Effects of the inoculation with arbuscular mycorrhizal (AM) fungus of the genus Glomus on growth and leaf mineral concentrations of grapevine (*Vitis vinifera* cv. Cabernet Sauvignon)

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Key words: AM-mycorrhiza, vineyard, nutrient uptake, growth parameters

Abstract

The effects of root inoculation with mycorrhizal fungi (genus Glomus) on growth and leaf mineral concentrations of grapevine (Vitis vinifera cv. Cabernet Sauvignon) were studied under the growth conditions of Central Chile. Inoculation enhanced the uptake of N and K and vegetative growth but decreased the foliar concentration, but not necessarily the uptake of P.

Introduction

The beneficial effects of arbuscular mycorrhizal inoculation with respect to grapevine growth and the nutritional status have been reported by several authors (Karagiannidis et al. 1995, Biricolti et al. 1997, Karagiannidis and Nikolaou 1999). No studies have been carried out in Chile about this symbiosis in grapevines. The objective of the present study was to assess the effects of root inoculation with mycorrhizal fungi (genus Glomus) on growth and leaf mineral concentrations of grapevine (*Vitis vinifera* cv. Cabernet Sauvignon) under the growth conditions of Central Chile.

Materials and methods

The study was carried out between 2005 and 2006 at the Department of Agronomy of the Universidad Católica del Maule at the City of Curicó. Curicó is situated in Central Chile. Geographic coordinates: 34°58' S; 71°14' E; 228 m above sea level. An unsterilized soil from the Research Station of the University was used for the study. It corresponds to a sandy clay soil from the Vertisols class with a total humus content of 6.31 % and pH/H₂O 5.62. The soil was sieved (2mm), not sterilized and mixed with sand at a ratio of 1:1. 20 kg of the resulted substrate was employed for every plant. The mineral content of the mixed substrate (mg kg⁻¹) was: P (35.0), K (149.0), Ca (672.0), Mg (102.0), Fe (113.5), Mn (3.18), Cu (2.78) and B (1.24). The plants were planted during late March 2005 in 20 L plastic pots. 10 grapevine plants (One year old) were utilized for every treatment, with every single plant as replicate. Plants were inoculated with a commercial granular inoculant (Mycosym Tri-ton®). The inoculant is composed of AM from the Glomus genus and is made with spores and small pieces of mycorrhizal roots fixed on a mix of perlite and sand. Similar Inoculants have been positively evaluated in apple trees (Von Bennewitz et al. 2000a, Von Bennewitz et al. 2000b)

When the grapevines were planted the inoculant was mixed with the soil at different depths to assure a direct contact with the roots. The treatments applied for the experiment were the following: Control, T1.- Inoculant 2,5 ml/plant, T2.- 5,0 ml/plant, T3.- 10,0 ml/plant. The following measurements were carried out: 1. Mycorrhizal

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infection. Samples of fresh roots were taken on 28.09.5, 28.11.05 and 16.01.06 and cut into 1.0-cm segments. All segments of each treatment were massed together, cleared with 2% KOH, and stained with tripan blue (Phillips and Hayman 1970). Fifty samples for each treatment were analyzed and the percentage of root length containing vesicles was assesed by the gridline-intersect method (Giovanetti and Mosse 1980), mounted on microscope slides and estimated by observation with 200 X magnification.

2. Foliar analysis. Included: N, P, K, Ca, Mg, Fe, Mn, Cu, and Zn (leaves sampled at veraison). Chemical methods. Leaf samples were cleaned; owen dried at 60 °C and reduced in particle size. Wet digestion was utilized for organic matter destruction in the case of N, P, K, Ca, Mg and dry ashing for Fe, Mn, Cu and Zn. N (Kjeldahl), P (Colorimetric method), K (Flame photometer), Ca, Mg, Zn, Fe, Cu, Mn (Atomic absorption spectrophotometer).

3. Vegetative and generative responses included measurements of shoot extension growth (two shoots per plant), increase in trunk diameter, length and diameter of nodes. Also number and area of leaves, according to the procedure of Gutierrez and Lavín (2000) and pruning weight (fresh and dry weight). Results were subjected to analysis of variance and the Tukey's multiple range test was employed in the case of significant differences to separate means.

Results and discussion

Root mycorrhizal infection. Results of this evaluation are presented in table 1.

Tab. 1: Mean mycorrhizal infection in roots of Cabernet Sauvignon grapevine (various dates)

Treatments Date of evaluation

| | 28.09.5 | 28.11.05 | 16.01.06 |
|------------------|---------|----------|----------|
| Control | 6.0b | 8.0c | 16.8bc |
| T1: 2,5 ml/plant | 6.0b | 17.2b | 14.8c |
| T2: 5,0 ml/plant | 14.4a | 29.6a | 23.2b |
| T3: 10 ml/plant | 17.6a | 24ab | 34.8a |

Values marked by the same letters in column are not statistically different (P \leq 0.05) according to Tukey's test.

