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ORIGINAL PAPER

Humic acid enhances growth, development, and nutrient absorption in the Zambesi lily cultivar

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Abstract

Humic acid, an environmentally friendly substance, directly or indirectly affects plant growth, and it is vital in various biological, physical, and chemical processes. In this study, humic acid was applied to lily plants at two-week intervals after plant emergence by three methods: soil (3.5 L da^{-1} and 7.5 L da^{-1}), foliar (1.75 L da^{-1} and 3.75 L da^{-1}) and soil + foliar combination (3.5 L da^{-1} + 1.75 L da^{-1}). The effects of these treatments on plant growth, development, and nutrient uptake were investigated in soilless culture. The soil humic acid application was found to be more effective than foliar application, and increasing humic acid doses were significantly associated with increased vegetative weight of lily plants. The humic acid application was conducted twice at two-week intervals after the plants reached approximately 20 cm height. The applications were as follows: control, 3.5 L da^{-1} from soil, 7 L da^{-1} from soil, 1.75 L da^{-1} foliar, 3.5 L da^{-1} foliar, and 3.5 L da^{-1} from soil + 1.75 L da^{-1} foliar. Harvesting of the plants began when at least two of the lower candles showed color. The soil application of 3.5 L da^{-1} humic acid gave superior leaf number, stem thickness, and full flower diameter results. Meanwhile, 7.5 L da^{-1} soil application showed better plant height, bud length, and branch weight results. Regarding nutrient uptake, the soil application of 3.5 L da^{-1} humic acid significantly increased nitrogen and potassium uptake. The study concluded that the soil application of humic acid is more favorable than foliar application, and exceeding a 3.5 L da^{-1} humic acid dose is unnecessary. Soil application of 3.5 L da^{-1} of humic acid significantly increased nitrogen and potassium uptake. Therefore, two different doses of humic acid applied to soil have shown the best performance in terms of plant development parameters in the Zambesi variety of the lily plant.

Keywords: bulb, cut flower, *Lilium,* soilless agriculture, vegetative

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INTRODUCTION

The cut flower industry is growing internationally, with an increasing impact on the global economy (Rogers et al. 2023). *Lilium longiflorum* is one of the most significant plants in the group of cut flowers (Lu et al. 2020). The genus *Lilium,* comprising about 110 varieties growing in a wide climatic range, is important for phylogenetics and gardening. Most of the varieties of *Lilium* are grown because of their esthetic value (Du et al. 2017). Lily is one of the primary flower bulbs worldwide in the exports of cut flowers, potted flowers, and bulbs (Kamenetsky 2014). It ranks fourth among the top ten cut flowers in the global floriculture trade (Bhandari, Ashwat 2018). Greenhouse gardening has been making increased use of agricultural inputs such as chemical fertilizers to produce a greater amount of products of higher quality in a unit area. However, these applications (fertilizer, pesticide, growth regulators) harm environmental and human health and reduce agricultural soil productivity by upsetting the ecological balance. Consequently, sustainable productivity is destroyed. To cope with such problems, sustainable methods must be preferred, and organic soil regulators must be used more (Ataklı et al. 2021). These organic soil regulators increase plant resistance to drought and salinity sensitivity by increasing the population of soil-borne living creatures, changing the physical conditions of enzymatic and hormonal effects, regulating the soil pH, and promoting plant growth. Humic substances are the leading ones among these materials. Humic substances affect plant nutrient uptake, inflorescence rate, root growth, and increased product quality and quantity (Tarakcioğlu et al. 2007). Humic acids play an important role in controlling different environmentally friendly biological, physical, and chemical processes that can directly or indirectly affect plant growth (Berbara, García 2014). Humic substances boost soil productivity, decrease soil intensity, stimulate microbial activities to increase some enzymes contained in soil, promote plant growth to increase the biomass amount of the plant, and may affect the soil organic substance (Nardi 2009). Humic substances in the soil may induce plant growth by synchronizing vegetative hormones (Nardi 2009, Jindo et al. 2012). Recent research on the effects of humic materials on plants has reported that using growth regulators and humic substances has positive effects on plant biomass and growth parameters (Suliman et al. 2020).

