
Influence of humic acid and macronutrients (MgSO₄ + S) application on growth and yield of petunia (*Petunia milliflora*)

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A field experiment was conducted during 2012-13 at the experimental fields of Horticulture Garden, Department of Horticulture, Sindh Agriculture University Tandojam to examine the effect of various levels of humic acid + MgSO₄ (Magnesium sulphate) +S (Sulphur) on growth and yield of petunia, cv. Fantasy pink morn. The experiment was laid out in a three replicated Randomized Complete Block design.

The results revealed that the growth and flower yield parameters of petunia were significantly affected by different levels of humic acid + S + MgSO₄ (P<0.05). The plants treated with 800 g Humic Acid + 60g S + 100g MgSO₄ produced 87.00 cm plant height, 12.40 branches plant⁻¹, 262.00 flowers plant⁻¹, 29.40 leaves branches⁻¹, 0.700 g weight of flowers plant⁻¹, took 18.40 days to opening of first flower and 5.40 days total blooming period. The Petunia treated with 400 g Humic Acid + 60g S + 100g MgSO₄ resulted in 83.20 cm plant height, 9.00 branches plant⁻¹, 232.67 flowers plant⁻¹, 27.40 leaves branches⁻¹, 0.630 g weight of flowers plant⁻¹, took 23.40 days to opening of first flower and 4.40 days total blooming period. The plants received only higher level of Humic Acid of 800 g without S + MgSO₄ resulted in 77.40 cm plant height, 7.40 branches plant⁻¹, 217.00 flowers plant⁻¹, 24.00 leaves branches⁻¹, 0.510 g weight of flowers plant⁻¹, took 30.00 days to opening of first flower and 4.00 days total blooming period. The application of humic acid @ 400 g showed a minor decrease in values for all the traits examined; while petunia cultivated without Humic Acid only given 60g S + 100g MgSO₄ resulted in overall poor performance. Similarly, the petunia cultivated under control also showed poor performance. It was concluded that the flower production as well as blooming period was highest in petunia given humic acid @ 800g + 60g sulphur + 100g Magnesium treatment; while decreasing humic acid rate resulted in decreased values for all the traits studied under combination of humic acid+S+MgSO₄. Apparently, the application of humic acid improved the soil organic matter that resulted in improved soil efficiency and hence increase in the flowers production and blooming period. However, Magnesium + Sulphur without humic acid did not show promising results.

Key words: Petunia (*Petunia milliflora*), humic acid, Magnesium sulphate + Sulphur, flower production

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Introduction

Petunia (*Petunia milliflora*), perennials in warm climates belong to the Solanaceae family and are used mainly as annual bedding and container plants in temperate zones (Baily and Baily, 1976). The origin of *P. × hybrida* is thought to be by hybridisation between *P. axillaris* and *P. integrifolia*. *P. axillaris* bears night-fragrant, buff-white blossoms with long, thin tubes and somewhat flattened openings. The species was first sent from South America to Paris in 1823. *P. integrifolia* has a somewhat weedy habit, spreading stems with upright tips, and small lavender to purple flowers. It was discovered in South America in 1831. Many open-pollinated species are also gaining popularity in the home garden (Ando *et al.* 2001). A wide range of flower colours, sizes, and plant architectures are available in both the hybrid and open-pollinated species (Ellis, 1999).

Indiscriminate use of chemical fertilizers destructs soil chemical and physical texture and in the long term, consequences will be irreversible which reduced plant yield (Astarai and Kouchaki, 1996, Makikouti, 1999). Humic acids are colloids and behave somewhat like clays, even though the nomenclature suggests that they are acids and form true salts (Griesbach, 2007). There is a growing interest in the use of organic materials as fertilizers or soil amendments. This may be attributed to: 1) an interest in the reduction of the use of chemical fertilizers; 2) public concern for the potential polluting effects of chemicals in the environment; and 3) a pressing need for energy conservation (Senn and Alta, 1973). Humic acid can be used as an alternative to synthetic fertilizers to increase crop production or an indirect effect, mainly by changing the soil structure (Biondi *et al.*, 1994). Humic acid (HA) is also proven to be effective in enhancing the mineral nutrient uptake (Pan and Dong, 1995). Humic acid comes from the highly compressed and biodegraded remains of ancient plants and animals. Over millions of years, plant and animal remains were converted into complex organic molecules and minerals. When this material is applied to soils, it helps the soil to promote better plant growth and productivity naturally. Humic acid helps chelate and improve the effects of many fertilizers. Humic acid also helps the soil retain the nutrients making plants healthier. Healthy plants are often less susceptible to insect and disease problems, a beneficial side effect (Henry Timrod, 2011). Use of humic acid and bio-fertilizers is proposed to modify soil texture, soil structure integrity, aeration and increase nutrient absorption. Humic acid is a commercial product containing abundant nutrients improves soil fertility and increase the availability of nutrients to plants and thus it influences plant growth and yield (Muhammdi Torkashrand, 2009, Salman *et al.*, 2005). Humic acid containing compounds in hoagland solution, increased nitrogen uptake and improved

