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## MINICUTTING OF CHARDONNAY GRAPEVINE AND SO4 GRAPEVINE ROOTSTOCK

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**ABSTRACT:** The use of grapevine minicutting is an alternative for quality plant production, being similar to conventional cutting, however, has advantages such as reduced physical space, higher rooting quality and reduced or eliminated use of plant growth regulators. This technique has been gaining ground as one of the most used forms of vegetative propagation. The objective of this study was evaluate the rooting of herbaceous minicuttings of Chardonnay grapevine and SO4 grapevine rootstock by using different concentrations of indolebutyric acid (IBA), verifying the viability of minicutting for clonal mini-garden formation. The experimental design was randomized blocks with four IBA concentrations (0, 500, 1000 and 1500 mg L<sup>-1</sup>). The minicuttings remained in trays containing vermiculite and kept in a micro sprinkler chamber. After 60 days, the survival percentage, rooting and callus formation, root number and average length, shoot number and average length were evaluated. For Chardonnay, the percentages of survival and rooting varied between 93 and 100% in all treatments. For SO4, the IBA concentration of 1500 mg L<sup>-1</sup> negatively affected the minicuttings survival and rooting. It is possible to root herbaceous Chardonnay grapevine minicuttings and SO4 grapevine rootstock without IBA application.

**Keywords:** *Vitis* spp., vegetative propagation, cuttings, indolebutyric acid.

## MINIESTAQUIA DE VIDEIRA CHARDONNAY E PORTA-ENXERTO DE VIDEIRA SO4

**RESUMO:** O uso da miniestaquia de videira é uma alternativa para a produção de plantas de qualidade, pois é semelhante à estaquia convencional, no entanto, apresenta vantagens como a redução do espaço físico, maior qualidade de enraizamento e redução ou eliminação do uso de reguladores de crescimento. Esta é uma técnica que vem ganhando espaço como uma das formas mais utilizadas de propagação vegetativa. O objetivo deste trabalho foi avaliar o enraizamento de miniestacas herbáceas de videira Chardonnay e do porta-enxerto de videira SO4 com o uso de diferentes concentrações de ácido indolbutírico (AIB), verificando a

*viabilidade da miniestaquia para formação de um minijardim clonal. O delineamento experimental utilizado foi blocos casualizados com quatro concentrações de AIB (0, 500, 1000 e 1500 mg L<sup>-1</sup>). As miniestacas permaneceram em bandejas contendo vermiculita e mantidas em câmara de microaspersão. Após 60 dias, foram avaliados os percentuais de sobrevivência, de enraizamento e de formação de calo, número e comprimento médio de raízes, número e comprimento médio de brotações. Para a Chardonnay, as porcentagens de sobrevivência e enraizamento foram entre 93 e 100% em todos os tratamentos. Para o SO4, a concentração de 1500 mg L<sup>-1</sup> de AIB afetou negativamente a sobrevivência e o enraizamento das miniestacas. É possível enraizar miniestacas herbáceas de videira Chardonnay e do porta-enxerto de videira SO4, sem necessidade de aplicação de AIB.*

*Palavras-chave: Vitis spp., propagação vegetativa, estaquia, ácido indolbutírico.*

## INTRODUCTION

Cutting has been the most used propagation technique in the production of grapevine plants, both for table grape and wine varieties for obtaining the rootstocks, with posterior grafting of the variety to obtain the finished plant (PIRES; BIASI, 2003; FRONZA; HAMANN, 2015). Although viticulture has expanded worldwide in the last decades, the system of production of plants has not shown great advances. However, to observe such progress, nurserymen will have to enhance plants production and quality (WAITE et al., 2015).

The formation of clonal mini-gardens in a protected cultivation system may be an alternative to obtain quality plants. The minicutting is the propagation of shoots (minicuttings) of plants obtained by cutting, minicutting or seeds (ALTOÉ et al., 2011; SÁ et al., 2018). This technique allows to obtain plants with a quality standard for the formation of clonal mini-gardens (stock plant), which will be the sources for propagating material for the production of new plants.

