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Original Research

Alleviating Effect of Bat Guano Against Negative Response of *Begonia semperflorens* to Different Saline Environments

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Abstract

This study was conducted to reveal the effects of bat guano on the growth, quality, content of photosynthetic pigments and nutrients of *Begonia semperflorens* L. under salt-free conditions, to determine the effects of salt stress on the related species, and to figure out whether there is an alleviating effect of bat guano on salt stress. For this purpose, 40 g (50 t ha^{-1}) bat guano per pot was used at various sodium chloride (NaCl) applications, including 0, 20, 40, 50, and 60 mM. The experiment was conducted in plastic pots with a volume of 2 L containing the peat+perlite mixture (3:1) by using a completely randomized experimental design with five replications. Bat guano had effects on all studied traits at a 0.001 probability level without Chl *a/b*. This study revealed that the contents of Chl *a*, Chl *b,* and the concentrations of all nutrients in leaves of *B. semperflorens* highly decreased at 60 mM NaCl compared to control treatments. Bat guano treatment without NaCl increased over 25% root fresh and dry weights. Bat guano was used to improve the effects of salt stress on *B. semperflorens*.

Keywords: begonia, bat guano, photosynthetic, salt stress, organic material

Introduction

Ornamental plants play a major role in horticulture owing to using for different purposes such as gardening, landscaping, and cut flowers. The production of these plants necessitates high-water consumption albeit the reduction of freshwater [1]. In this sense, irrigation with salt water can be an alternative yet it is important to know how the salinity affects ornamental plants [1, 2]. Salt stress is one of the most important negative abiotic drivers [3-6]. It has adverse effects on growth [4, 6], photosynthesis [1], nutrient content [3], quality, and enzymatic activity. Some physiological mechanisms can mitigate these effects in plants but sometimes not enough, especially for the aesthetic appearance of ornamental plants [1].

One of the methods to improve the effect of salt stress on plants in recent years is the use of organic materials. Bat guano, one of the important organic materials, enhances the aeration, porosity, infiltration and water-holding capacity of soils thanks to its microbes with bioremediation capabilities [7]. It is widely used as a fertilizer in agriculture and horticulture due to its inclusion of too much nitrogen and phosphorous [8-10].

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Begonia semperflorens L. is the most common species within the Begonia genus. It has a resistance to drought and moisture [11]. It is an ornamental plant growing in shady and sunny areas, and blooming continuously [12]. Until now, some studies have been carried out not only on the cultivation of ornamental plants with different organic materials [13-16] but also on the usage of organic materials on alleviating salt stress in ornamental plants [17-20]. However, there is no study used organic materials to improve salt stress in begonia species. The purposes of this study are (i) to reveal the effects of bat guano on the growth, quality, contents of photosynthetic pigments and nutrients *B. semperflorens* under salt-free conditions, (ii) to determine the effects of salt stress on the related species and (iii) to figure out whether there is an alleviating effect of bat guano on salt stress.

Materials and Methods

Greenhouse Experiment and Growth Conditions

The study was carried out in the Research and Practice Greenhouse of Çankırı Karatekin University (40°37′32″N, 33°36′30″E; 884 m asl). *B. semperflorens* was selected as the test plant. Its seedlings were grown under 450-500 µmol m^{-2} s^{-1} light intensity at 30-35ºC with 65-70% of relative humidity in the greenhouse.

Ready-to-use bat guano was provided by private companies. pH, electrical conductivity (EC), humidity, total humic + fulvic acid [15, 21], organic matter (OM) and total nitrogen (N) of bat guano were 5.00, 7.3 dS m^{-1}, 20%, 50 mg g^{-1}, 300 mg g^{-1}, and 15 mg g^{-1}, respectively.

The experiment was conducted in plastic pots with a volume of 2 liters containing the peat+perlite mixture (3:1) by using a completely randomized experimental design with five replications. pH, EC, organic material [15, 21] and extractable ammonium (NH_4)-N, nitrate (NO_3)-N, phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg) and iron (Fe) [21] of the peat+perlite mixture in the experiment were noted as 6.0, 0.5 dS m^{-1}, 950 mg g^{-1}, 0.7 µg g^{-1}, 5.8 µg g^{-1}, 5.5 µg g^{-1}, 51 µg g^{-1}, 178 µg g^{-1}, 56 µg g^{-1} and 0.1 µg g^{-1}, respectively. No plant nutrients were used in the experiment. 40 g (50 t ha^{-1}) bat guano per pot was used at various NaCl applications including 0, 20, 40, 50, and 60 mM.

