Physiological Responses of Hybrid Rose (Rosa hybrida L.) to Foliar Glycine and Licorice Root Extract Under Heat Stress Conditions

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Abstract

Roses Hybrid (*Rosa hybrida* L.) have shown sensitivity during development and blooming with regard to high temperature and high light. Foliar biostimulants can help in maintaining performance of plants under these stresses. This study evaluated the effects of shading, glycine, and licorice root extract on the vegetative growth and flowering of hybrid rose. Factorial experiments were carried out during both spring and fall seasons in which 162 potted plants were used. Treatment combinations included two shading rates (0 and 50%), three rates of glycine (0, 5, and 10 mg L^{-1}), and three rates of licorice root extract (0, 50, and 100 g L^{-1}). At 50% shading, there was an increase in flower number, branch number, leaf number, fresh and dry weight, length of main branch, and root length. Addition of glycine caused an increase in flower number, flower diameter, fresh and dry weight, and petals number. The licorice root extract caused an increase in leaf number, stem diameter, fresh and dry weight, and petals number. The highest flower number (8.33 $plant^{-1}$), branch number (3.33 $plant^{-1}$), leaf number (119.7 $plant^{-1}$), and root length (26.3 cm) were achieved when 50% shading, 10 mg L^{-1} glycine, and 100 g L^{-1} licorice root extract were combined. Moderately shaded plants, supplemented with glycine and licorice root extract, performed well in terms of growth and flowering of the hybrid rose.

Keywords: *Rosa hybrida* L.; shading; glycine; licorice root extract; foliar biostimulants.

Introduction

Rosa hybrida is a very important ornamental flower species. The scientific importance of *Rosa hybrida* lies in three major aspects, namely the high amount of biologically relevant phenolics and pigments found in the petals of *Rosa hybrida*, the presence of diverse coloration and scent properties in these flowers and the vulnerability of flower tissues towards postharvest infections [1], [2].

High temperature is a major limitation on the growth of hybrid roses. Hybrid tea roses are susceptible to high temperature stress conditions, which affect flower production. Heat stress is also known to induce the response to heat shock proteins [3]. Elevated temperatures in *Rosa hybrida* 'Carolla' were associated with negative effects on the chlorophyll concentration and photosynthesis process. Furthermore, heat stress impaired the stability of membranes, antioxidative properties, and osmoregulation. Such changes could lead to decreased growth and flowering of plants. However, proper foliar applications could alleviate heat stress [4].

The application of amino acids as foliar biostimulants is becoming more widespread. Glycine, an amino acid, is applied either alone or in mixtures with

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other amino acids. Applications containing glycine have proved effective for improving growth, fruit setting, production, fruit quality, and leaf mineral composition in guava trees [5]. The growth, pigment, yield, biochemical characteristics, and nutritional content of celery improved with glycine. This effect was concentration-dependent and depended on the interaction of glycine with tryptophan [6]. Therefore, the effects of glycine may vary from one plant species to another.

Plants' extracts act as biostimulators in nature. Some of them include the licorice root extract which has been found to have positive impacts on the growth and productivity of plants. In the case of heat-stressed mango trees, fruit production and quality was increased when the plants were sprayed with licorice root extract [7]. These findings support its evaluation in ornamental plants under stressful conditions.

Prior studies have considered heat tolerance of hybrid roses. Also, studies have been conducted on the use of either glycine or licorice root extract in agricultural plants. Nevertheless, there is inadequate data on the combination of these two methods for *Rosa hybrida*. In addition, the application of the two methods can be influenced by the environment [8].

In this research, the objective was to determine the individual and combined effects of spraying of glycine and licorice root extract on hybrid rose under heat stress. The parameters considered in the study included flower numbers, branch numbers, leaf numbers, petal number, flower diameter, stem diameter, main branch length, root length, plant fresh weight, and dry weight.

Material and Methods

This experiment was carried out inside the plastic-enclosed structure located in Ibaa Center for Agricultural Research, which is part of the Horticulture Department of the Ministry of Agriculture,in the 2025-2026 crop season.

Description of the Plant Used in the Study:

The research was conducted on the hybrid rose plant (*Rosa hybrida*). The complete plant seedlings (propagation seed) were obtained from Al-Krayat Nursery of Baghdad, Iraq. Upon harvesting, the seedlings had an estimated age of 6-12 months and reached a height of 65-70 cm, possessing two to three main branches.