The composition and chemical structure of the organic matter in the soil is not uniform. Organic matter is mainly composed of such compounds as carbohydrates, resins, oils, waxes, tannins and proteins. The structure of mineralized organic matter mainly contains humic and fulvic acids. Humic substances are heterogeneous natural resources with high molecular weight, colors ranging from yellow to black, and resistance to degradation. By creating attachment surfaces in the soil, humic substances can increase the uptake of nutrients such as phosphorus, zinc and iron, which are difficult for plants to take up from the soil (Hua et al. 2008). This situation arises from the increase in the cation exchange capacity in the soil. Humic acid acts as a buffer in the external pH range and since the soil is neutralized, various microelements can be taken up by plants.

This study aimed to assess the impact of humic acid on the growth, development, and nutrient uptake of the Zambesi lilium variety. Various doses of humic acid were applied through soil, foliar, combined methods, with the appropriate amount of commercial fertilizer added in the growing medium.

MATERIALS AND METHODS

The study was conducted in a plastic greenhouse with roof ventilation (40°19'46"N 36°28'43" E), but without a heating and cooling system. The investigations regarding vase life were carried out in a laboratory setting. The conditions in the vase room were as follows: temp. was set at 21±2°C, humidity level at %52±5 (Hobo Data Logger U12-012, Onset, United States), light intensity at 482 lux, and the day length was determined as 11 h of light and 13 h of darkness.

Material

Bulbs of the Zambesi (Oriental) variety were transported from the Netherlands using vehicles equipped with cold storage. The bulbs were received in a rooted form with an 18-20 cm calibration. The humic acid (Biomol, Genta) used in the study has a total (humic + fulvic) acid content of 15%, organic matter of 15%, water-soluble potassium oxide (K_2O) 3%, with a pH of 8-10, and leonardite is the substance used in its production. The dimensions of the crates used in the soilless agriculture where the study was conducted are 20 cm in depth, 40 cm in width, and 60 cm in length.

Methods

In the study, crate culture was preferred in soilless farming. Before planting the bulbs, a growing medium with a thickness of 6 cm was placed at the bottom of the crates, and the bulbs were then planted in groups of eight. The planted bulbs were covered with at least 10 cm of growing medium, and water was supplied. The growing medium for the plants consisted of a mixture of peat and perlite in a ratio of 2:1 (v/v). The plants were irrigated using a drip irrigation system. Each container had two drip irrigation tubes (with a flow rate of 4 L s^{-1}). In the early morning hours (08:00), irrigation was carried out for 5 min per plant, ensuring 50-80 mL of water per plant. Fertilization was initiated in the study when the lily plants reached approximately 20 cm in height. Two weeks after the emergence of the plants, fertilization was carried out weekly using Hoagland solution, with an EC value adjusted to 1.4 mhos cm^{-1} (Hamurcu et al. 2016). The prepared containers were placed on the floor inside the greenhouse. The climate conditions within the greenhouse were recorded using a data logger (Hobo Data Logger U12-012, Onset, United States). Throughout the study, the greenhouse temp. varied with an average of 25.16°C ± 2°C; the highest temp. recorded was 37.49 ± 2°C, and the lowest temp. recorded was 14.86 ± 1°C. The relative humidity inside the greenhouse was measured at 54.47% ± 2%.

The humic acid application was conducted twice at two-week intervals, first when the plants reached approximately 20 cm height. The applications were as follows: control, 3.5 L da^{-1} from soil, 7 L da^{-1} from soil, 1.75 L da^{-1} foliar, 3.5 L da^{-1} foliar, and 3.5 L da^{-1} from soil + 1.75 L da^{-1} foliar. Harvesting of the plants began when at least two of the lower candles showed color.

Parameters for post-study analysis

At the end of the study, parameters such as the length of the flower stem (cm), thickness of the flower stem (mm), height of the plant (cm), number of leaves (pieces per plant), bud length (cm), number of buds (pieces/per plant), weight of the branch (g), vase life (day) and total blossom flower diameter (cm) were analyzed. The vase life is defined as the time elapsed from the moment the flowers are placed in a vase until the leaves begin to yellow, the petals start to fade and shed, and concurrently, the flower stem begins to bend (Aziz et al. 2020).