Harvest and Plant Analysis

Aesthetic appearance, number of main shoots, buds and flowers, flower weight, and crown width (cm) as quality traits were recorded. For the aesthetic appearance of each plant, the plant condition, its appearance, its representation in the pot, vegetative component structure, its vitality, and brightness were assessed by a 5-member jury, with a score between 1 and 10 was used to assess plant condition, appearance, representation in the pot, vegetative component structure, its vitality and brightness and aesthetic appearance of each plant before harvesting. Here, a final rating of 1-4, 5, and 6-10 are considered as slightly sensitive, moderately sensitive, and tolerant to salinity, respectively. Crown width was measured as the projection diameter of the plant crown vertically in two directions [19, 21, 22].

Growth traits including shoot length (sL, cm), shoot fresh weight (sFW, g), shoot dry weight (sDW, g), root length (rL, cm), root fresh weight (rFW, g), and root dry weight (rDW, g) were measured after 12 weeks of sowing. After harvesting, plants were washed with tap and deionized water to avoid soil-induced contamination. The harvested plants were desiccated at 70±5ºC for 72 h to determine shoot and root dry weights [15, 19, 21].

The fresh leaf samples (250 mg) were homogenized in 10 mL of acetone (90%, v/v). The absorbance of the photosynthetic pigments was determined at 663, 645, and 470 nm using a spectrophotometer (UV/VIS-1201, Shimadzu Corp., Kyoto, Japan). The concentrations of chlorophylls (Chl) and carotenoids (Car) were calculated following [15, 21].

Fresh leaf samples (250 mg) were homogenized in 10 mL of 3% sulphosalicylic acid and then filtered through Whatman No 2-filter paper. The proline was detected from the obtained extract based on the method of [15, 19, 21].

Lipid peroxidation of plants determined at 532 and 600 nm by a spectrophotometer [22]. Then, lipid peroxidation values were calculated as malondialdehyde (MDA) by the following equation.

$$MDA\ (nmol\ mL^{-1}) = [(A_{532}-A_{600}) / 155000] \times 10^6 \quad (1)$$

To determine the amount of H_2O_2, fresh leaf samples were measured at 415 nm by a spectrophotometer [23-24]. The conversions of the measured values were calculated with the standard curve prepared with H_2O_2.

After harvesting, plant samples were dried in a plant drying cabinet with air circulation at 65-70ºC until constant weight. The dried leaf samples were grounded using the dry-ashing method in a muffle furnace at 500ºC for 6 hours, then dissolved in 10 mol L^{-1} nitric acid (HNO_3), and extracts were finally filtered and stored in plastic jars for nutrient analysis [15, 19, 21]. The concentrations of P, K, Ca, Mg, sodium (Na), copper (Cu), manganese (Mn), Fe and zinc (Zn) were detected by ICP-OES (Optima®2100 DV, Perkin-Elmer, Waltham, MA, USA). Nitrogen concentration was determined following [15, 21].

Statistical Analysis

Kolmogorov-Smirnov test was used to determine whether data shows the normal distribution. As having a normal distribution, parametric methods were performed for the analyses. Firstly, the mean, standard error, *F* ratio, and probability (P-value) of all

the traits were calculated. Moreover, analysis of variance (ANOVA) was used to reveal whether the means of the related traits are different among treatments. If there is a difference, homogenic and different treatments were determined by Duncan's multiple range tests (α: 0.05). All analyses were performed using SPSS statistical program.

Results and Discussion

Bat guano (BG) had effects on all quality traits at a 0.001 probability level. In current study, maximum and minimum mean values for quality traits resulted from the treatments of control+bat guano (C+BG) and 60 mM NaCl, respectively. Aesthetic appearance, the number of main shoots, the number of buds, the number of flowers, flower weight, and crown width were reduced by 35.6%, 51.8%, 56.0%, 50.5%, 44.2%, and 32.5%, respectively, at the highest NaCl level (60 mM) compared to control (non-saline). The number of buds (>30%) and aesthetic appearance (<10%) in bat guano and salt treated plants had higher and lower increase rates than only salt treated plants when compared to the rates of increase in other quality traits, respectively (Table 1).