The use of clonal micro-gardens results in a reduction in the physical space used for the maintenance of these stock plant (mother plants), a greater efficiency in the management, an increase in the percentage of rooting of the minicuttings, even with the possibility of reducing or eliminating the use of plant regulators for rooting induction (CARVALHO; SILVA, 2012; FERRIANI et al., 2010).

However, success in rooting of a particular species may vary depending on the physiological conditions, nutritional status and age of the mother plant, the type of

cutting (woody, semi-woody or herbaceous), season, genetic potential of rooting and hormonal balance (HARTMANN et al., 2011).

Exogenous application of growth regulators, such as IBA (indolebutyric acid), NAA (naphthaleneacetic acid) and IAA (indoleacetic acid) is one of the most used methods to promote the hormonal balance and, consequently, to promote rooting. indolebutyric acid (IBA) is most used one because it is a photostable substance with a local action and less sensitive to biological degradation, in relation to other synthetic auxins (PIRES; BIASI, 2003; HAN et al., 2009; STUEPP et al., 2017). However, the ideal concentration to promote rooting may vary between species and between genotypes of the same species, so, it is necessary to test them for each material used.

Chardonnay is considered the “queen of white grapes” for providing complex, rich and well-structured wines. It is one of the most important varieties for the Campanha region, and in terms of production and planted area, ranks second in the region, afterwards only to Cabernet Sauvignon. SO4 is also among the most used rootstocks in Rio Grande do Sul (MELLO; MACHADO, 2017).

In order to determine the viability of minicutting for propagation of plants, it is essential to verify the rooting potential of minicuttings, the need of using exogenous auxins (OLIVEIRA et al., 2015) and the application to different genotypes. Minicuttings can be a promising alternative in the production of grapevine plants. Furthermore, the formation of clonal mini-gardens would enable the availability of uniform material with greater propagation potential.

The objective of this study was to evaluate the rooting of herbaceous minicuttings of Chardonnay grapevine and SO4 grapevine rootstock by using different concentrations of indolebutyric acid (IBA), verifying the minicutting viability for a clonal mini-garden formation.

## MATERIAL AND METHODS

The plant material used for the experiment was collected from a vineyard located in the municipality of Dom Pedrito, state of Rio Grande do Sul, Brazil (31°01'13" S, 54°36'13" W, 131 m above sea level). Besides the importance of the varieties, another factor that determined the choice of these genotypes was the suggestion of a local vine grower who works with the Chardonnay variety, using SO4 as rootstock.

Being this, who made available material for this trial. The herbaceous branches were collected, wrapped in paper moistened with water and taken to the nursery. The minicuttings were standardized at 5 to 8 cm in length, containing two buds and two leaves with their limb reduced to half.

The experimental was arranged in a randomized complete block design with four levels of IBA concentrations (0, 500, 1000 and 1500 mg L<sup>-1</sup>), containing four replications composed of fifteen minicuttings, constituting four treatments for each of the varieties, Chardonnay grapevine and the SO4 grapevine rootstock.

In the treatments with use of regulator, the base of the minicuttings was immersed for 10 seconds in IBA solution, with the respective concentrations. Then, the material was transplanted to trays (72 cells) containing vermiculite as substrate. The trays were placed in a micro sprinkler chamber in the nursery. The nursery is built with a 50% shading cloth, with dimensions (L x W x H) of 16 x 32 x 3.5 m.

The micro-sprinkler system was programmed using a timer, with pre-set time intervals with duration of 5 minutes and 8-hour intervals. The average minimum and maximum temperatures over the experiment period were 13 °C and 26 °C, respectively. The minimum recorded was 7 °C and the maximum 32 °C. Temperature measurements were performed daily using a digital thermo-hygrometer.