Leaf analyses: Leaf samples from lily plants were collected at the bud stage and prepared for analysis by drying in an oven at 80°C until the samples reached constant weight. Following the method reported by Miller (1998), macro-nutrient elements such as phosphorus (P), potassium (K), calcium (Ca), and magnesium (Mg) in the ash obtained after dry combustion were measured using an ICP-AES device. The analysis of nitrogen (N) elements was determined using the standard Kjeldahl fresh combustion method. According to the methods reported by Bowman et al. (1988) and Jones (2001), nitrogen in a plant sample, obtained by fresh combustion with sulfuric acid and Kjeldahl flasks with catalysts, was measured using the distillation method for the ammonium (NH_4) formed by the breakdown of nitrogen in the plant sample.

Experiment design and assessment of data

The experiment was set up according to a randomized plot experimental design with three repetitions and 8 bulbs for each repetition. The results were analyzed using the IBM SPSS Statistics 26.0 software package according to one-way ANOVA variance analysis. The statistical differences between the applications were defined using the Duncan test, and the degree of significance was determined according to 5%.

RESULTS AND DISCUSSION

An analysis of the effects of various humic acid applications on the plant growth of lily plants (*Lilium* sp.) has revealed that parameters such as the bud length, bud number, and vase life are statistically significantly affected ($p<0.01$). According to the study results, the lilies of the Zambesi variety exhibited the highest number of leaves (66.39) after a soil application of 3.5 L da^{-1} humic acid. The lowest leaf count (57.33) was observed with a combined application of humic acid from soil and foliar. In cv. Zambesi lilies, the most extended flower stem (64.36 cm) was obtained in the control treatment, while the shortest flower stem (58.72 cm) was determined in the variant with a foliar application of 3.75 L da^{-1} humic acid. In cv. Zambesi lilies, the thickest flower stem (6.63 mm) was obtained in the variant with a 3.5 L da^{-1} humic acid application to the soil. In comparison, the thinnest flower stem (5.98 mm) was obtained after the 3.75 L da^{-1} humic acid application sprayed over leaves. In cv. Zambesi, the highest lily plants (64.36 cm) were obtained in the variant providing 7 L da^{-1} humic acid to the soil. In comparison, the lowest plant height (58.72 cm) was recorded after a 3.75 L da^{-1} humic acid foliar application (Figure 1).

The soil application of humic acid at 7 L da^{-1} resulted in the most extended bud length (95.76 mm), while the shortest bud length was in the control group – NPK (70.76 mm). Examination of the effect of humic acid applications on the number of buds shows that the control group and the soil application of humic acid at a 7 L da^{-1} dose produce the highest number of buds (4.72 pieces – 4.67 pieces). Foliar humic acid application of 3.5 L da^{-1} resulted in the lowest number of buds (3.67 pieces). The effect of humic acid applications on the lily plants of the variety Zambesi in terms of the branch weight was also examined. The highest stem weight was obtained after the soil application of 7 L da^{-1} humic acid, while the lowest branch weight was observed as a result of the foliar application of 3.75 L da^{-1} humic acid. An analysis of the effect of humic acid applications to soil, to leaves or to both soil and leaves on the vase life of lilies (*Lilium* sp.) showed that vase life was the longest (17 days) for plants in the control group, and that humic acid applications reduced vase life, with the shortest vase life being 13 days, that is 24% shorter. In cv. Zambesi, the highest fully opened flower diameter (26.67 cm) was obtained in the variant with a 3.5 L da^{-1} humic acid application, while the smallest flower diameter (22.67 cm) was recorded in lilies given 7 L da^{-1} humic acid (Figure 2).

In the study, when the nutrient contents of leaf samples of the Zambesi variety were analyzed, nitrogen and potassium contents were found to be statistically significant ($p<0.01$), while phosphorus, calcium, and magnesium contents were not significant ($p<0.05$). The highest nitrogen content (3.75%) was obtained in the 7 L da^{-1} application treatment, while the lowest nitrogen content (3.23%) was found in the control group. The highest phosphorus con-