ANOVA revealed significant differences among treatments with and without bat guano for growth traits at 0.001 probability level. C+BG and 60 mM NaCl resulted in maximum and minimum mean values for growth traits, respectively. Shoot length, shoot fresh and dry weight, root length, root fresh and dry weight were decreased by 44.4%, 28.2%, 29.4%, 42.5%, 37.2%, and 48.8%, respectively, at the highest NaCl level (60 mM) over control (non-saline). Shoot length (>20%) and shoot dry weight (<10%) in bat guano and salt treated plants had higher and lower increase rates than only salt treated plants when compared to the rates of increase in other growth traits, respectively (Table 2).

Significant differences were found among treatments with and without bat guano for the contents of photosynthetic pigments, proline, MDA, and H_2O_2 apart from Chl *a/b* ratio at 0.001 probability level (Table 3 and 4). C+BG showed maximum mean contents of Chl *a*, Chl *b*, Chl *a*+*b*, Chl *a*+*b*/Car. Chl *a*, Chl *b*, Car, Chl *a*+*b*, Chl *a/b* and Chl *a*+*b*/Car were decreased by 39.2%, 35.4%, 26.5%, 38.1%, 5.6% and 15.6%, respectively; however, the contents of proline, MDA and H_2O_2 were increased by 4.22-, 1.71- and 2.39-fold, respectively, at the highest NaCl level (60 mM) compared to control (non-saline). Chl *a* content (>20%) and Chl *a/b* ratio (<10%) in bat guano and salt treated plants had higher and lower increase rates than only salt treated plants when compared to the rates of increase in other contents, respectively (Table 3 and 4).

Bat guano affected on the concentrations of all plant nutrients at a 0.001 probability level. C+BG and 60 mM NaCl resulted in the maximum and minimum mean values for plant nutrients, respectively. The concentrations of N, P, K, Ca, Mg, Fe, Cu, Zn and Mn were decreased by 31.2%, 54.4%, 29.3%, 52.3%, 49.3%, 38.7%, 51.2%, 44.7%, and 40.6%, respectively; however, the concentration of Na was increased by 5.69-fold at the highest NaCl level (60 mM) compared to control (non-saline). Besides, the concentrations of N, P, K, Ca, Mg, Fe, Cu, Zn and Mn increased by 1.37-, 2.79-, 1.79-, 2.00-, 1.92-, 1.86-, 1.50-, 1.86-, and 1.33-fold, respectively at the treatment of control+bat guano (C+BG) compared to the treatment of control (non-bat guano). The concentrations of Cu and Zn (>30%) and the concentrations of other plant nutrients (<20%) in bat guano and salt-treated plants had higher and lower increase rates than only salt treated plants

Table 1. Effects of NaCl concentration and bat guano on quality traits.

Treatments	Aesthetic appearance score (1-10)	Number of main shoots	Number of buds	Number of flowers	Flower weight (g)	Crown width (cm)
C	[#]$9.0^{\pm0.3}$ab	$11.2^{\pm0.6}$b	$15.0^{\pm0.3}$b	$18.2^{\pm0.6}$b	$7.62^{\pm0.24}$b	$20.63^{\pm0.41}$b
C+BG	$9.6^{\pm0.3}$a	$13.4^{\pm0.7}$a	$18.2^{\pm0.7}$a	$21.4^{\pm0.8}$a	$8.45^{\pm0.18}$a	$22.87^{\pm0.59}$a
20 mM NaCl	$8.4^{\pm0.3}$bc	$9.0^{\pm0.6}$cd	$11.6^{\pm0.5}$c	$15.6^{\pm0.5}$cd	$6.41^{\pm0.22}$c	$17.96^{\pm0.63}$cd
40 mM NaCl	$7.6^{\pm0.4}$cd	$6.4^{\pm0.5}$fgh	$8.4^{\pm0.5}$de	$11.8^{\pm0.6}$e	$5.04^{\pm0.19}$de	$15.77^{\pm0.74}$ef
50 mM NaCl	$6.2^{\pm0.4}$e	$5.6^{\pm0.5}$gh	$7.8^{\pm0.6}$ef	$10.6^{\pm0.7}$ef	$4.51^{\pm0.23}$ef	$14.61^{\pm0.62}$fg
60 mM NaCl	$5.8^{\pm0.4}$e	$5.4^{\pm0.5}$h	$6.6^{\pm0.5}$f	$9.0^{\pm0.5}$f	$4.25^{\pm0.11}$f	$13.93^{\pm0.54}$g
20 mM NaCl+BG	$8.6^{\pm0.3}$abc	$10.6^{\pm0.7}$bc	$14.8^{\pm0.4}$b	$16.4^{\pm0.7}$c	$7.27^{\pm0.17}$b	$19.18^{\pm0.52}$bc
40 mM NaCl+BG	$8.0^{\pm0.3}$bc	$8.2^{\pm0.7}$de	$12.4^{\pm0.5}$c	$14.0^{\pm0.5}$d	$5.30^{\pm0.20}$d	$17.91^{\pm0.54}$cd
50 mM NaCl+BG	$6.4^{\pm0.5}$e	$8.0^{\pm0.5}$def	$12.8^{\pm0.6}$c	$13.8^{\pm0.6}$d	$5.33^{\pm0.25}$d	$17.37^{\pm0.46d}$e
60 mM NaCl+BG	$6.8^{\pm0.2}$de	$7.2^{\pm0.5}$efg	$9.6^{\pm0.5}$d	$11.2^{\pm0.7}$e	$5.02^{\pm0.13d}$e	$16.16^{\pm0.61}$ef
F value	14.9^{***}	20.9^{***}	50.5^{***}	37.9^{***}	53.2^{***}	22.9^{***}