After 60 days, the following were evaluated: percentage of survival (living minicuttings) rooting (minicuttings with roots  $\geq$  1 mm) and callus formation (with undifferentiated cell mass formation at the base), number and average root length (based on the three largest roots), number and average shoot length. The percentages of rooting and callus formation were calculated using the total number of surviving minicuttings.

Data were submitted to analysis of variance (ANOVA) and, when significant, the means of the treatments were statistically compared by Tukey's test at  $p \leq 0.05$ . For the statistical analysis, the data expressed as a percentage were transformed into  $\sqrt{x} / 100$ , to meet the assumptions of homogeneity of the variance and normality of the residues.

## RESULTS

For Chardonnay, means of survival and rooting varied between 93 and 100% for all treatments. All surviving minicuttings rooted regardless of the use of IBA or the concentration tested in the experiment. No statistically significant differences were observed between treatments in any of the variables analyzed, with all treatments being equally effective (Table 1).

Table 1. Root number (RN), average root length (ARL), shoot number (SN) and average shoot length (ASL) of minicuttings of Chardonnay grapevine and SO4 rootstock under different concentrations of IBA for rooting.

*Tabela 1. Número de raízes (NR), comprimento médio de raízes (CMR), número de brotações (NB), comprimento médio de brotações (CMB) e número de folhas (NF) de miniestacas de videira Chardonnay e porta-enxerto SO4 sob diferentes concentrações de AIB para o enraizamento.*

<b>Chardonnay</b>				
<b>IBA (mg L<sup>-1</sup>)</b>	<b>RN</b>	<b>ARL (cm)</b>	<b>SN</b>	<b>ASL (cm)</b>
0	8.33 <sup>ns</sup>	12.88 <sup>ns</sup>	0.90 <sup>ns</sup>	1.28 <sup>ns</sup>
500	8.28	14.55	1.13	1.30
1000	8.13	15.40	1.13	1.50
1500	7.73	15.55	1.18	1.50
<i>p</i> -value	0.9986	0.7019	0.1448	0.5950
CV (%)	22.63	20.12	17.04	18.22
<b>SO4</b>				
0	3.58 a	7.73 a	0.53 <sup>ns</sup>	0.50 <sup>ns</sup>
500	2.15 b	6.55 ab	0.45	0.50
1000	2.23 b	6.63 ab	0.65	0.68
1500	1.63 b	5.10 b	0.25	0.38
<i>p</i> -value	0.0065	0.0073	0.1694	0.0134
CV (%)	19.75	15.00	59.95	40.75

<sup>ns</sup> = not-significant by the F-test at  $p \leq 0.05$ . Means followed by different letters within columns are significantly different according to Tukey's test at  $p \leq 0.05$ .

Regarding SO4, the best results for percentage of survival (between 67 and 77%) and rooting (between 67 and 75%) were obtained using treatments of 0, 500 and 1000 mg L<sup>-1</sup> IBA. In both variables, there were no statistical differences between these treatments, however, showed better results than treatment receiving 1500 mg L<sup>-1</sup> of IBA, with a difference of at least 24% for survival and 34% for rooting (Fig. 1).