overall yield. The use of humic acid and chemical fertilizers improves plant nutrient absorption (Ayas and Gulser, 2005). Humic acid is a commercial product contains many elements which improve the soil fertility and increasing the availability of nutrient elements and consequently affected plant growth and yield (Hartwigson and Evans, 2000). Humic substance supply growing plants with food; makes soil more fertile and productive, increases the water holding capacity of soil; therefore, it helps plants resist droughts and stimulates seed germination (Hartwigson and Evans, 2000). The indirect effects of humic compounds have been attributed to the improvement of physical, chemical and biological conditions of soil (Mallikarjuna *et al.*, 1987). MacCarthy *et al.* (2001) reported that humates enhanced nutrient uptake, improve soil structure, and increase crop yields (Maggioni *et al.*, 1987; De Kreij and Basar 1995; Mackowiak *et al.*, 2001), to promote the root length (Vaughan and Malcolm, 1979), and to increase the fresh and dry weights of crops and ornamental plants (Kausar *et al.*, 1985; Chen *et al.*, 2004a, b). Due to the positive effect of humic substances on the visible growth of plants, these chemicals have been widely used by the growers instead of other substances such as pesticides etc.

Similarly, among micronutrients, $MgSO_4$ is an inorganic salt containing magnesium, sulfur and oxygen. It is often encountered as the heptahydrate sulfate mineral epsomite ($MgSO_4 \cdot 7H_2O$), commonly called Epsom salt (Peterson *et al.*, 2006). Magnesium sulfate, known in most households as Epsom salts, is a combination of magnesium and sulfur, both nutrients beneficial for plant health. Magnesium is involved in photosynthesis, so deficiency of this nutrient affects leaf color. Sulfur activates plant enzyme systems. A deficiency of sulfur results in growth and color changes. Plant requirements for sulfur are equal to those for phosphorus, although sulfur is not considered one of the primary plant nutrients. Observation of plant leaves provides information needed to diagnose nutrient deficiencies. However, because magnesium and sulfur symptoms look very similar to symptoms related to other nutrients, it is necessary to distinguish between the symptoms of various nutrient deficiencies (Kilpatrick, 2013).

Sulfur contributes to plant growth and health. Organic and humus-rich soils are enriched with sulfur from its release during decomposition of organic matter. Because sulfur leaches out of soil very easily, the top layer of garden soil can be deficient while deeper layers of soil have sufficient amounts of this nutrient. Plants with deep roots can benefit from nutrients in lower reaches of the soil (Kilpatrick, 2013; Mohanapriya *et al.*, 2006). Krug *et al.* (2009) indicated a positive impact of $MgSO_4$ and Sulphur application of Petunia growth and flowering. In view of the facts stated above, the experiment was

conducted to investigate the influence of humic acid and macronutrients ($\text{MgSO}_4 + \text{S}$) application on yield and growth of Petunia.

Materials and methods

The experiment was conducted at Horticulture Garden, Department of Horticulture, Sindh Agriculture University Tandojam to assess the influence of humic acid and $\text{MgSO}_4 + \text{Sulphur}$ application on the growth and yield of Petunia cv. Fantasy pink morn. The experiment was laid out in a three replicated Randomized Complete Block Design (RCBD) and six treatments. The treatments included, T1=control, T2=400 g Humic Acid, T3=800 g Humic Acid, T4= 60 g Sulphur + 100 g Magnesium, T5= 400 g Humic Acid + 60 g Sulphur + 100 g Magnesium, T6= 800 g Humic Acid + 60 g Sulphur + 100 g Magnesium.

For conducting this experiment, the whole plot was properly worked and leveled for even distribution of water. Thereafter the plot was divided into 18 sub-plots measuring in a plot size of 4ft x 5ft (20ft²). Each bed was separated by developing 30 cm bund, and these sub-plots/beds were prepared in such a way to be irrigated feasibly and uniformly. The petunia seedlings were maintained at a distance of 30 cm between rows and 20 cm between plants. All cultural practices were carried out throughout the growing season as recommended.

Being ornamental plant, petunia are naturally sensitive to some extent and probably do not withstand better under water stress and weedy conditions. Hence, the flower beds were strictly monitored for development of any weeds. Beds were kept clean, and a periodical weeding was performed to allow the experimental plants to utilize nutrients and moisture optimally. Moreover, the presence of weeds could also cause insect pest infestation; so cultural practices were performed in each bed uniformly.

When the plants attained establishment, five normal looking plants in each sub-plot were randomly selected and marked by different labels to avoid any confusion in recording observations. The observations on the number of days taken to opening of first flower of petunia and total blooming period in days were recorded carefully for the petunia in each plot at the field. Observations were recorded, the plant height (cm), number of branches plant⁻¹, number of flowers plant⁻¹, number of leaves branches⁻¹, weight of flowers (g), days taken to opening of first flower and total blooming period (days). The data of all the parameters were individually subjected to the analysis of variance techniques (Steel *et al.*, 1997). Subsequently, the significant means were separated by the least significant difference (LSD) test by using the MSTAT-C computer program.