Planting and Cultural Practices:

A total of 162 pots with *Rosa hybrida* L. hybrids were obtained and transferred to the experimental station. The experiment was carried out for two growing seasons: the spring season (February – May) and the autumn season (September - December).

These plants were then equally divided into two groups for experiments. Eighty-one plants were assigned in the greenhouse and exposed to the shading treatment at 50%, while the other eighty-one plants were placed outside the greenhouse in an open field environment. These plants were provided with equal treatments throughout the experiment. Winter pruning was performed once, trimming the plants to a height of about 50 cm. Weeding was done frequently in order to avoid competition among the crop plants and the weeds. Biological pest management techniques were used for the control of spider mites and aphids infestation. Since the plant grew in pots, there was little that could be done in terms of soil management other than turning the soil in the pots.

The production and testing of rose plants in hybrid form within greenhouse

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environment including overall growth of the plant and collection and measurement of the flower diameter through digital caliper have been demonstrated in (Figure 1).



Figure 1. Typical examples of hybrid rose (*Rosa hybrida* L.) flowers grown under different environmental conditions: (A) flowers grown outside in full sun; (B) flowers grown inside under greenhouse conditions with 50% shading; and (C-D) magnified images demonstrating the difference in flower structure.

Factors Studied and Experimental Design

Three factors were used in the experiment. First, the light factor had two levels, which were 50% shading in the greenhouse and full light (0% shading) from outside. The second factor was foliar spraying with glycine at three concentrations of 0, 5, and 10 mg/L. Third, the licorice root extract was applied to the leaves of hybrid rose plants at three levels: 0, 50, and 100 g L^{-1}. Foliar treatments were done following a weekly pattern, with the use of either glycine or licorice root extract. In particular, a foliar application of glycine was done during one week, after which another foliar application of the licorice

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root extract was made the next week. This means that each solution was sprayed once every two weeks. The spray treatment was applied mainly on the abaxial side of the leaves, which contained most of the stomata. Treatment was done until the leaves were saturated. Diagrams of the experimental procedure are shown in (Figure 2).