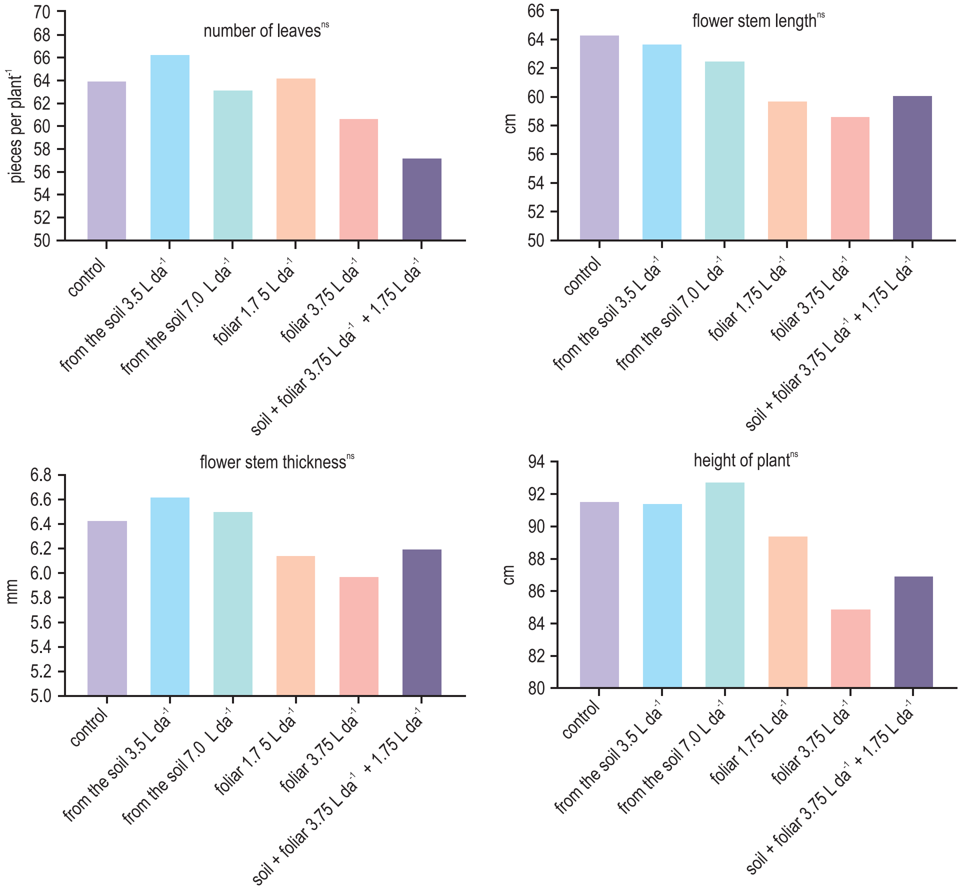

Fig. 1. Effects of humic acid applications on leaf number, flower stem length, flower stem thickness and plant height, ns – non-significant

tent (3.81%) was recorded in the variants with the soil and foliar humic acid application, while the lowest phosphorus content (3.51%) was observed in the control (Figure 3).

The study has demonstrated that different concentrations and application modes of humic acid have significant effects, especially on the length of buds, number of buds, and the vase life of lily plants, while humic acid (HA) yielded better results than determined in the control group as regards the number of leaves, thickness of the flower stem, height of the plant, weight of branch, and the diameter of full blossom flower. However, these positive effects vary depending on the different concentrations and application methods of humic acid. This fact is inconsistent with some of the earlier studies that showed that HA improved the plant's photosynthesis and productivity (Liu et al. 1998, Yang et al. 2004). This may be related to plant varieties and the origin of HA, or the plant's reduced growth at higher HA