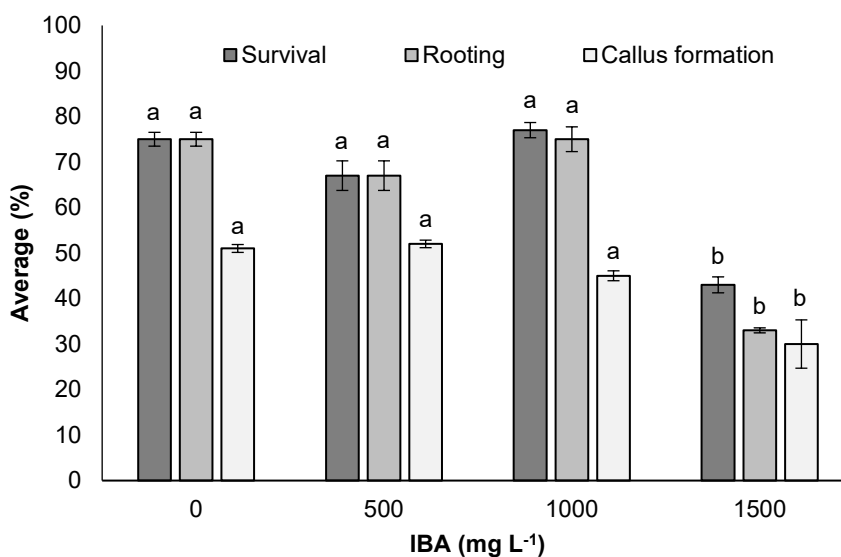


Figure 1. Percentages of survival, rooting and callus formation of minicuttings of the SO4 grapevine rootstock, under IBA treatments. Columns of the same color with different letters are significantly different according to Tukey's test ( $p \leq 0.05$ ); the bars represent the standard error of the mean.

*Figura 1. Porcentagens de sobrevivência, enraizamento e formação de calo de miniestacas do porta-enxerto de videira SO4, sob tratamentos de AIB. Colunas com a mesma cor com letras diferentes são significativamente diferentes de acordo com o teste de Tukey ( $p \leq 0.05$ ); as barras representam o erro padrão da média.*

The percentage of callus formation in the SO4 minicuttings ranged between 30 and 52%, with no statistical differences between the treatments 0, 500 and 1000 mg L<sup>-1</sup> IBA (Fig. 1). There was at least a 15% difference compared to the treatment with 1500 mg L<sup>-1</sup> of IBA. There was no callus formation in the minicuttings of the Chardonnay variety.

For the number and average root length in the SO4, the highest means were observed in treatments with no IBA (3.58 and 7.73 cm, respectively). However, for the average root length, no statistical differences were found in relation to treatments 500 and 1000 mg L<sup>-1</sup> (6.55 and 6.63 cm, respectively). Therefore, treatment 1500 mg L<sup>-1</sup> presented the lowest mean (5.10 cm), without showing significant differences with the two treatments mentioned previously (Table 1).

In relation to the number and average shoot length, no significant differences were observed among treatments. Also, the means obtained ranged from 0.25 to 0.65 for number of shoots and, from 0.38 to 0.68 cm for the average shoot length (Table 1).

## DISCUSSION

For the Chardonnay variety, it was possible to promote the rooting of minicuttings, even without the use of IBA. In contrast, for the SO4 rootstock, it's observed that the concentration of 1500 mg L<sup>-1</sup> of IBA appears to have been detrimental to the survival and rooting of the minicuttings. In this case, it is possible to observe that the genotype is one of the factors that make the physiological responses.

In the rooting of semi-woody cuttings of grapevine rootstocks, Biasi et al. (1997) did not observe any significant differences between the evaluated IBA concentrations (0 to 2000 mg L<sup>-1</sup>), obtaining rooting averages between 75 and 100%. The authors found that the increase in IBA concentration increased mortality in four rootstock varieties out of the five evaluated in the study. This show that the plant tissue of minicuttings is very sensitive to plant growth regulators application and auxins can have a phytotoxic effect. Hence, they claimed that the use of auxins is not necessary to stimulate the rooting of the rootstocks.

Botelho et al. (2005) obtained a mean of 92% of rooting in herbaceous cuttings of VR 43-43 grapevine rootstock, with no use of growth regulators. In the same study, the treatment 1000 mg L<sup>-1</sup> IBA increased the root number; however, when associated with PBZ (paclobutrazol), it reduced the percentage of rooting, which was attributed to a possible phytotoxic effect of these substances.

In another study using rooting of herbaceous cuttings of the VR 43-43 grapevine rootstock, Lone et al. (2010) verified that the highest percentages of survival and rooting were obtained by using regulators (66% and 41%, respectively). The authors also conclude that the application of IBA was not needed for the rooting of this rootstock.