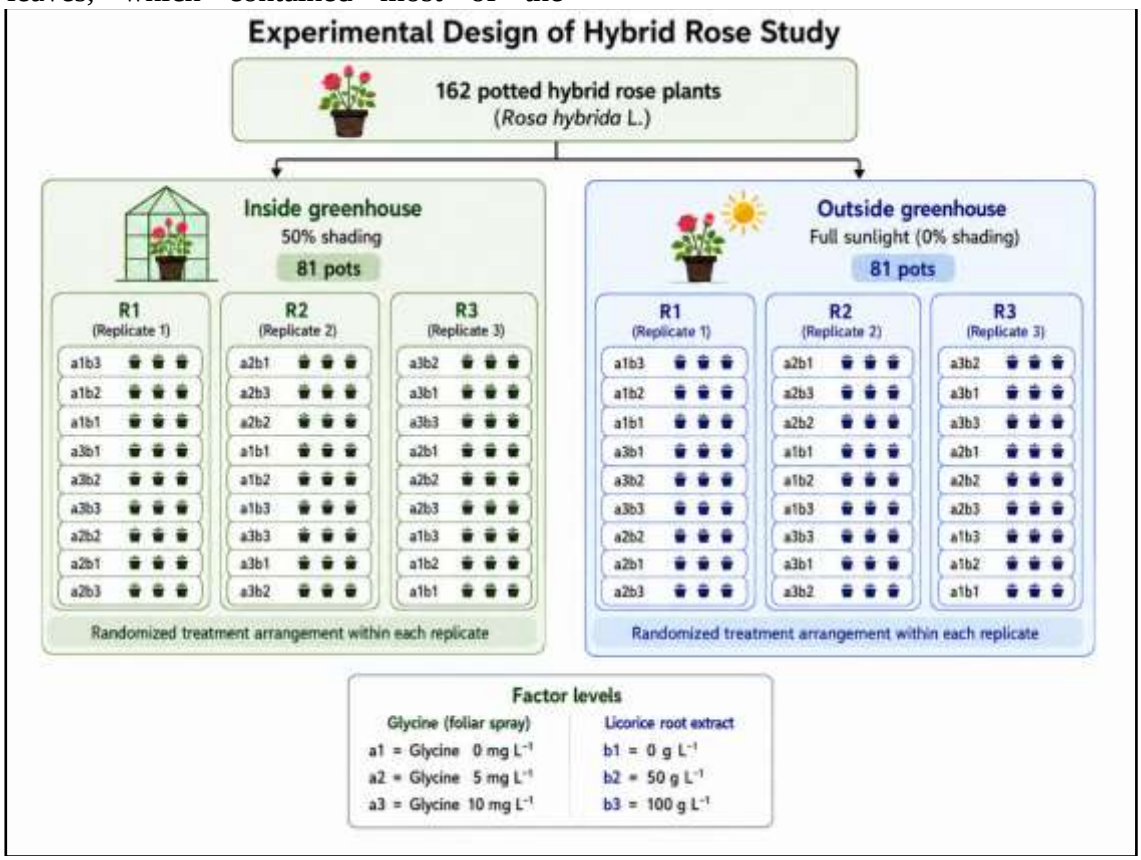


Figure 2. Design of study.

Preparation of Glycine Solutions

Solutions of glycine were made using HiMedia Hi-AR/ACS grade glycine (GRM199-100G). The concentrations 5 and 10 mg L^{-1} were made by adding 5 and 10 mg of glycine, respectively, to 1 L of distilled water, while distilled water was used as a control solution containing 0 mg L^{-1} glycine.

Preparation of Licorice Root Extract

The roots of licorice were thoroughly cleaned, dried both by air and oven drying, and then pulverized. A 500 g portion was immersed in distilled water for 72 hours. It was filtered using sterile gauze and then centrifuged. It was clarified further using filter paper. Vacuum concentration using rotary evaporation followed by drying at 50°C yielded the dry extract [9].

Studied Traits

The studied traits comprised vegetative growth traits, including number of branches (branch $plant^{-1}$), number of leaves (leaf $plant^{-1}$), main branch length (cm), stem diameter (mm), plant fresh weight (g), plant dry weight (g), and root length (cm), and flowering traits, including number of flowers (flower $plant^{-1}$), flower diameter (mm), and number of petals (petal $flower^{-1}$).

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Statistical Analysis

The experiment was conducted using a three-factor factorial arrangement (shading, glycine and licorice root extract) in a Randomized Complete Block Design (RCBD). Each trait's data was analyzed through ANOVA employing a three-way design with the use of the General Linear Model. The statistical model includes the main effects of light regime, glycine concentration, and licorice root extract concentration. It also included all the interactions up to the three-way interaction among the factors. Replicate was included in the model as a random blocking factor. The analyses were done for each season separately – fall and spring. When needed, a pooled analysis for the two seasons could be undertaken when there is equality of error variances. Normality of residuals and equality of variance assumptions were assessed through the Shapiro-Wilk and Levene's tests, respectively, prior to the analysis. Whenever the ANOVA results showed a significant effect of treatment, Tukey's HSD test was carried out for comparing the means of treatments at $P \leq 0.05$ significance level. The data is expressed as mean ± standard error. Statistical calculations were performed using IBM SPSS Statistics, version 26.0.

Results and Discussion

Figure 3 shows that 50% shading increased the mean values of all studied traits compared with 0% shading. Significant increases were recorded in the number of flowers (6.41 vs. 4.19; $P < 0.001$), number of branches (2.52 vs. 2.07; $P < 0.05$), number of leaves (93.82 vs. 72.44; $P < 0.05$), dry weight (13.81 vs. 8.89 g; $P < 0.001$), fresh weight (41.13 vs. 29.66 g; $P < 0.05$), main branch length (111.61 vs. 92.72 cm; $P < 0.01$), and root length (23.81 vs. 18.06 cm; $P < 0.001$). Flower diameter, stem diameter, and number of petals also increased under 50% shading, but these differences were not statistically significant.

Foliar glycine application significantly affected several vegetative and flowering traits of hybrid rose. Increasing the glycine concentration from 0 to 10 mg L^{-1} significantly increased the number of flowers from 4.33 to 6.17 flowers $plant^{-1}$ ($P < 0.01$), dry weight from 8.43 to 14.58 g, fresh weight from 22.35 to 47.77 g, and number of petals from 21.17 to 32.92 petals $flower^{-1}$ ($P < 0.001$). Flower diameter also increased significantly from 33.62 mm in the control to 55.43 and 55.30 mm at 5 and 10 mg L^{-1}, respectively ($P < 0.001$). Although the number of branches, stem diameter, and root length generally increased with glycine concentration, these differences were not significant. The number of leaves and main branch length also varied among concentrations without significant differences (Figure 4).

