Effect of Mycorrhizal Fungi on Fertilization, Growth and Essential Oil of Taif Rose under Salinity Stress in KSA

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Abstract: *Arbuscular mycorrhizal* fungi (AMF) establishing beneficial symbiosis with most plant roots have gained a growing interest as soil providers able to increase crop productivity and quality. Our results stated that AMF with NPK improve agriculture productivity of Taif rose (*Rosa damascena trigintipetala* Dieck), by enhancing plant growth and quantity of some essential oils. The tested NPK concentrations enhanced the all growth parameters of rose plant in the presence of mycorrhizal fungi compared with the control. In contrast, increasing of NaCl as a stress treatment to rose development decreased all development parameters except flower length. Combination of NPK and NaCl of Taif rose plant decreased survival percentage, plant height and flower length of rose plants under the infection of mycorrhizal fungi.
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1.Introduction

Taif rose (*Rosa damascena trigintipetala* Dieck) is considered one of the most important economic products of Taif, Saudi Arabia. There are almost 200 species and more than 18000 cultivars in the genus Rosa (1). They are mostly shrubs, distributed in the temperate zones of the Northern hemisphere (2). In recent years, antioxidant, antibacterial and antimicrobial activities of *R. damascena* essential oil have been demonstrated (3-6).

Salinity is a major a biotic environmental stress that is reported to be responsible for reducing plant growth across the globe. Sea water intrusion, in coastal states, has imposed salinity problems in turf grass culture (7, 8). Sodium chloride (NaCl) is the major compound contributing salinity in soils, and more salt-tolerant turf grasses are required to cope this problem (9).

Salt accumulation in soils, limitations on use of groundwater, and salt water intrusion into groundwater may restrict cultivation of glycophytic crops in these areas (10). Salinity lowers water potential and restricts of water to plants (11). Presence of excessive salt (NaCl) outside the cell can induce an osmotic stress, which may adversely affect the plant growth (12).

Salinity limits crop metabolic activities, hampers plant growth and synthesis of secondary metabolites. It also affects the osmotic potential in the root zone of plants (13). It induces ion toxicity due to excessive uptake of sodium ions (14). The suitability of water for irrigation is determined not only by the total amount of salt present but also by the kind of salt (15).

Many plant species suffer a decline in growth when exposed to NaCl stress. The inhibition of growth in long-term exposures (days) can result from osmotic effects on water availability, reduction in net $C0_2$ assimilation, specific ion effects, or ion imbalance due to interference with uptake of essential nutrient ions (16).

Sodium chloride (NaCl) is the major compound contributing salinity in soils, and more salt-tolerant turfgrasses are required to cope this problem, (17).

Generally, plants have developed different adaptive mechanisms to mitigate salinity under the saline environments, (18).

Arbuscular mycorrhizal (AM) symbioses occur between most terrestrial plants and fungi of the phylum Glomeromycota . The interaction starts with a fine-tuned signal exchange between the two symbiotic partners. Prior to physical contact, the host secretes signal molecules, termed branching factors, that stimulate hyphal growth and branching, while the fungi synthesize and release the so-called Myc factors, which trigger host responses. Despite the recent identification of the major signals used by the host and micro symbionts, the complete spectrum of signaling molecules remains elusive.

The identification of the Myc factors used by mycorrhizal fungi to induce host responses involved a tremendous amount of effort.

It have been found that *Arbuscular mycorrhizal* fungi (AMF) stimulate growth and improve pathogen, heavy metal, and salinity resistance, as well as to influence the level of secondary metabolites in plants (19).

It has been successfully utilized for the evaluation of the role of AMF inoculation on plants, including those of medicinal importance (20-23). (24) mentioned that a mixture of clay + peat was the most effective medium for increasing the percentage of N, P and K in leaves of *Eucalyptus camaldulensis*. (25) found that planting the cutting in sand or clay composted leaves (1: 1) increased the content of N in the leaves and stems of *Philodendron erubesens* c.v. (Emerald Queen). The lowest value of N or P content was recorded with sandy medium.

(26) stated that leaf nutrients (N, P, K, Ca and Na) of *Adhatoda vasica, Nerium oleander* and *Lantana camara*, tended to increase with sand/clay 1:1 compared to sand/clay at 2:1.

Several components were isolated from flowers, petals and hips (seed-pot) of *R. damascena* including terpenes, glycosides, flavonoids, and anthocyanins. This plant contains carboxylic acid , myrcene , vitamin C , kaempferol and quarcetin.. The identified compounds were; β-citronellol, geraniol , nerol and phenyl ethylalcohol .