Results

Plant height (cm)

The effect of humic acid (HA) levels and different concentrations of Magnesium sulphate (MgSO_4) + Sulphur (S) on the plant height of petunia as compared with control and such results are presented in Table-1. The petunia plants grew tallest (87.00 cm) on average when supplied with 800 g Humic Acid + 60g Sulphur + 100g Magnesium (T_6). By reduction of humic acid (400 g Humic Acid + 60g Sulphur + 100g Magnesium, T_5), the plant height decreased to 83.20 cm. In the absence of humic acid the plant height was badly affected and the petunia plant height was decreased to 64.00 cm when no humic acid was given and the plants were supplied only with 60g Sulphur + 100g Magnesium (T_4). However, absence of MgSO_4 or S did a little damage to this trait when supplied with highest humic acid level of 800 g (T_3) resulting average plant height of 77.40 cm, while decreasing half of the humic acid level upto 400 g (T_2) caused only a minor reduction in the petunia plant height (74.40 cm) when compared with 800 g humic acid treatment. However, the plant height declined to its lowest i.e. 54.40 cm (T_1) when the petunia plants were left untreated of humic acid, Magnesium or sulphur (control). The LSD test showed that differences in plant height amongst all the treatments as well as when compared with control were statistically significant ($P < 0.05$) with the exception of T_3 (humic acid 800 g) and T_2 (humic acid 400g) where the differences in plant height were non-significant ($P > 0.05$). The results clearly indicated that application of Magnesium + Sulphur was more effective for improving plant height of petunia when applied in addition to humic acid. However, 400g humic acid would be adequate, because on 800g humic acid application, the plant height did not increase economically.

Number of Branches plant⁻¹

The results in regards to the number of branches plant⁻¹ of petunia as affected by different levels of humic acid, Sulphur + Magnesium sulphate are given in Table-1. The analysis of variance demonstrated that the branches plant⁻¹ were significantly ($P < 0.05$) influenced by various levels of humic acid + Sulphur + Magnesium sulphate combination. The plants fertilized with 800 g Humic Acid + 60g Sulphur + 100g Magnesium (T_6) resulted in maximum number of branches plant⁻¹ (12.40); while the number of branches plant⁻¹ decreased to 9.00 when decreased level of humic acid (400 g Humic Acid + 60g Sulphur + 100g Magnesium, T_5). Absence of humic acid in the treatment plan showed negative effects on this trait and branches plant⁻¹ decreased to 6.00 when no humic acid was given and the plants were supplied only with 60g Sulphur + 100g

Magnesium (T₄). Higher humic acid level of 800 g (T₃) even in the absence of MgSO₄ or S compensated very well resulting 7.40 branches plant⁻¹, while decreasing humic acid to 400g level (T₂) caused a little decrease in branches (6.07) plant⁻¹ when compared with 800g humic acid treatment. The branches plant⁻¹ declined to minimum of 4.40 (T₁) in control where the plants were left untreated of humic acid, Magnesium or sulphur. The LSD test indicated that differences in branches plant⁻¹ between T₂ (400g humic acid), T₃ (800g humic acid) and T₄ (60g sulphur + 100g Magnesium) were statistically non-significant (P>0.05), while significant (P<0.05) when compared with rest of the treatments and control. It was evident from the results that humic acid resulted in a remarkable increase in the branches plant⁻¹ either when applied alone or in combination with Magnesium + Sulphur; while Magnesium + Sulphur more effective when applied in addition to humic acid.

Number of flowers plant⁻¹

The data in relation to number of flowers plant⁻¹ of petunia as influenced by various levels of humic acid and Sulphur + Magnesium are presented in Table-1. The analysis of variance revealed that the flowers plant⁻¹ were significantly (P<0.05) affected by varying levels of humic acid + Sulphur + Magnesium combination. The plants fertilized with 800 g Humic Acid + 60g Sulphur + 100g Magnesium (T₆) produced maximum flowers plant⁻¹ (262.00); and the flowers plant⁻¹ reduced to 232.67 under reduced humic acid level (400 g Humic Acid + 60g Sulphur + 100g Magnesium, T₅). Leaving petunia without humic acid resulted adverse effects on flowers plant⁻¹ that decreased to 152.00 when plants were supplied only with 60g Sulphur + 100g Magnesium (T₄). Higher humic acid level of 800 g (T₃) even without adding MgSO₄ or S recompensed rightly by increasing flowers plant⁻¹ (217.00); and decreased humic acid to 400g level (T₂) resulted in a minor decrease in flowers plant⁻¹ (204.00) when comparison was made with 800g humic acid treatment. The flowers plant⁻¹ diminished to a minimum of 132.00 (T₁) in plots left untreated of humic acid, Magnesium or sulphur (control). The LSD test suggested that differences in flowers plant⁻¹ between T₃ (800g humic acid) and T₅ (400g humic acid + 60g sulphur + 100g Magnesium) were statistically non-significant (P>0.05), while significant (P<0.05) when compared with rest of the treatments as well as control. The results clearly suggested that application of humic acid was probably improved the soil organic matter that resulted in improved soil efficiency and hence increase in the flowers plant⁻¹ was noted. However, Magnesium + Sulphur without humic acid did not show promising results for this parameter.