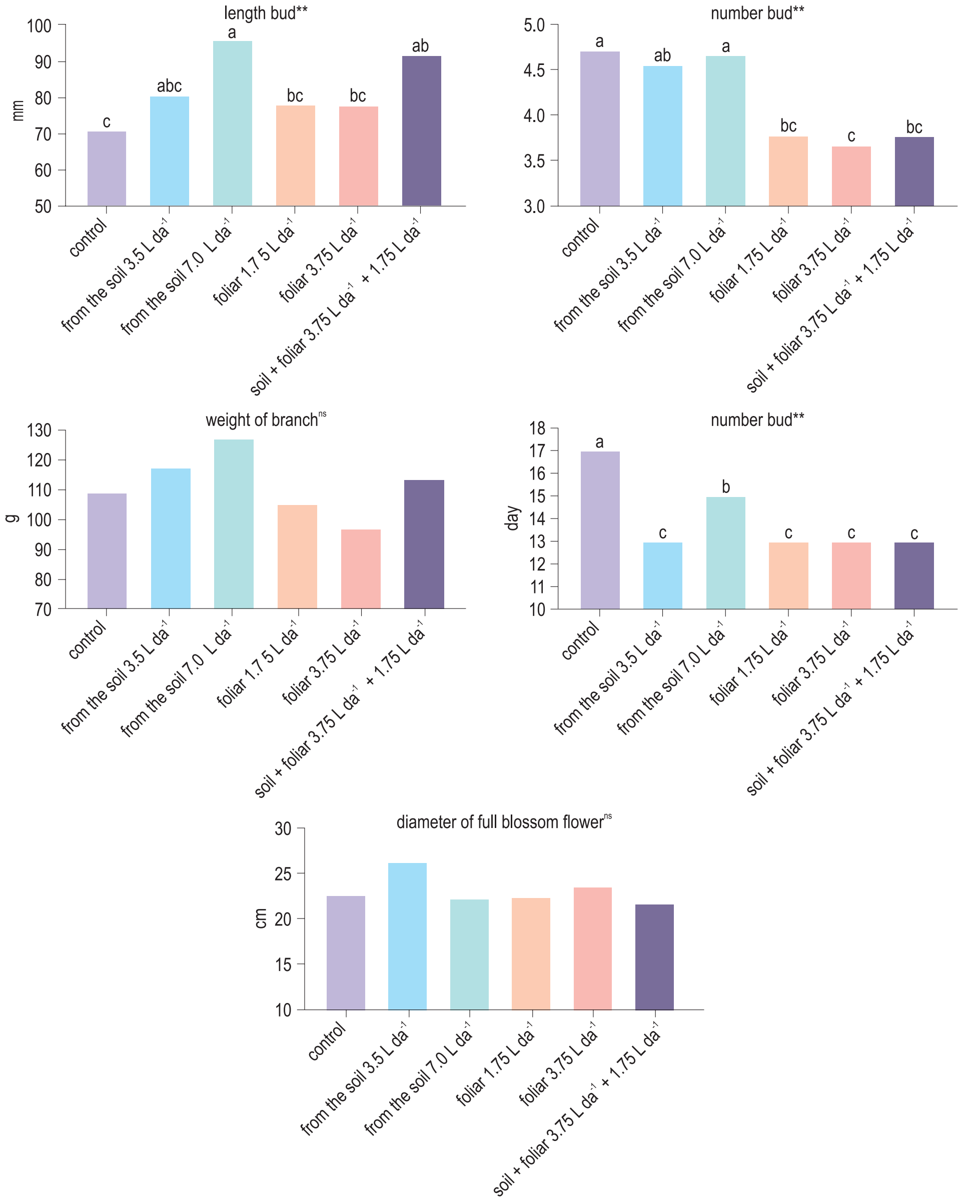

Fig. 2. Effects of humic acid applications on length bud, number bud, weight of branch, vase life and diameter of full blossom flower, ** p<0.001, ns – non-significant

concentrations (Chen and Aviad 1990). Better results have been obtained for the number of leaves in variants with soil applications of humic acid, particularly of a dose of 3.5 L da^{-1}. The higher number of leaves is believed to result from the indirect positive effect of humic acid on the amount of chlorophyll. It is also estimated that a sufficient amount of chlorophyll can in-