[#]: Different letters in the columns indicate that the means of treatments are significantly different in terms of the related trait ($P<0.05$). C: Control, BG: Bat guano.

Table 2. Effects of NaCl concentration and bat guano on growth traits.

Treatments	sL (cm)	sFW (g)	sDW (g)	rL (cm)	rFW (g)	rDW (g)
C	#22.02±0.57b	129.6±1.8b	6.74±0.22ab	19.32±0.75b	19.01±0.4b	1.612±0.030b
C+BG	26.11±0.44a	141.2±1.9a	6.94±0.24a	23.82±1.40a	25.16±1.3a	2.164±0.120a
20 mM NaCl	17.45±0.76d	119.2±1.7c	6.20±0.19bcd	16.62±0.68cd	16.21±0.4cde	1.359±0.030bcd
40 mM NaCl	13.95±0.81e	104.6±2.2e	5.44±0.24ef	14.66±0.85d	14.81±0.4def	1.233±0.040cd
50 mM NaCl	13.86±0.31e	100.4±1.1e	5.71±0.17def	14.74±0.36d	14.19±0.5ef	1.190±0.070cd
60 mM NaCl	12.25±0.56e	93.0±3.1f	4.76±0.30g	11.10±0.61e	11.94±0.7g	0.826±0.190e
20 mM NaCl+BG	19.88±0.57c	127.9±1.2b	6.49±0.26abc	18.94±1.02bc	18.07±0.9bc	1.526±0.080b
40 mM NaCl+BG	17.96±0.51d	123.8±2.1bc	6.36±0.21abcd	17.98±0.76bc	16.97±0.9bcd	1.428±0.080bc
50 mM NaCl+BG	17.86±0.48d	112.5±2.3d	5.98±0.11cde	17.60±0.61bc	15.77±0.7de	1.457±0.070bc
60 mM NaCl+BG	16.20±0.70d	103.3±2.5e	5.05±0.25fg	16.88±0.58bcd	13.52±0.4fg	1.117±0.040d
F value	49.7***	54.7***	10.3***	17.5***	26.0***	15.6***

#: Different letters in the columns indicate that the means of treatments are significantly different in terms of the related trait (P<0.05). C: Control, BG: Bat guano, sL: shoot length, sFW: shoot fresh weight, sDW: shoot dry weight, rL: root length, rFW: root fresh weight, rDW: root dry weight.

Table 3. Effects of NaCl concentration and bat guano on contents of photosynthetic pigments.