Table 1. Influence of humic acid and macronutrients (MgSO₄+ S) application on mean plant height (cm), number of branches plant⁻¹, number of flowers plant⁻¹, number of leaves branch⁻¹ of petunia

Treatments	Plant height (cm)	Number of branches plant ⁻¹	Number of flowers plant ⁻¹	Number of leaves branch ⁻¹
Control	54.40 e	4.40 d	132.00 e	20.40 c
400 g Humic Acid	74.40 c	6.07 c	204.00 c	22.40 b
800 g Humic Acid	77.40 c	7.40 c	217.00 b	24.00 b
60g Sulphur +100g Magnesium	64.00 d	6.00 c	152.00 d	21.40 b
400 g Humic Acid + 60 g Sulphur +100g Magnesium	83.20 b	9.00 b	232.67 b	27.40 a
800 g Humic Acid + 60 g Sulphur +100g Magnesium	87.00 a	12.40 a	262.00 a	29.40 a
S.E.±	1.5420	0.9506	9.0746	1.8915
LSD 0.05	3.4358	2.1181	20.219	4.2145
LSD 0.01	4.8870	3.0128	28.760	5.9947

In a column means followed by same letters are not significantly different at P=0.05 as suggested by LSD test.

Number of leaves branch⁻¹

The results related to number of leaves branch⁻¹ of petunia as affected by different levels of humic acid and Sulphur + Magnesium are shown in Table-1. The analysis of variance demonstrated that the leaves branch⁻¹ were significantly (P<0.05) affected by varying humic acid + Sulphur + Magnesium combination levels. The petunia plants receiving 800 g Humic Acid + 60g Sulphur + 100g Magnesium (T₆) resulted in maximum leaves branch⁻¹ (29.40); followed by 27.40 and 24.00 leaves branch⁻¹ recorded in the plants given reduced humic acid level (400 g Humic Acid + 60g Sulphur + 100g Magnesium, T₅) and Higher humic acid level of 800 g (T₃) without MgSO₄ + S, respectively. Growing petunia without humic acid (T₄= 60g Sulphur + 100g Magnesium) showed adverse effects on leaves branch⁻¹ (21.40). The decreased humic acid to 400g level (T₂) resulted in a slight reduction in leaves branch⁻¹ (22.40) as compared

with 800g humic acid treatment. However, leaves branch⁻¹ declined to its lowest of 20.40 (T₁) in plots left untreated (control). The LSD test demonstrated that differences in leaves branch⁻¹ between T₅ and T₆ or amongst T₂-T₃-T₄ were statistically non-significant (P>0.05), while significant (P<0.05) when these treatment groups were compared with each or compared with control. Hence, 400g humic acid + 60g Sulphur and 100g Magnesium would be adequate and optimum level; while further increase in humic acid was not economically beneficial for this trait of petunia.

Weight of flowers plant⁻¹ (g)

The data in regards to weight of flowers plant⁻¹ of petunia as influenced by various levels of humic acid and Sulphur + Magnesium are presented in Table-2. The analysis of variance indicated that the weight of flowers plant⁻¹ was significantly (P<0.05) influenced by varying levels of humic acid + Sulphur + Magnesium. The petunia given 800 g Humic Acid + 60 g Sulphur + 100 g Magnesium (T₆) produced highest weight of flowers plant⁻¹ (0.70 g); and the weight of flowers reduced to 0.63 g under reduced humic acid level (400 g Humic Acid + 60g Sulphur + 100g Magnesium, T₅). Leaving petunia without humic acid resulted adverse effects on weight of flowers plant⁻¹ that decreased to 0.29 g when plants were supplied only with 60g Sulphur + 100g Magnesium (T₄). Higher humic acid level of 800 g (T₃) even without adding MgSO₄ or S compensated well by increasing weight of flowers plant⁻¹ (0.51 g); and decreased humic acid to 400g level (T₂) resulted in a minor decrease in weight of flowers (0.49 g) plant⁻¹ when comparison was made with 800g humic acid treatment. The weight of flowers plant⁻¹ declined to a minimum of 0.22 g (T₁) in plots left untreated of humic acid, Magnesium or sulphur (control). The LSD test suggested that differences in weight of flowers plant⁻¹ between T₃ (800g humic acid only) and T₂ (400g humic acid only) were statistically non-significant (P>0.05), while significant (P<0.05) when compared with rest of the treatments as well as control. The results clearly indicated that the application of humic acid was markedly beneficial for the experimental soil and under higher humic acid level of 800 g in addition to 60 g sulphur and 100 g magnesium resulted in healthier foliage and flowers. However, Magnesium + Sulphur without humic acid did not show hopeful results for the weight of flowers plant⁻¹.

Days to opening of first flower

The results in regards to days to opening of first flower of petunia as affected by different levels of humic acid, Sulphur + Magnesium are given in Table-2. The analysis of variance demonstrated that the days to opening of first

flower were significantly ($P < 0.05$) influenced by various levels of humic acid + Sulphur + Magnesium combination. The plants fertilized with 800 g Humic Acid + 60g Sulphur + 100g Magnesium (T_6) resulted in early flowering with only 18.40 days to opening of first flower; while the days to opening of first flower increased to 23.40 when humic acid was decreased to 400 g in addition to 60g Sulphur + 100g Magnesium (T_5). Absence of humic acid prolonged the opening of first flower upto 36 days when plants were supplied only with 60g Sulphur + 100g Magnesium (T_4). Higher humic acid level of 800 g (T_3) and lower humic acid level of 400 g even in the absence of $MgSO_4$ or S resulted in early opening of first flower in 30.00 and 31.40 days, respectively. The period to opening of first flower prolonged upto 41.00 days (T_1) in control plots. The LSD test indicated that differences in days to opening of first flower between T_2 (400g humic acid), T_3 (800g humic acid) and T_4 (60g sulphur + 100g Magnesium) or between 800 g Humic Acid + 60g Sulphur + 100g Magnesium (T_6) and 400 g humic acid + 60g Sulphur + 100g Magnesium (T_5) were statistically non-significant ($P > 0.05$), and significant ($P < 0.05$) when these treatment groups were compared with each other and control. The results suggested that increasing humic acid levels simultaneously reduced to days to opening of first flower.