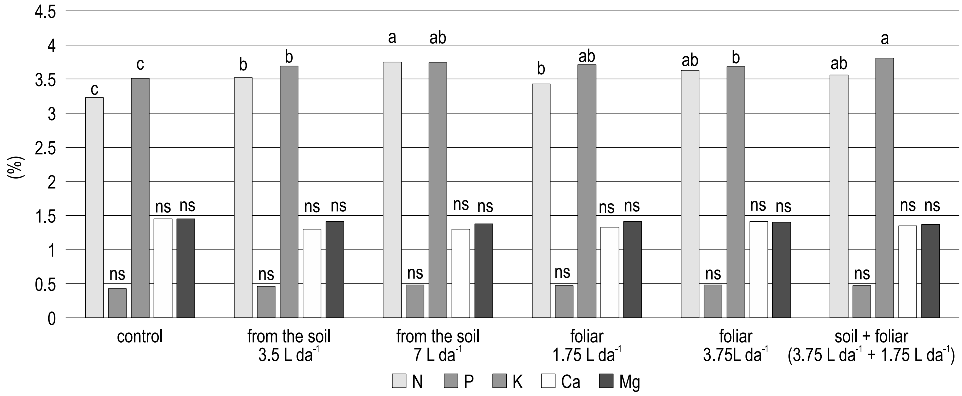

Fig. 3. Effects of different humic acid treatments on macronutrient content in leaves of the Zambesi lily cultivar (ns – non-significant)

crease photosynthetic activities, leading to a higher degree of photo assimilation, producing a greater number of fresh leaves (Meganid et al. 2015, Ullah et al. 2021). It has been seen that soil application of humic acid increases water retention capacity. Using humic acid in addition to the nutrient solution given to plants as a standard promotes root growth. This may depend on varieties, growth conditions, and different doses of humic acid applied (Wagas et al. 2014). Also, in coherence with the previous studies, humic acid application to soil gives similar results to those obtained for corn and wheat, increasing the number of ears compared to the control group (Ullah et al. 2021). The increased growth and flowering of lily plants brought by humic acid is supported by the previous findings concerning humic acid application (Khan et al. 2010).

Humic acid improves the root development of Lilium bulbs by creating a suitable medium for them and increasing their intake of nutrients. Photosynthesis is thus increased, producing more energy (Atiyeh et al. 2002). This energy boosts plant development and results in the growth of plants. In conclusion, humic acid application through soil promotes more photosynthesis and better plant development than control (Ahmad et al. 2013). Having such positive effects, humic acid is estimated to have affected the growth rate and length of the buds. Research has also proven that humic acid application plays an important role in cell enlargement and cell elongation; results similar to this study have been put forward (Khodakhah et al. 2014).

A positive correlation has been observed between humic acid application and the number of buds. However, this application had a positive effect when made through the soil. The reason may be that humic acid activity, which significantly increases the number of buds, may have resulted in the maximum absorption of nutrients from the soil. Studies have also reported that humic acid application of 2000 mg L^{-1} yielded better results than the control group (Bryan 1976). Memon et al. (Memon et al. 2014) reported having obtained similar results with a humic acid application of 4 g 2 m^{-2}. The findings support our results, where humic acid applications of 3.5 L da^{-1} and 7 L da^{-1}, administered through the soil, significantly increase the number of buds of the plant (Mohammadipour et al. 2012).

It has been seen that humic acid applications reduce vase life compared to control. However, a case to the contrary has been observed in studies about the effect of humic acid on vase life. The longest vase life (18.0 days) was determined by the combined application of potassium and calcium fertilizers (Çakıcı et al. 2022). In the studies done with ornamental plants such as zinnia, gerbera, tulip, and tuberose, increased doses of humic acid have retarded the aging of flowers and extended the vase life of flowers (Kumar et al. 2003, Yazdani et al. 2014).

CONCLUSIONS

The study's results indicate that soil applications of humic acid give better results than foliar applications. In soil application of humic acid as 3.5 L da^{-1}, parameters such as the number of leaves, the thickness of the stem, and the diameter of the full blossom flower have given the best results compared to the other applications. In the variant with the soil application of humic acid at 7.5 L da^{-1}, parameters such as the height of the plant, length of buds, and the branch's weight have yielded better results than after the other applications. Soil application of 3.5 L da^{-1} of humic acid significantly increased nitrogen and potassium uptake. Therefore, two different doses rates of humic acid application from soil have shown the best performance on plant development parameters in the Zambesi variety of the lily plant.

Author contributions

S.Ş. and O.S.A. – came up with the experiment's idea, designed the study, supervised the experiments, field analysis, data collection analysis, and wrote the manuscript, S.B. – contributed to leaf analysis. All these authors contributed substantially to the final manuscript and approved this submission. All the authors are aware of the authorship rights and that no further change in authorship will be performed after submission except those previously authorized by the editor-in-chief.

Conflicts of interest

The authors declare no conflict of interest.

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