Treatments	Chl *a*	Chl *b*	Car	Chl *a*+*b*	Chl *a/b*	Chl a+b/Car
	(mg g^{-1} FW)					
C	#0.480±0.016c	0.192±0.006bc	0.245±0.008bc	0.672±0.019c	2.51±0.081	2.76±0.015a
C+BG	0.687±0.015a	0.265±0.013a	0.278±0.007a	0.952±0.024a	2.62±0.126	3.43±0.029bc
20 mM NaCl	0.450±0.016cd	0.176±0.005cd	0.230±0.007cd	0.626±0.021cd	2.56±0.058	2.73±0.023bc
40 mM NaCl	0.392±0.004e	0.162±0.006d	0.220±0.003d	0.554±0.009e	2.43±0.067	2.52±0.011cd
50 mM NaCl	0.342±0.006f	0.140±0.005e	0.192±0.006e	0.482±0.010f	2.45±0.055	2.51±0.015cd
60 mM NaCl	0.292±0.009g	0.124±0.005e	0.180±0.007e	0.416±0.009g	2.37±0.129	2.33±0.010d
20 mM NaCl+BG	0.550±0.015b	0.210±0.005b	0.254±0.008b	0.760±0.015b	2.63±0.101	3.00±0.015b
40 mM NaCl+BG	0.472±0.014c	0.180±0.008cd	0.234±0.009bcd	0.652±0.021c	2.64±0.101	2.80±0.025bc
50 mM NaCl+BG	0.432±0.013d	0.166±0.005d	0.220±0.007d	0.598±0.017de	2.61±0.063	2.73±0.021bc
60 mM NaCl+BG	0.388±0.013e	0.160±0.005d	0.216±0.005c	0.548±0.016e	2.43±0.077	2.54±0.018cd
F value	75.5***	34.0***	17.7***	81.80***	1.20ns	9.60***

#: Different letters in the columns indicate that the means of treatments are significantly different in terms of the related trait (P<0.05). C: Control, BG: Bat guano. Chl *a*: chlorophyll *a*, Chl *b*: chlorophyll *b*, Car: carotenoid, Chl *a*+*b*: chlorophyll *a*+chlorophyll *b*, Chl *a/b*: chlorophyll *a*/chlorophyll *b*, Chl *a*+*b*/Car: chlorophyll *a*+*b*/carotenoid.

when compared to the rates of increase in other nutrients, respectively. In addition, C+BG) treatment increased all nutrients to critical values (Table 5 and 6).

Bat guano-supplemented salt concentrations improved the harmful effects of NaCl on the quality, growth and contents of photosynthetic pigments and the concentration of nutrients of *B. semperflorens*. All studied traits had higher values with increasing bat guano and lower values with increasing NaCl concentration.

Quality traits of *B. semperflorens* showed an overall reduction of over 30% at 60 mM NaCl compared to control treatments. Similarly, the flower weight of *Tagetes patula* decreased at a salinity of 100 mM NaCl [25]. Bat guano treatment without NaCl increased all quality traits in the present study. Although the number

Table 4. Effects of NaCl concentration and bat guano on content of proline, MDA and H_2O_2.

Treatments	Proline (mmol kg^{-1} FW)	MDA (nmol g^{-1} FW)	H_2O_2 (nmol g^{-1} FW)
C	$^{\#}0.119^{\pm0.001}$f	$7.01^{\pm0.15}$f	$4.87^{\pm0.13}$h
C+BG	$0.116^{\pm0.003}$f	$6.85^{\pm0.17}$f	$5.11^{\pm0.14}$h
20 mM NaCl	$0.233^{\pm0.006}$d	$7.98^{\pm0.11}$e	$7.17^{\pm0.13}$f
40 mM NaCl	$0.309^{\pm0.009}$c	$9.72^{\pm0.24}$cd	$8.82^{\pm0.14}$d
50 mM NaCl	$0.422^{\pm0.011}$b	$11.04^{\pm0.10}$b	$10.86^{\pm0.16}$b
60 mM NaCl	$0.502^{\pm0.007}$a	$11.97^{\pm0.29}$a	$11.62^{\pm0.26}$a
20 mM NaCl+BG	$0.189^{\pm0.005}$e	$7.13^{\pm0.09}$f	$6.34^{\pm0.21}$g
40 mM NaCl+BG	$0.252^{\pm0.011}$d	$8.24^{\pm0.11}$e	$7.40^{\pm0.26}$f
50 mM NaCl+BG	$0.329^{\pm0.012}$c	$9.49^{\pm0.17}$d	$8.17^{\pm0.20}$e
60 mM NaCl+BG	$0.410^{\pm0.010}$b	$10.04^{\pm0.15}$c	$9.90^{\pm0.18}$c
F value	252.0^{***}	111.2^{***}	153.1^{***}

$^{\#}$: Different letters in the columns indicate that the means of treatments are significantly different in terms of the related trait ($P < 0.05$). C:Control, BG: Bat guano, MDA: malondialdehyde.

Table 5. Effects of NaCl concentration and bat guano on concentrations of nitrogen, phosphorus and alkaline metal-nutrients.