Total blooming period (days)

The data pertaining to total blooming period of petunia as affected by different levels of humic acid, Sulphur + Magnesium are shown in Table-2. The analysis of variance indicated that the total blooming period was significantly ($P < 0.05$) influenced by humic acid + Sulphur + Magnesium combination levels. The petunia fertilized with 800 g Humic Acid + 60g Sulphur + 100g Magnesium (T_6) resulted in maximum blooming period of 5.40 days; while the total blooming period decreased to 4.40 when humic acid was decreased to 400 g in addition to 60g Sulphur + 100g Magnesium (T_5). In case of no humic acid, blooming period was reduced upto 3.40 days (only 60g Sulphur + 100g Magnesium (T_4)). Higher humic acid level of 800 g (T_3) and lower humic acid level of 400 g even in the absence of $MgSO_4$ or S resulted in equal blooming period of 4.00 days, respectively. The total blooming decreased to minimum (3.00 days) in control plots (T_1). The LSD test indicated that differences in total blooming period between T_2 (400g humic acid), T_3 (800g humic acid) and T_5 (400 g humic acid + 60g Sulphur + 100g Magnesium) were statistically non-significant ($P > 0.05$), and significant ($P < 0.05$) when these treatments were compared with rest of the treatments and control. The results suggested that increasing humic acid levels caused a simultaneous increased in the total blooming period in petunia.

Table 2. Influence of humic acid and macronutrients (MgSO₄ + S) application on mean weight of flowers plant⁻¹(g), number of days to opening of first flower, Total blooming period (days) to opening of first flower of petunia

Treatments	Weight of flowers plant ⁻¹ (g)	Number of days to opening of first flower	Total blooming period (days)
Control	0.22 e	41.00 c	3.00 c
400 g Humic Acid	0.49 c	31.40 b	4.00 b
800 g Humic Acid	0.51 c	30.00 b	4.00 b
60 g Sulphur + 100 g Magnesium	0.29 d	36.00 b	3.40 c
400 g Humic Acid + 60 g Sulphur + 100 g Magnesium	0.63 b	23.40 a	4.40 b
800 g Humic Acid + 60 g Sulphur + 100 g Magnesium	0.70 a	18.40 a	5.40 a
S.E.±	0.014	2.4449	0.2459
LSD 0.05	0.058	5.4477	0.5478
LSD 0.01	0.080	7.7487	0.7792

In a column means followed by same letters are not significantly different at P=0.05 as suggested by LSD test.

Discussion

The agriculture soils have become poor in organic matter due to continuous cropping and non-use of organic sources that contains humus to improve the soil productivity efficiency. Humic acid contains many elements which improve the soil fertility and increasing the contents of soil organic matter and consequently affect plant the growth and yield (Hartwigson and Evans 2000). The use of humic acid has been found in different fertilizers forms. The indirect effects of humic compounds have been attributed to the improvement of physical, chemical and biological conditions of soil (Mallikarjuna *et al.*, 1987); while its direct effect on plant growth has been attributed to the increase in chlorophyll content, the acceleration of the respiration process, hormonal growth responses, increasing penetration in plant membranes or a combination of these processes (Mallikarjuna *et al.*, 1987). The present study was carried out to examine the effect of various levels of humic

acid +S (Sulphur) + MgSO₄ (Magnesium sulphate) on the growth and yield of petunia.

The findings of the present study indicated that plants given 800 g Humic Acid + 60g S + 100g MgSO₄ produced 87.00 cm plant height, 12.40 branches plant⁻¹, 262.00 flowers plant⁻¹, 29.40 leaves branches⁻¹, 0.700 g weight of flowers plant⁻¹, took 18.40 days to opening of first flower and 5.40 days total blooming period. The Petunia treated with 400 g Humic Acid + 60g S + 100g MgSO₄ resulted in 83.20 cm plant height, 9.00 branches plant⁻¹, 232.67 flowers plant⁻¹, 27.40 leaves branches⁻¹, 0.630 g weight of flowers plant⁻¹, took 23.40 days to opening of first flower and 4.40 days total blooming period. The plants received only higher level of Humic Acid of 800 g without S + MgSO₄ resulted in 77.40 cm plant height, 7.40 branches plant⁻¹, 217.00 flowers plant⁻¹, 24.00 leaves branches⁻¹, 0.510 g weight of flowers plant⁻¹, took 30.00 days to opening of first flower and 4.00 days total blooming period. The application of humic acid @ 400 g showed a minor decrease in values for all the traits examined; while petunia cultivated without Humic Acid only given 60g S + 100g MgSO₄ resulted in overall poor performance. Similarly, the petunia cultivated under control also showed poor performance. It was concluded that the flower production as well as blooming period was highest in petunia given humic acid @ 800g + 60g sulphur + 100g Magnesium treatment; while decreasing humic acid rate resulted in decreased values for all the traits studied under combination of humic acid+S+MgSO₄. Apparently, the application of humic acid improved the soil organic matter that resulted in improved soil efficiency and hence increase in the flowers production and blooming period. However, Magnesium + Sulphur without humic acid did not show promising results. These results are further supported by Chamani *et al.* (2008) who examined the effects of humic acid in the form of vermicompost of an animal manure origin on the growth and flowering of *Petunia hybrida* 'Dream Neon Rose' grown under glasshouse conditions. Plant performance was best in the 20% vermicompost medium. Further increasing the vermicompost content in the base media decreased flower numbers, leaf growth rates and shoot fresh and dry weights. Thus, the beneficial effects of humic acid as vermicompost were associated with elevated tissue concentrations of the macronutrients. Jack and Evans (2000) who reported that the humic acid treatments increased root fresh weight of marigold seedlings, and all increased geranium root fresh weight. Kutuk *et al.* (2000) reported that the application of humic acid resulted in an improved plant growth and flower yield in a number of flower species. Atiyeh *et al.* (2002) reported that humic acid concentrations more than 40% (50-100%) significantly decreased shoot weight and height and total number of flower buds. Dudley *et al.* (2004) reported that application of humic substances