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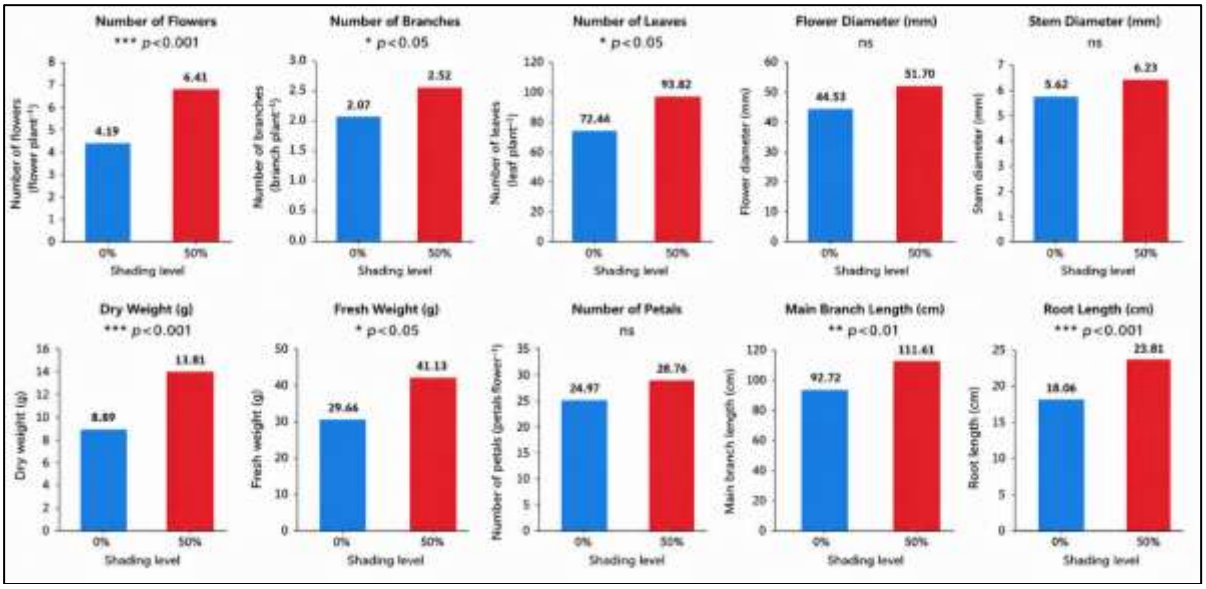


Figure 3. Effect of Shading Level on Vegetative Growth and Flowering Traits of Hybrid Rose (*Rosa hybrida* L.).