Treatments	N	P	K	Ca	Mg	Na
	($\mu g\ g^{-1}$)					
C	$^{\#}29.30^{\pm0.07}$c	$4.34^{\pm0.05}$c	$16.86^{\pm0.04}$c	$8.42^{\pm0.03}$b	$2.80^{\pm0.01}$b	$2.98^{\pm0.01}$g
C+BG	$40.18^{\pm0.136}$a	$12.12^{\pm0.04}$a	$30.10^{\pm0.05}$a	$16.84^{\pm0.03}$a	$5.38^{\pm0.02}$a	$3.26^{\pm0.01}$g
20 mM NaCl	$27.42^{\pm0.10}$cd	$3.60^{\pm0.02}$cd	$14.86^{\pm0.04}$d	$6.24^{\pm0.03}$d	$2.30^{\pm0.01}$c	$9.88^{\pm0.02}$de
40 mM NaCl	$26.16^{\pm0.13}$de	$2.80^{\pm0.02}$def	$13.44^{\pm0.07}$e	$5.38^{\pm0.03}$ef	$2.02^{\pm0.01}$cd	$12.20^{\pm0.02}$c
50 mM NaCl	$24.42^{\pm0.11}$e	$2.60^{\pm0.01}$ef	$13.20^{\pm0.025}$e	$4.84^{\pm0.03}$f	$1.72^{\pm0.01}$de	$14.98^{\pm0.02}$b
60 mM NaCl	$20.16^{\pm0.05}$f	$1.98^{\pm0.01}$f	$11.92^{\pm0.035}$f	$4.02^{\pm0.02}$g	$1.42^{\pm0.01}$e	$16.96^{\pm0.023}$a
20 mM NaCl+BG	$38.40^{\pm0.03}$a	$6.80^{\pm0.07}$b	$20.80^{\pm0.048}$b	$7.58^{\pm0.04}$c	$3.00^{\pm0.02}$b	$7.34^{\pm0.02}$f
40 mM NaCl+BG	$32.54^{\pm0.08}$b	$3.76^{\pm0.02}$cd	$15.50^{\pm0.033}$d	$6.34^{\pm0.03}$d	$2.28^{\pm0.01}$c	$9.58^{\pm0.02}$e
50 mM NaCl+BG	$28.80^{\pm0.13}$cd	$3.22^{\pm0.02}$de	$14.84^{\pm0.024}$d	$5.84^{\pm0.02}$de	$1.96^{\pm0.01}$cd	$10.42^{\pm0.03}$d
60 mM NaCl+BG	$24.48^{\pm0.07}$e	$2.54^{\pm0.01}$ef	$14.28^{\pm0.036}$de	$5.36^{\pm0.02}$ef	$1.90^{\pm0.01}$d	$11.90^{\pm0.04}$c
F value	44.0^{***}	91.0^{***}	156.1^{***}	196.6^{***}	90.5^{***}	397.5^{***}
Critical range*	35-60	3-8	25-60	10-30	0-8	0-20

$^{\#}$: Different letters in the columns indicate that the means of treatments were significantly different in terms of the related trait ($P<0.05$). C: Control, BG: Bat guano. *Apart from Na [23], critical ranges of macro-nutrients were provided from Jones et al. [24].

of flowers increased slightly (15%) with bat guano in this study, hazelnut husk, an organic material, increased 56% in *Salvia splendens* [15] and 34% in *Viola tricolor* [16]. Akintoye et al. [26] also reported that *B. erythrophylla* planted in topsoil + poultry manure (4:1) produced maximum number of flowers per plant.

Plant growth is inhibited by salt stress due to decreased photosynthesis [6]. In the current study, the growth of *B. semperflorens* reduced by up to 30% at 60 mM NaCl compared to control treatments. Root dry weight and shoot length decreased under salt stress. Roots are the most vulnerable part of the plant as they are directly exposed to the salt [27]. Ornamental plants grown under saline conditions exhibited reduced plant weight and height [28]. Bat guano treatment without NaCl increased over 25% root fresh and dry weights

Table 6. Effects of NaCl concentration and bat guano on concentrations of heavy metal-nutrients.