showed positive impacts on the growth and flower production of *Zinnia elegans* Jacq. and *Tagetes patula* L. Hank *et al.* (2006) reported that the effect of $MgSO_4$ on the growth and alkaloid production of genetically modified ornamental plant roots. Arancon *et al.* (2008) used humic acid in the form of vermicompost to flowering plants and found that vermicompost produced improvements in the physical structure of the growth medium such as aeration and drainage. Krug *et al.* (2009) reported that petunia (*Petunia x hybrida*, *Viola x vittrrockiana* and *Gerbera janzezonii* plants were grown and their performance was associated with the soil organic matter contents. The results clearly indicated that the soil rich in humus produced improved plant foliage and healthy flowering. Santos *et al.* (2011) reported that effect of micronutrients which also included Magnesium and sulphur resulted in improved plant propagation in petunia. Le Chang *et al.* (2011) investigated that the effect of calcium and humic acid treatment on the growth and nutrient uptake of Oriental Lily and reported that humic acid may facilitate plant growth by improving the nutrient uptake as well as through hormonal effects. Khosa *et al.* (2011) evaluated the effect of macro and micro nutrients subjective the vegetative growth and flowering superiority of gerbera and under higher concentrations the plant height, number of branches per plant, length of branches per plant, number of leave per plant, leaf area, stock length, days to first flower emergence, flower diameter and flower quality increased with increasing fertilization level and began to turn down when fertilization level exceed beyond the above given levels of macro and micro nutrients.

Humic acid has also been found to have positive effects on ornamentals such as geranium (Chand *et al.*, 2007), marigolds (Atiyeh *et al.*, 2002), petunia (Arancon *et al.*, 2008), chrysanthemum (Hidalgo and Harkess 2002) and poinsettia (Hidalgo and Harkess, 2002). Positive effects of humic acid have also been observed in forestry species (Lazcano *et al.*, 2010). Le Chang *et al.* (2011) reported that humic acid (HA) may facilitate plant growth by improving the nutrient uptake as well as through hormonal effects. Sarhan (2011) reported that the interaction between treatments also caused a significant effect and the plants which got a humic acid and sprayed with mixture of Alga 600 and sea force 2 gave the highest values of vegetative characters as compared with lowest values of control plants. Also the results appeared a positive significant effect of humic acid and seaweed extracts and their interactions on all yield quantitative characters. Azarpour *et al.* (2012) showed that the effect of foliar humic acid spraying on all measured traits had significant differences at 1% probability level. Also, effect of nitrogen fertilizer management on fruit yield, number of fruit per square meter, plant height, fruit length and fruit width at 1% and on number of branches per plant at 5% was significant. Interaction effect of

foliar humic acid spraying and nitrogen fertilizer management on fruit yield at 1% and on number of fruit per square meter, plant height, fruit length and fruit width at 5% was significant and on number of branches per plant was non significant. The highest fruit yield was obtained from use of 50 mg L⁻¹ humic acid spraying and also from 80 kg ha⁻¹ nitrogen fertilizer. Bangulzai (2013) indicated that all the zinnia traits examined including plant height, number of branches plant⁻¹, number of leaves branch⁻¹, days taken to first flower emergence, number of flowers plant⁻¹, size of flower, average flower life and total blooming period with the exception of days to first flower emergence, followed similar growth and flower production trend and similar response to various humic acid and FeSO₄. However, days taken to first flower emergence were inversely proportional to increasing humic acid or combination of FeSO₄ which was in contrast to rest of all the characters investigated. Generally, T₅, where 4 g Humic acid + 40 g FeSO₄ per 2m² proved to be an optimum level for zinnia production, while increasing humic acid upto 40 g Humic acid in addition to 40 g FeSO₄ per 2m² showed adverse effects on all the parameters when compared with the optimum level.