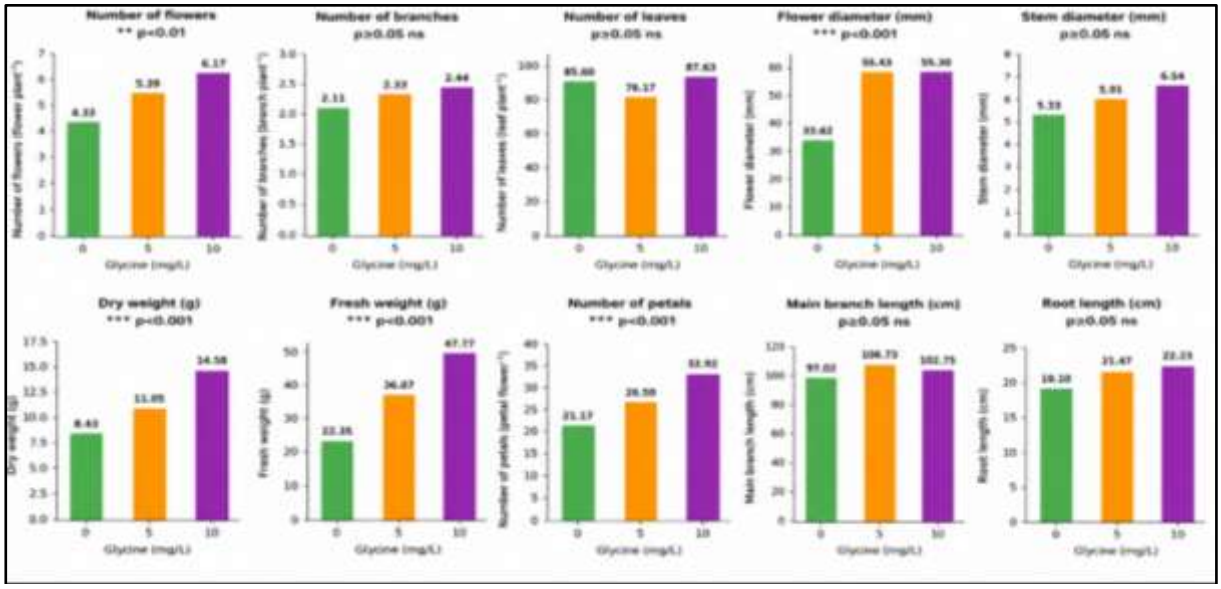


Figure 4. Effect of Foliar-Applied Glycine on the Vegetative Growth and Flowering Traits of Hybrid Rose (*Rosa hybrida* L.).

Increasing the licorice root extract concentration from 0 to 100 g L^{-1} improved most measured traits. Significant increases were observed in the number of leaves, which rose from 65.27 to 98.18 leaves $plant^{-1}$ ($P < 0.01$), stem diameter from 5.08 to 7.02 mm ($P < 0.001$), dry weight from 9.53 to 13.52 g ($P < 0.05$), fresh weight from 25.98 to 49.54 g ($P < 0.001$), and number of petals from 20.12 to 33.60 petals $flower^{-1}$ ($P < 0.001$). The numbers of flowers and branches, flower diameter, and root length also increased with higher extract concentrations, but these differences were not statistically significant. Main branch length showed only slight variation and reached its highest value at 50 g L^{-1} (Figure 5).

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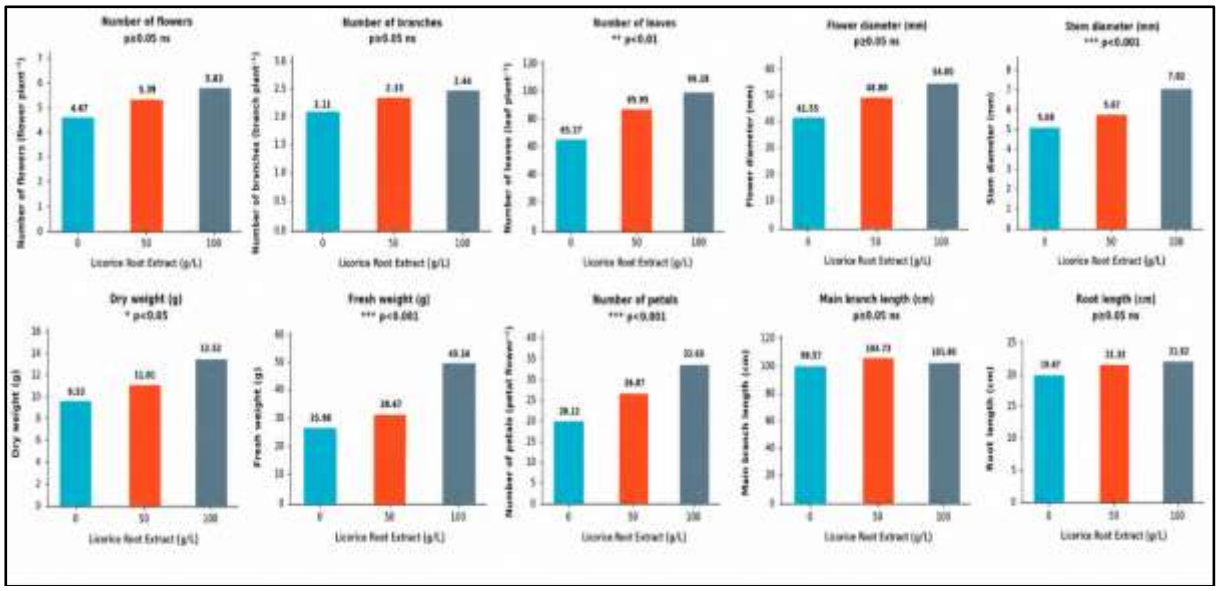


Figure 5. Effect of Foliar-Applied Licorice Root Extract on the Vegetative Growth and Flowering Traits of Hybrid Rose (*Rosa hybrida* L.)