Treatments	Fe	Mn	Zn	Cu
	($\mu g\ g^{-1}$)			
C	$^{\#}111^{\pm 2.9}$d	$52.28^{\pm 1.33}$c	$12.96^{\pm 0.96}$d	$4.55^{\pm 0.23}$b
C+BG	$207^{\pm 5.7}$a	$69.71^{\pm 2.57}$a	$24.08^{\pm 1.25}$a	$6.82^{\pm 0.33}$a
20 mM NaCl	$90^{\pm 2.4}$f	$47.04^{\pm 1.51}$d	$11.63^{\pm 0.48}$de	$3.67^{\pm 0.18}$d
40 mM NaCl	$81^{\pm 2.1}$g	$40.69^{\pm 1.57}$ef	$10.58^{\pm 0.49}$ef	$2.96^{\pm 0.13}$e
50 mM NaCl	$74^{\pm 1.7}$gh	$36.84^{\pm 0.94}$f	$8.74^{\pm 0.49}$fg	$2.71^{\pm 0.18}$ef
60 mM NaCl	$68^{\pm 1.8}$h	$31.03^{\pm 0.90}$g	$7.17^{\pm 0.57}$g	$2.22^{\pm 0.11}$f
20 mM NaCl+BG	$142^{\pm 2.8}$b	$63.55^{\pm 2.28}$b	$16.67^{\pm 0.50}$b	$5.14^{\pm 0.30}$b
40 mM NaCl+BG	$121^{\pm 1.9}$c	$55.29^{\pm 0.70}$c	$15.40^{\pm 0.42}$bc	$4.63^{\pm 0.24}$b
50 mM NaCl+BG	$101^{\pm 1.6}$e	$47.46^{\pm 1.05}$d	$15.29^{\pm 0.34}$bc	$4.46^{\pm 0.25}$bc
60 mM NaCl+BG	$93^{\pm 2.0}$f	$41.83^{\pm 1.93}$e	$13.56^{\pm 0.37}$cd	$3.85^{\pm 0.14}$cd
F value	225.6^{***}	56.5^{***}	53.9^{***}	37.1^{***}
Critical range*	50-200	50-200	25-200	7-30

$^{\#}$: Different letters in the columns indicate that the means of treatments were significantly different in terms of the related trait ($P<0.05$). C: Control, BG: Bat guano.* Critical ranges of micro-nutrients were provided from Jones et al. [24].

in the present study. *B. semperflorens* grown in 25% green waste compost showed maximum dry weight [29]. In contrast to these results, Crutchfield et al. [30] stated that biochar did not affect the growth of *B. semperflorens*. Shetty et al. [8] reported that bat guano was required in minute quantity to improve plant growth.

Earlier studies reported a decrease in chlorophyll content of ornamental plants grown under saline conditions [1]. Similarly, the present study showed maximum reduction in the contents of Chl *a* and Chl *b* at 60 mM NaCl compared to control treatments. Chrysargyris et al. [25] found that salinity increased total carotenoids in *Tagetes patula*. In contrast to the contents of proline, MDA and H_2O_2 showed markedly the effect of bat guano on salt stress in the plant.

Ornamental plants grown under saline conditions had a decrease of the concentrations of N, P, K and Ca concentration [1]. Similarly, this study revealed that all nutrients in leaves of *B. semperflorens* decreased over 50% at 60 mM NaCl compared to control treatments. As considered control and C+BG together, bat guano mainly contributed nutrients to the plant. But this contribution was especially much higher in terms of P (2.79-fold) and Ca (2.00-fold). Jayasvasti and Jayasvasti [9] reported that bat guano was rich in P content. A study [31] conducted to determine several organic materials on the growth of *Begonia eliator* also showed that the concentrations of nutrients were higher in media containing coco peat and tea waste than the control treatment.

Conclusions

Bat guano had effects on all studied traits at a 0.001 probability level without Chl *a/b*. This study revealed that the contents of Chl *a*, Chl *b*, and the concentrations of all nutrients in leaves of *B. semperflorens* highly decreased at 60 mM NaCl compared to control treatments. Bat guano was an alternative as suitable organic material as it was used to improve quality, growth, contents of photosynthetic pigments, and nutrients of *B. semperflorens* under salt stress. It also enhanced the nutrient content to the critical ranges. The conclusions of the study suggested regular application of bat guano, rich in nutrients, to alleviate the detrimental aspects of salinity in salinized soils of arid and semi-arid areas.

Availability of Data and Material

The author confirms that the data supporting the findings of this study are available within the article.

Conflict of Interest

The authors declare no conflict of interest.

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