During both years (2005 and 2006) all treatments showed mycorrhizal infection, even the Control treatment where no AM were applied. These results reflect the presence of native mycorrhizas in the soil of the study, with activity in roots of grapevines. The greater degree of root infection was observed in treatment T3 (Highest doses of innoculum). The percentage of root length infected increased markedly during the second year in most of the cases, even in the control treatment (6.0%-16.8%). These results showed that the natural population of mycorrhiza of the soil can increase from year to year if appropriate conditions are given (minimal soil disturbance, no application of soil herbicides, adequate fertilization among others). According to the results of this study the AM fungus contained in the inoculant seem therefore to be able to infect grapevine roots, and the responses are significantly greater than the control without application. It seems that one application of the inoculant at the

moment of plantation is sufficient, if colonization has been taken part successfully, to secure mycorrhizal root colonization, but not great differences (increase or decrease of the infection) occur from year to year once the products have been applied.

Foliar mineral content. Results of this evaluation are presented in table 2.

Tab. 2: Foliar mineral content

| Т. | % | | | | mg kg⁻¹ | | | | |
|----|-------|-------|-------|--------|---------|--------|--------|------|-------|
| | N | Р | К | Са | Mg | Fe | Mn | Cu | Zn |
| C. | 1.59c | 0.22a | 0.86b | 1.17ab | 0.27ab | 117.6a | 120.0a | 5.0a | 11.3a |
| T1 | 1.81b | 0.18b | 1.03a | 1.06ab | 0.27b | 121.6a | 85.6b | 5.0a | 11.3a |
| T2 | 1.96a | 0.18b | 1.07a | 0.95c | 0.24c | 118.0a | 74.0b | 6.0a | 12.3a |
| Т3 | 1.60c | 0.19b | 0.86b | 1.26a | 0.28a | 116.3a | 126.3a | 4.6a | 12.0a |

Values marked by the same letters in column are not statistically different (P \leq 0.05) according to Tukey's test.

N, K: Values were significantly higher in the case of treatments T1 and T2. These results are similar with those reached by Alarcón et al. (2001). These authors observed significantly higher foliar concentrations of N and K in treatments with AM in grapevine. P: Values were significantly lower in those treatments where AM were applied. In these cases the inoculation could have decreased the concentration, but not necessarily the uptake of P. We have to consider the dilution effect produced by the great shoot growth (table 4). If the P foliar content and the pruning dry weight, are considered together, we could estimate the total P concentration of the above ground section of the plant. In this case the concentration of P is significantly higher in inoculated plants (table 3). Ca: Was significantly lower in the case of treatment T2. Fe, Cu y Zn: Were not significantly affected by the treatments. Mg y Mn: Concentration decreased in the case of treatments T1 y T2.

Tab. 3: Estimation of the total P concentration in the above ground section of the plant

| Treatments | mg |
|------------------|--------|
| Control | 19,2 c |
| T1: 2,5 ml/plant | 25,1 b |
| T2: 5,0 ml/plant | 30,4 a |
| T3: 10 ml/plant | 30,1 a |

Values marked by the same letters in column are not statistically different ($P \le 0.05$) according to Tukey's test.

Vegetative responses. Results of this evaluation are presented in table 4.

Significant differences were detected for the evaluations of pruning weight (up to 93% increase). No significant differences were detected for trunk diameter, length and diameter of nodes, number and area of leaves.

Tab. 4: Pruning weight (dry weight)

| Treatments | |
|---|-----------------------------------|
| Control | 8,72 b |
| T1: 2,5 ml/plant | 13,68 a |
| T2: 5,0 ml/plant | 16,89 a |
| T3: 10 ml/plant | 15,34 a |
| Values marked by the same latters is column are not statistically | different (D < 0.05) according to |

Values marked by the same letters in column are not statistically different (P \leq 0.05) according to Tukey's test.

Conclusions

The studied inoculant has the capacity to colonize and persist in the roots of grapevine (AM-mycorrhizal fungus), enhance the concentration of P of the above ground section of the plant, the foliar concentration of N and K and improve the vegetative growth. They could decrease the foliar concentration, but not necessarily the uptake of P. These results obtained in experimental conditions and young plants have a practical importance. The studied inoculant could be applied in the nursery, where moderate amounts of colonization are often naturally achieved and in the vineyard after the transfer of these plants to a low-nutrient environment they could spread and enhance plant growth and production.

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