Conclusions

The flower production as well as blooming period was highest in petunia plantation given humic acid @ 800g + 60g sulphur + 100g Magnesium treatment; while decreasing humic acid rate resulted in decreased values for all the traits studied under combination of humic acid+S+MgSO₄. Apparently, the application of humic acid improved the soil organic matter that resulted in improved soil efficiency and hence increase in the flowers production and blooming period. However, Magnesium + Sulphur without humic acid did not show promising results.

References

- Ando, T., M. Nomura, J. Tsukahara, H. Watanabe, H. Kokubun, T. Tsukamoto, G. Hashimoto, E. Marchesi and I. Kitching. (2001). Reproductive isolation in a native population of *Petunia sensu Jussieu* (Solanaceae) *Ann. Bot. (Lond.)*, 88:403–413.
- Arancon, N.Q., C.A. Edwards, A. Babenko, J. Cannon, P. Galvis and J.D. Metzger. (2008). Influences of vermicomposts, produced by earthworms and microorganisms from cattle manure, food waste and paper waste, on the germination, growth and flowering of petunias in the greenhouse. *Appl. Soil Eco.*, 39 : 91-99.
- Astaraei, A.R. and A. Kouchaki. (1996). Indiscriminate use of chemical fertilizers. *Bulletin of the Ferdowsi University Publication, Tehran (Iran)*, Pp. 168.
- Atiyeh, R.M., C.A. Edwards, S. Subler and J.D. Metzger. 2002. Pig manure vermicompost as a component of a horticultural bedding plant medium: effects on physicochemical properties and plant growth. *Biores. Tech.* 78 : 11-20.

- Ayas, H. and F. Gulser. (2005). Use of humic acid for improving soil organic matter and increasing crop yield. *Journal of Biological Sciences.*, 5(6): 801-804.
- Azarpour, I., M.K. Motamed, M. Moraditochae and H.R. Bozorgi. 2012. Effects of bio, mineral nitrogen fertilizer management, under humic acid foliar spraying on fruit yield and several traits of eggplant (*Solanum melongena* L.). *African Journal of Agricultural Research*, 7(7) : 1104-1109.
- Baily, L.H. and E.Z. Baily. (1976). *Petunia*. In: *Hortus Third: A Concise Dictionary of Plants Cultivated in the United States and Canada*. Macmillan Publishing, New York, Pp: 850-851.
- Bangulzai, F.M. (2013). Effect of humic acid and iron sulphate on growth and yield of zinnia (*Zinnia elegans*). M.Sc. thesis submitted to Sindh Agriculture University Tandojam.
- Biondi, F. A., A. Figholia, R. Indiati and C. Izza. (1994). Effects of fertilization with humic acids on soil and plant metabolism: a multidisciplinary approach. Note III: phosphorus dynamics and behaviour of some plant enzymatic activities. In *Humic Substances in the Global Environment and Implications on Human Health*, ed. Senesi N & Miano TM. Elsevier, pp. 239-244.
- Chamani, E., D.C. Joyce and A. Reihanytabar. (2008). Vermicompost Effects on the Growth and Flowering of *Petunia hybrida* 'Dream Neon Rose'. *American-Eurasian J. Agric. & Environ. Sci.*, 3 (3): 506-512.
- Chand, S., Pande, P., Prasad, A., Anwar, M. and Patra, D.D. (2007). Influence of integrated supply of vermicompost and zinc-enriched compost with two graded levels of iron and zinc on the productivity of geranium. *Communications in Soil Science and Plant Analysis*, 38, 2581–2599.
- Chen, Y., C. E. Clapp and H. Magen. (2004a). Mechanisms of plant growth stimulation by humic substances: The role of organic-iron complexes. *Soil Sci. and Plant Nutr.*, 50 (7) : 1089–1095.
- Chen, Y., M. Nobili and T. Aviad. (2004b). Stimulatory effect of humic substances on plant growth. "Soil Organic Matter in Sustainable Agriculture". CRC Press, Washington. In: Magdoff F., Ray R. (eds): 103-130 Boca Raton, FL.
- De Kreij, C. and H. Basar. (1995). Effect of humic substances in nutrient film technique on nutrient uptake. *J. Plant Nutr.*, 18 (4) : 793–802.
- Dudley, J.B., A.J. Pertuit and J. E. Toler. (2004). Leonardite Influences Zinnia and Marigold Growth. *Hort Sci.*, 39 (2) : 251-255.
- Ellis, B.W. (1999). *Taylor's Guide to Annuals*. Boston. Houghton Mifflin Co.
- Griesbach, R.J. (2007). In *Flower breeding and genetics: Issues, challenges and opportunities for the 21st century, Petunia*, ed Anderson N.O. (Springer, Dordrecht, The Netherlands), Pp. 301–336.
- Hartwigson, J.A. and M.R. Evans. (2000). Humic acid seed and substrate treatments promote seedling root development. *Hort Sci.*, 35 (7) : 1231-1233.
- Hank, H., I. László, I. Bálványos, G. Kursinszki, E. Kovács, S.E. Tóth. 2006. Effect of magnesium on the growth and alkaloid production of hairy root cultures. *ISHS Acta Horticulturae 597: International Conference on Medicinal and Aromatic Plants (Part II)*, Pp. 546.
- Henry Timrod, (2011). Spring is a true re-constructionist: J&L Garden Center's News. <http://jlgardencenter.com/news/11/15>.
- Hidalgo, P.R. and Harkess, R.L. (2002). Earthworm casting as a substrate for Poinsettia production. *Hort Sci.*, 37 (2), 304-308.
- Jack A., H. and M. R. Evans. (2000). Humic Acid Seed and Substrate Treatments Promote Seedling Root Development. *Hort Sci.*, 35 (7) : 1231–1233.