On the other hand, when testing IBA concentrations and cutting types in Jales rootstock, Faria et al. (2007) found that the use of this regulator had a positive influence on the rooting of cuttings with leaves. Considering that in the cuttings not treated with IBA, rooting was of 69% and, in the cuttings treated with concentrations 1500 and 2000 mg L<sup>-1</sup>, the rooting was 97%.



The use of synthetic auxins may inhibit or stimulate tissue growth and differentiation, and physiological responses depend directly on endogenous auxin levels (BOTELHO et al., 2005). It can be seen that those responses can also vary according to the genotypes. In this experiment, although the material was collected under the same conditions and the characteristics of minicuttings are standardized, because they are different genotypes, they may differ in relation to the endogenous auxin levels. Nevertheless, the objective of this study was not to compare them, but to find the best treatment for each of them.

In relation callus formation, for some species, this may occur before the formation of roots; however, for others, the rooting occurs independently of this condition (SAMPAIO et al., 2010). Such condition was not related in this experiment as the rooted Chardonnay minicuttings and the presence of callus was not verified. In relation to SO<sub>4</sub>, callus was formed, which was a disadvantage since the excess of callus at the base of the minicuttings may have impaired root emission.

Another factor that can influence rooting is the presence and maintenance of leaves after minicutting. In the case of herbaceous minicuttings, which are in full vegetative growth, the leaves are a reserve of growth regulators, among other substances such as carbohydrates and nutrients, besides synthesizing cofactors that contribute to the formation of roots (ROBERTO et al., 2006; HARTMANN et al., 2011).

Roberto et al. (2006) evaluated types of cuttings with Campinas and Jales rootstocks, obtaining averages of 77% and 81% of rooting, respectively. According to the authors, the preparation of cuttings has great importance in the process of plants production. They emphasize the importance of maintaining leaves for the formation of adventitious roots over propagation of herbaceous cuttings. However, grapevine is a species with large leaves, which may hinder rooting due to excessive dehydration. Therefore, in this experiment, it was decided to use minicuttings with leaves with their limb reduced by half.

The use of minicuttings by maintaining one or two half leaves has already been studied and used for other species, such as guava (*Psidium guajava* L.) (ALTOÉ et al., 2011), blueberry (*Vaccinium* spp.) (NASCIMENTO et al., 2011; FISCHER et al., 2013), peach (*Prunus persica*) (TIMM et al., 2015), "ipê-roxo" (*Handroanthus heptaphyllus*



Mattos) (OLIVEIRA et al., 2015), yerba mate (*Ilex paraguariensis*) (SÁ et al., 2018) and grapevine rootstocks (ROBERTO et al., 2006). The reduction in leaf size reduces water loss, therefore, facilitating the handling of the material (FISCHER et al., 2013), and enabling the storage of a greater number of minicuttings in a particular container or space intended for rooting.

The season of year can also influence minicutting. Cuttings collected between spring and summer, when the branches are in full growth, tend to have a higher concentration of auxins in relation to those that are collected in autumn and winter. Consequently, these variations may influence rooting (DIAS et al., 2012). As clonal mini-gardens are formed in protected cultivation, there is a greater chance of controlling the productivity of stock plant (mother plant), allowing the propagation of material at different times in the year.

The application of IBA did not influence the rooting of Chardonnay minicuttings, in which all surviving minicuttings rooted, even without the use of this regulator. For SO4, the concentration 1500 mg L<sup>-1</sup> seems to have been detrimental to the survival and rooting of the minicuttings. Also, between 0 and 1000 mg L<sup>-1</sup> concentrations, no significant differences were found in the majority of the variables. As a result, it is verified that the use of growth regulator for the rooting of these materials is not needed.

## CONCLUSIONS

It is possible to root herbaceous Chardonnay grapevine minicuttings and SO4 grapevine rootstock without IBA application.

Minicutting is a viable technique for propagation and can be applied to the formation of clonal mini-gardens of these genotypes.

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