assumed to have specific importance for the transport and availability of micro- and macroelements in the plants (El-Nemer et al., 2012; Kazemi, 2013). It has also been reported that humic substances in soil provide the compounds that affect root growth and the distribution of nutrients absorbed by plants (Chen and Aviad, 1990).

4.3. Soil chemical properties and nutrient status

Postharvest soil analysis showed that Ca actosol, B actosol, and actosol applications significantly affected soil chemical properties, as well as soil nutrient status, when compared to the control. The increase in soil macro- and micronutrient status with HA application was also reported by other researchers (Sharif et al., 2002; Tahir et al., 2011). The application of HA at the rate of 20 kg ha^{-1} resulted in an increase in soil N content from 28% to 29% (Tenshia et al., 2005). The increase in soil N content with different HA treatments observed in this study may be attributed to the inhibition of urease activity by HA (Vaughan and Ord, 1991), resulting in reduced N losses by volatilization (Flaig, 1984) and thereby in increased N content of the soil.

A significant increase in soil available P recorded in this study is in conformity with the results of Sharif et al. (2002), who reported 31% increase in soil P status with HA application over the control. Humic acid has the potential to reduce P fixation and solubilize insoluble P, thus increasing P concentration in the soil (Sibanda and Young, 1986). A study conducted in a greenhouse by Bermudez et al. (1993) showed 8% –24% increase in soil P availability due to reduced P fixation by HA.

Likewise, increased available soil K with HA applications documented in our results is in accordance with the findings of Tahir et al. (2011) and Tenshia et al. (2005). This increase in available K might be due to reduced K fixation along with the release of fixed K by HA (Tan, 1978). Olk and Cassman (1995) reported reduced K fixation that resulted in increased K availability by applying HA in vermiculite soils. Increased micronutrient (Fe, Mn, and Zn) status of soil with HA was also reported by Samir and Sengupta (1985).

Increase in HA concentration resulted in a significant increase in total marketable yield and fruit weight as compared to the control. However, as the concentration of

HA increased further, the total marketable yield and fruit weight decreased, especially in response to 5000 mg of HA application. As regards the nutrient contents in different plant parts (leaf and fruit), the highest values were recorded mostly for 3000 mg kg^{-1}, and concentrations beyond this rate did not prove very effective. On the other hand, soil chemical and nutrient contents were found to be higher after the application rate of 5000 mg kg^{-1}. It has been pointed out that the characteristic growth-response curves as a consequence of exposing plants to humic substances display a progressive increase in growth with the increase in the concentration of humic substances; however, there is usually a decrease in growth at higher concentrations of the humic materials (Chen and Aviad, 1990; Zandonadi et al., 2007).

The results conclude that the application of HA with Ca and B might be efficiently utilized in obtaining maximum fruit yield and in stimulating the nutrient contents and Ca and B translocation of tomato and cucumber. The applications also upgraded the health of the soil by modifying soil chemical characteristics and enhancing its nutrient status. These nutrient sources could offer an economical and simple application for tomato and cucumber plants grown under greenhouse conditions.

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