- Kauser, A. M. and F. Azam. (1985). Effect of humic acid on corn seedling growth. *Env. and Exp. Bot.*, 25: 245–252.
- Kilpatrick, J. (2013). Magnesium Sulfate for plant care. Home guides. Driven by Demand Median Hearst Newspaper. <http://homeguides.sfgate.com.html>.
- Khosa, S.S., A. Younis, A. Rayit, S. Yasmeen and A. Riaz. 2011. Effect of Foliar Application of Macro and Micro Nutrients on Growth and Flowering of *Gerbera jamesonii* L. *American-Eurasian J. Agric. & Environ. Sci.*, 11 (5): 736-757
- Krug, B.A., B.E. Whipker and J. Frantz and I. McCall. 2009. Characterization of Calcium and Boron Deficiency and the Effects of Temporal Disruption of Calcium and Boron Supply on Pansy, Petunia, and Gerbera Plugs. *Hortscience*, 44(6):1566-1572.
- Kutuk, C., C. Gokhan, B. Abdullah and O. Baskan. (2000). Effect of Humic Acid on some soil properties. *Bildiri Ozetleri, Soil Science Department, Agricultural Faculty, Ankara University, 06110- Turkey.*
- Lazcano, C., Sampredo, L., Zas, R. and Domínguez, J. (2010). Vermicompost enhances germination of the maritime pine (*Pinus pinaster* Ait.). *New Forest*. 39 : 387-400.
- Le Chang, Yun Wu, Wei-wei XU, Ali Nikbakht and Yi-ping Xia. (2011). Effects of calcium and humic acid treatment on the growth and nutrient uptake of Oriental lily, *African Journal of Biotechnology*, 11 (9): 2218-2222.
- MacCarthy, P., C.E. Clapp, R.L. Malcolm and P.R. Bloom. (2001). Humic substances in soil and crop sciences: selected readings. *Am Soc. of Agronomy and Soil Sci. Soc. of Am, Madison, WI.*
- Mackowiak C.L., P. R. Grossl and B. G. Bugbee. (2001). Beneficial effects of humic acid on micronutrient availability to wheat. *J. Soil Sci. Soc. Am.*, 56 : 1744–1750.
- Maggioni A., Z. Varanini, S. Nardi and R. Pinton. (1987). Action of soil humic matter on plant roots: Stimulation of ion uptake and effects on (Mg²⁺, K⁺) ATPase activity. *Science of the Total Environment*, 62: 355–363.
- Malakouti, (1999). Dissemination of Agri. Edy-168 pages.
- Mallikarjuna M, Govindasamy R, Chandrasekaran S (1987). Effect of humic acid on Sorghum vulgare var. CSH-9. *Curr. Sci.* 56(24): 1273-1276.
- Mohanapriya, S., P. Senthilkumar, S. Sivakumar, M. Dineshkumar and C.V. Subbhuraam. (2006). Effects of copper sulfate and copper nitrate in aquatic medium on the restoration potential and accumulation of copper in stem cuttings of the terrestrial medicinal plant, *Portulaca oleracea* linn. *Environ Monit Assess*, 121(1-3):233-44.
- Mohammadi Torkashvand, A. (2009). Islamic Azad University of Rasht Press. 264 pages.
- Pan, R. C. and Y. D. Dong. (1995). *Plant physiology* (third edition). High education Press, Beijing, China. (in Chinese).
- Peterson, R.C., J.M. Hammarstrom and I.R.R. Seal. (2006). Alpersite (Mg,Cu)SO₄·7H₂O, a new mineral of the melanterite group, and cuprian pentahydrate: Their occurrence within mine waste. *American Mineralogist*, 91 (2-3): 261–269.
- Salman, S.R., Abou-Hussein S.D., Abdel-Mawgoud, A.M.P., El-Nemr, M.A. 2005. *Journal of Applied Sciences Research.*, 1: 51-58.
- Santos, K.M., P.R. Fisher, T.H. Yeager, E.H. Simonne, H.S. Carter and W.R. Argo. (2011). Effect of petunia stock plant nutritional status on fertilizer response during propagation. *Journal of Plant Nutrition*, 34:1424-1436.
- Sarhan, T.Z. 2011. Effect of humic acid and seaweed extracts on growth and yield of potato plant (*Solanum tuberosum* L) Desiree cv. *Mesopotamia J. of Agric.*, 39 (2) : 19-27
- Senn, T.L. And Alta. R. Kingman. (1973). A review of humus and humic acid research series number 145. S.C. Agricultural. Experimental Station. Clemson, South Carolina

- Steel, R.G.D., J.H. Torrie and D.A. Dicky. (1997). Principles and procedures of Statistics A Biometric Approach. 3rd Ed. McGraw Hill Book Co. Inc., New York.
- Vaughan D., Malcolm R.E. (1979): Effect of humic acid on invertase synthesis in roots of plants. Soil Biology and Biochemistry, 11: 247–252.

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