It is clear from ANOVA F-values that the effects of shading, glycine, and licorice root extract differed among the various growth and flowering characters studied. Shading had the strongest significant effects on flower number, dry weight, root length, and, to some extent, on branch number, leaf number, fresh weight, and branch length since the values of F exceed the critical value for $\alpha = 0.05$. The significant effects of glycine were found in the flower number, flower diameter, dry weight, fresh weight, and petal number. In its turn, licorice root extract had significant effects on leaf number, stem diameter, fresh weight, and especially petal number (Figure 6).

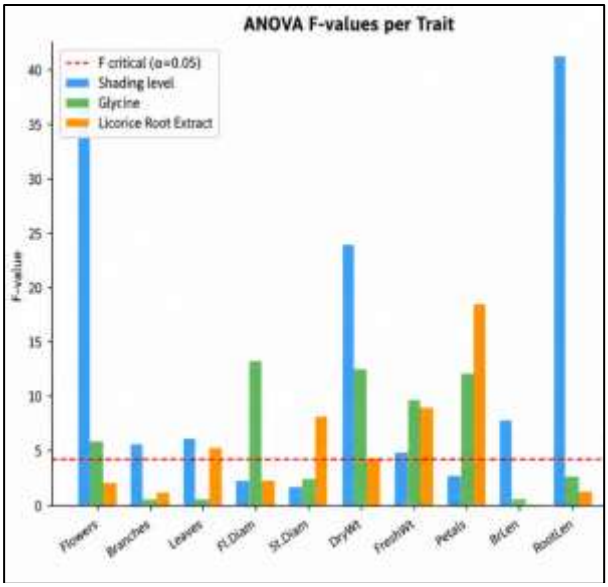


Figure 6. Comparative ANOVA F-Values of Patterns of Shading, Glycine, and Licorice Root Extract Effects on Hybrid Rose (*Rosa hybrida* L.) Traits.

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The heatmaps (Figure 7) show clear variation among the combined shading, glycine, and licorice root extract treatments. The most prominent response was observed under 50% shading combined with 10 mg L^{-1} glycine and 100 g L^{-1} licorice root extract, which produced the highest number of flowers (8.33 flowers $plant^{-1}$), branches (3.33 branches $plant^{-1}$), leaves (119.7 leaves $plant^{-1}$), and root length (26.3 cm). The largest flower diameter was recorded under 0% shading with 5 mg L^{-1} glycine and 100 g L^{-1} licorice root extract (71.8 mm). The greatest stem diameter (9.10 mm) and main branch length (141 cm) were obtained under 50% shading with 0 mg L^{-1} glycine and 100 g L^{-1} licorice root extract. The highest dry weight (18.23 g) and petal number (44.3 petals $flower^{-1}$) were recorded under 0% shading with 10 mg L^{-1} glycine and 100 g L^{-1} licorice root extract, whereas the maximum fresh weight was observed under 50% shading with 5 mg L^{-1} glycine and 100 g L^{-1} licorice root extract (65.05 g).