This study was conducted to determine the effects of NPK as well as NaCl at different concentrations as well as mycorrhizal treatment on growth and chemical composition of Taif rose in KSA.

2.Material and Methods

Pot experiment:

This investigation was carried out in Biology Department, Faculty of Sci., Taif Univ., during 2012 and 2013. This study designed to investigate the growth nodes of rose plants cultured in Taif region in KSA. Nodes were cultured on pots (3L) containing sand and peatmoss (1:1). Each pot infected with 25g of mycorrhizae inoculum. These native *Arbuscular mycorrhizal* fungi (AMF) isolated from the rhizosphere of maize (*Zea mays* L.) grown in agricultural soils of National Research Centre, Sakha, Egypt. After two weeks, pots were subjected to different concentrations of NPK (0.0, 250, 500, 750 and 1000 ppm) and/or NaCl (0.0, 250, 500, 750 and 1000ppm).

After 6 months of cultivation some morphological, physiological and chemical composition parameters like plant height, shoot length, number of flowers, size of flowers and plant survival were recorded.

Chemical analysis:

a-Rose essential oil was extracted by using 50 gm sample from whole flower (petals and sepals) dissolved in N- hexane and taken 20 mg +1 ml hexane to make analysis .

b- Gas chromatography –Mass spectrometry analysis:

The analysis of the samples were performed using a Varian GC-MS system (Model Varian CP 3800, Varian Saturn 2200 and Combi Pal auto sampler system, USA) equipped with VF-5 fused silica capillary column (30m x 0.25 i.d. mm film thickness0.25um, Varian). For GC-MS detection, an electron ionization system with ionization energy of 70e V was used. Helium gas was a carrier gas at low rate of 1ml/min. injector and mass transfer line temperature were set at 250 and 300ºC, respectively. The oven temperature was programmed for 5 minute at 60ºC, 60ºC to 250ºC/minute and held for 5 minute finally at 260ºC, solvent delay time 3 min. the injection of the all samples were carried out with the auto-sampler for 1 µl with a split ratio 1/20. The conditions of analysis and specification of the instrument were optimized for better separation and resolution. Identification of components of concrete and absolute oils was based on matching with Wiley and NIST electronic library.

Statistical analysis:

All results were subjected to one-way ANOVA and the means were compared according to the Student–Newman–Keuls (SNK) multiple range test ($P \leq 0.05$).

3.Results

Effect of NPK and NaCl concentrations on growth and development of Taif rose infected with mycorrhizal fungi.

Many parameters were used to evaluate the effect of both NPK and NaCl on *Rosa damascena trigintipetala* development in Table (1) . Concerning of NPK fertilizations, survival (%), leave number and flower length varied insignificantly compared with the control. In contrast, significant increase in flower number was observed by increasing NPK concentration. The highest value obtained at 1000ppm. About 76.6% increases of flower number were recorded more than the control. All tested NPK concentrations enhanced the height of rose plant compared with the control. the highest value was stated at 750ppm (101.3 cm). Generally, addition of NaCl as a stress treatment to rose development decreased all development parameters except flower length. A significant reduction in survival (%) of rose plants was recorded by all NaCl treatments. About 46.6% of reduction was observed using 750ppm as a salinity stress agent (Table 1). Also, 39% reduction in rose height was recorded by 750ppm of NaCl. In contrast, the plant height when used 250ppm varied insignificantly compared with the control. Leaves number was decreased significantly by increasing

NaCl concentration. The maximum reduction in leaf number was observed by 1000ppm, it was 59.6% compared with the control. While, insignificant variations were observed in flower number among NaCl concentrations. Among the tested NaCl concentration, 1000ppm exhibited the inhibitoriest effect of flower length of rose, that caused 53% reduction compared with the non-treated control.

These results indicate that the vegetative and growth characters of Taif rosa are affected by using various doses of NPK and found much helpful to improve plants and found low effect by using Nacl treatments. Similar finding was recorded by some investigators (27,28) .

Effect of combination of both NPK and NaCl concentrations on growth and development of Taif rose under the infection of mycorrhizal fungi .

Combination of NPK and NaCl decreased survival percentage, plant height and flower length of rose plants under the infection of mycorrhizal fungi. Survival percentage decreased insignificantly among the treatments(Table 2). The highest survival percentage (98%) obtained by the control. Plant height at 1000/1000ppm decreased significantly by 24% over than those of infested non-treated control. Also, about 27% reduction in leave number was recorded 1000/1000ppm in comparison with untreated control.