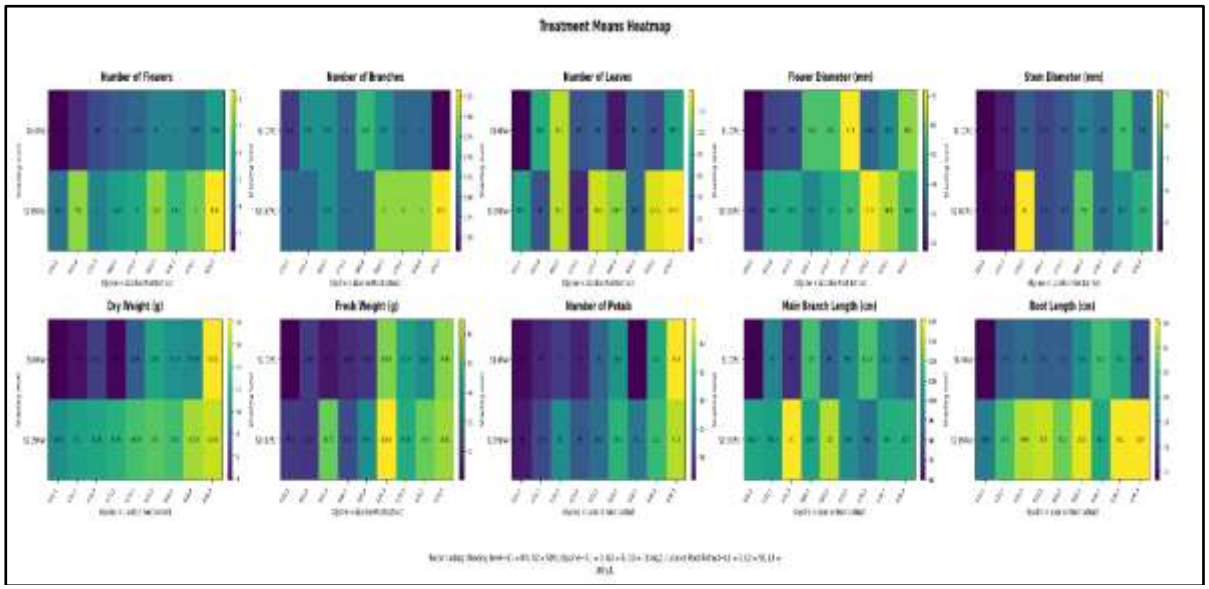


Figure 7. Heatmaps of Treatment Means for Vegetative Growth and Flowering Traits of Hybrid Rose (*Rosa hybrida* L.) under Different Shading, Glycine, and Licorice Root Extract Treatments.

This study highlights that 50% shading results in significantly higher development of most vegetative and reproductive traits in hybrid roses compared to plants grown without any shading. The considerable increase in flowers, branches, leaves, fresh/dry weight, length of main branches, and roots indicates that moderate shading provides a better growth environment for rose plants. This observation is consistent with findings by Al-Rawi & Al-Marsoumi [10], [11], who found that 50% shading, especially when combined with biostimulants and fertilizers, increased the diameter of the flower, the number of petals, flowering stem length, flower longevity, and anthocyanin production in the shrub roses. They suggested that shading reduces the environmental stress that facilitates vegetative and reproductive development of rose plants under nutrient conditions.

The current shading experiment is not in line with previous results. In accordance with Rupasinghe *et al.* [12], the application of 25% Aluminet shading increased flower production and flower size, while 50%

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black shading decreased the flower production and flower diameter. Similarly, Hou *et al.* [13] noted that shading resulted in delayed flowering, lowered flower fresh weight, and anthocyanins and flavonoids. Moreover, El-Naggar *et al.* [8] determined that full sunlight resulted in the highest flower production and fresh weight of *Rosa hybrida*. Possible reasons for such discrepancies may include varying shading levels and materials, specific reaction of the selected cultivar, ambient temperatures, and interrelation between lighting and heat stress. In the current experiment, it seems that a shade of 50% can solve problems such as too much heat and water loss without hindering growth of the plant in relation to its light needs. This can mean that the increase in biomass, number of leaves, and root length is because of proper regulation of water, thermal stress relief, and better assimilation of nutrients. The lack of significance in response to flower diameter, stem diameter, and petal number to shading alone implies that shading contributes only to plant and flower growth but not flower quality.

Glycine spray increased a number of agronomic properties including flower number, fresh biomass, dry biomass, flower diameter, and flower petal numbers. The increase in the flower diameter due to glycine spray along with an increase in the flower petals indicated that glycine not only increased the biomass production but also the development of flowers. These findings are similar to the observations of Salih *et al.* [14] where glycine was found to increase the flower number, flower diameter, flower stalk length, flowering period, and flower clusters in *Jasminum sambac*. This reaction was said to result from the role of amino acids in providing nitrogen and in osmotic regulation, oxidation resistance, membrane stability, and protein/enzyme stabilization. Similarly, the increase in fresh and dry weight can be explained in the same light since glycine could possibly have promoted cell growth and metabolism, thus dry matter production.

Positive impact of glycine is consistent with the results reported by Khattab *et al.* [15] in which amino acids were found to enhance floret numbers, floret diameters, flower duration, floret dry weights, spike lengths, and spike dry weights of gladiolus flowers. Their reasoning that amino acids serve as the source of nitrogen, carbon, and energy besides improving the enzyme activities and hormone processes is in line with the current experiment. Likewise, Abd-Elkader *et al.* [16] and Geshnizjani & Khosh-Khui [17] noted that flowers treated with amino acids exhibited improvements in flower diameter, flower numbers, stem lengths, photosynthetic pigments, and physiological performance in gerbera. Therefore, the effect of glycine on the hybrid rose in the current experiment can be understood in terms of higher nitrogen metabolism, increased photosynthesis, and better translocation of source-sink towards developing florets. The fact that there are no statistically significant differences in the number of branches, stem thickness, root length, number of leaves, and the length of the main branch shows that glycine is better in modifying reproductive and biomass characteristics than plant structure.