It has been shown that, the combination of NPK and NaCl at 250/250ppm enhanced flower formation of rose plants, but it varied significantly compared with the control. The other treatments decreased flower number. But this reduction varied insignificantly comparing with the control.

The treatments of 250/250 and 500/500ppm decreased flower length of rose plants insignificantly in comparison with non treated control, but significant reductions were observed by both 750/750 and 1000/1000ppm of NPK/NaCl. About 42% reduction in flower length was recorded by 750/750ppm compared with non treated control. In general, the difference between growth parameters of taif rose at low salinities and that in deionized water is insignificant, but they gradually decrease with further increase of salinity . A similar findings was recorded by some investigators(29,30) .

Effect of some NPK and Nacl concentrations on chemical compositon of Taif rose under the infection of mycorrhizal fungi .

Table (3) reflected the effect of different NPK and Nacl concentrations on chemical composition. It is showed that phenyl ethyl alcohol percentage was recorded 5.2% with control plants ,and increased to 8.5% with NPK at 500ppm. However, increasing Nacl to 250 ppm caused decrease to 2.1% . Data also showed that B-citronellol +Nerol was increased to 23.85% with plants fertilized with NPK at 500 ppm than control plants which recorded 16.6% or plants exposed to 250 ppm of Nacl which reached 9.9 % .

Above results indicates that there was a direct relation between the adding Nacl concentrations to media and low accumulations of important rose component in the cultured plantlets. These results explain that NPK at 500 ppm in the medium encouraged the cultured plantlets to increase the phenyl ethanol and B.citronllol +Nerol . These results are in agreement with the findings with (31) on geranium plants.

Table (1): Effect of NPK and NaCl concentration on growth and development of Taif rose under the infection of mycorrhizal fungi.

Concentration	Survival (%)		Plant height (cm)		Leaves number		Flower number		Flower length (cm)	
(ppm)	NPK	NaCl	NPK	NaCl	NPK	NaCl	NPK	NaCl	NPK	NaCl
Control	96.7^{a}	97.0^{a}	76.7^{b}	60.7^{a}	43.0^{a}	42.3^{a}	4.7^{c}	3.7^{a}	1.9^{a}	1.9^{a}
250	96.0^{a}	77.7^{b}	96.3^{a}	54.7^{a}	48.0^{a}	40.0^{a}	5.7^{bc}	3.7^{a}	2.2^{a}	1.3^{b}
500	97.7^{a}	73.0^{b}	100.7^{a}	42.0^{b}	40.0^{a}	40.0^{a}	6.3^{bc}	4.0^{a}	2.0^{a}	1.0^{b}
750	93.3^{a}	52.7^{c}	101.3^{a}	37.0^{b}	38.0^{a}	25.3^{b}	5.7^{bc}	3.7^{a}	2.2^{a}	1.0^{b}
1000	92.0^{a}	57.3^{c}	96.0^{a}	37.7^{b}	42.0^{a}	17.7^{b}	8.3^{a}	3.0^{a}	1.7^{a}	0.9^{b}

Different letters on the same column are differ significantly at $p \leq 0.05$.

Table (2): Effect of the combination of both NPK and NaCl on growth and development of Taif rose under the infection of mycorrhizal fungi.

Concentration (ppm)NPK/NaCl	Survival (%)	Plant height (cm)	Leaves number	Flower number	Flower length (cm)
Control	98.0^{a}	82.7^{a}	32.0^{ab}	4.0^{a}	1.9^{a}
250/250	93.0^{a}	78.3^{ab}	35.7^{a}	4.7^{a}	1.6^{ab}
500/500	91.3^{a}	65.3^{b}	31.0^{ab}	4.3^{a}	1.4^{ab}
750/750	84.0^{a}	64.7^{b}	29.3^{ab}	4.0^{a}	1.1^{b}
1000/1000	84.3^{a}	63.0^{b}	23.3^{b}	3.0^{a}	1.2^{b}

Different letters on the same column are differ significantly at $p \leq 0.05$.

Table (3): Effect NPK and NaCl on chemical composition percentage of Taif rose under the infection of mycorrhizal fungi.

Chemical composition %	Untreated plants		Plants with 250 ppm Nacl		Plants with 500 ppm NPK	
Phenyl Ethyl Alcohol	5.2		2.1		8.5	
B.citronellol +Nerol	16.6		9.9		23.85	

Conclusion

According to the results obtained in the present study ,it could be concluded that exposing the Taif rose explants under this study to NPK caused improve in the growth parameters and chemical composition under using mycorrhizal fungi. , also using NaCl with or without NPK concentrations decrease in growth and chemical contents under using mycorrhizal fungi.

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