Licorice root extract increased all parameters except for scape length, with significantly higher values being obtained for number of leaves, stem diameter, fresh weight, dry weight, and petals. This is in agreement with Babilie *et al.* [18], who found that the use of licorice-root extract through foliar feeding resulted in increased height of scape, umbel diameter, number of flowers in an umbel, and flower set in onion seed production. As far as their interpretation of these findings was that licorice root extract contains growth-promoting compounds and nutrients thus

promoting leaf growth and redirecting carbohydrates to reproduction, it can be considered highly relevant to the findings of the present research.

This current result is supported by Mostafa and Rida [19], who indicated that natural extracts such as licorice increased vegetative growth and flowering activity in Pentas lanceolata. Also, Alwan *et al.* (2026) indicated that using licorice extract along with garlic extract resulted in increasing flower diameter, flower weight, plant height, number of leaves, and chlorophyll concentration in chrysanthemums. Furthermore, Ahmed *et al.* [20] stated that using licorice root increased branching, leaves development, leaves area, flower diameter, flowers number, and chlorophyll concentration in Chinese carnation. Such studies associate the positive impacts of licorice with the presence of compounds such as sugars, minerals, flavonoids, glycyrrhizin, and gibberellins, which may stimulate cell division, photosynthesis, and general plant health. Such findings are consistent with those of the current study, considering the fact that licorice extract greatly contributed to increasing the number of leaves, diameter of stem, and weight of the plant, thereby implying improved vegetative growth and development for flowering. The fact that licorice did not significantly increase the number and size of flowers may suggest that licorice extracts only stimulate general plant health and petal development.

The following partial contradiction can be drawn from the works by Al-Ma'athedi *et al.* [21]. The researchers found that many growth regulators increased the productivity of carnations and the greatest productivity was achieved through the use of salicylic acid, rather than licorice extract. Even though it does not contradict the findings of the present research directly, this information indicates that the efficiency of the usage of licorice in comparison to other regulators depends on many factors, including the plant variety, concentration of licorice, stress, and other growth regulators used for comparisons. Therefore, licorice extract is an efficient biostimulant.

The interaction between shading, glycine, and the licorice root extract is such that the highest positive results in terms of vegetative and reproductive growth were achieved by using 50% shading in conjunction with glycine and licorice root extract based on the flower count, branch count, leaf count, and root length. Such interaction may be interpreted as synergistic interaction where stress is alleviated by environmental factor while biochemical factor stimulates the physiological processes. It can be argued that shading helps to reduce heat stress and transpiration, glycine assists in nitrogen metabolism and osmoregulation, and licorice extract provides necessary nutrients.

At the same time, it is not true that the maximum values of all the characteristics have been reached as a result of the sole interaction. Thus, the maximum flower size, the maximum dry mass and also the maximum number of petals have been reached due to the presence of no shade combined with glycine and licorice. Meanwhile, the maximum value of fresh mass was reached due to 50% shade with glycine and licorice. These results indicate that some quality features of flowers can be further developed with increasing light exposure provided that proper biostimulants are used. This explanation facilitates the synthesis of the present results with the literature recommending full sun exposure, including that reported by El-Naggar *et al.* [8] and Al-Rawi and Al-Marsoumi [10], [11]. On a broader scale, the data clearly show that the performance of hybrid roses depends on

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the interaction of light, heat stress reduction, and biostimulant availability rather than on one component alone.

Conclusion

In the given experimental condition, the application of 50% shading gave rise to better values in several characteristics related to vegetation and flowering for the rose hybrid in comparison with full sunlight conditions. While glycine at a concentration of 10 mg L^{-1} increased flower number, flower diameter, fresh and dry weight, and petal number, licorice root extract at 100 g L^{-1} increased leaf number, stem diameter, fresh and dry weight, and petal number. When 50% shading along with 10 mg L^{-1} glycine and 100 g L^{-1} licorice root extract were used together, the best results were observed in case of flower number, branch number, leaf number, and root length.

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