

## Rooting behavior and biochemical changes in relation to IBA concentrations in different grape rootstocks

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### ABSTRACT

Indole-3-butyric acid (IBA) treatments at different concentrations (750, 1000, 1500 and 2000 ppm) were given to the hardwood stem cuttings of six grape rootstocks before planting in rooting medium. The bud sprouting success of cuttings was not significantly influenced by the IBA treatments. However, the days taken for bud sprout were significantly reduced with 750 ppm concentration. Shoot length, root length, leaf area and number of root primordia exhibited varied response among rootstocks. Biochemical analysis showed significant changes in sugar and protein content of leaves. Higher protein content was recorded at 750 ppm IBA in 1613C, St. George and 140-Ru rootstocks and was superior over other concentrations. Total phenolics content increased significantly with increase in IBA concentration in St. George. Considering the days taken for bud sprout, sprouting success and root primordia, 2000 ppm IBA was better for Dog Ridge. It can be concluded that 1000 ppm IBA concentration was for rooting success and growth in 1613C and 140-Ru. In 41-B and Freedom, the sprouting success was higher at 1500 & 2000 ppm. The numbers of root primordia also varied significantly among the rootstocks. Maximum numbers of root primordia were recorded in 1613C and St. George rootstocks as compared to all other rootstocks indicating their distinct genotypic behavior.

**Key words:** Auxin, grape, rooting, rootstocks.

### INTRODUCTION

Grape rootstocks have become indispensable under adverse conditions like soil salinity and poor quality irrigation water. The rootstocks have potential to combat the soil problems and also act as a tool for manipulating the vine growth and productivity. This helps in altering the shoot vigour, thus bringing equilibrium between the growth and yield. With the increase in area under grape cultivation on rootstock, it became necessary to fulfill the demand for rootstock plants. The grape rootstocks are generally propagated through hardwood cuttings, though propagation by seed, soft wood cuttings and layering is specific to certain situations. The use of rooting hormones for propagation in fruit through hardwood stem cuttings has been studied widely by many workers. Indole-3-butyric acid (IBA) is widely used auxin to stimulate rooting due to its ability to promote root initiation, low toxicity and good stability in comparison with naphthalene acetic acid and indole-3-acetic acid (Blazich, 2; Singh *et al.*, 10; Wiesman and Lavee, 15). Among the different grape rootstocks, Dog Ridge (*Vitis champini*) is one of the most popular rootstock with the grape growers in India. There are different rootstocks, which vary in their capacity to root in response to various treatments. Basically, it involves water treatment and leaching out the rooting inhibitors

in the cuttings (Satisha and Adsule, 8). Hence, an experiment was conducted to study the different IBA concentrations required to get maximum rooting and also to study its effect on biochemical changes induced during the rooting process in different grape rootstocks.

### MATERIALS AND METHODS

The study was conducted at the Nursery of NRCG, Pune during the year 2009-10. Six rootstocks (Dog Ridge, 1613 C, St. George, 140-Ru, 41-B and Freedom) representing different wild *Vitis* species were selected. From the 6-8 year-old mother vines of rootstocks, well matured one season old hardwood stem cuttings having 4-5 nodes were prepared. These cuttings were then tied in bundles and soaked in tap water for about 24 h to leach out the rooting inhibitors. Thereafter, a fresh slanting cut was given at the basal portion of the cuttings and then dipped in different concentrations of IBA (2000, 1500, 1000 and 750 ppm) for about 30 sec. and compared with the control where only distilled water was used. Cuttings were planted in black polythene bags of 5" × 7" size (1 l capacity) filled with potting mixture of soil, sand and compost (2:1:1). The poly bags were watered thoroughly and all the cultural operations like weeding and plant protection were carried out at different growth stages.

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The observations on days taken for bud sprouting were recorded every day morning and the percent success was recorded at 75<sup>th</sup> day after planting. The growth parameters like shoot length, root length and number of root primordia were recorded at 120 days after planting by following the standard methods. The total carbohydrates content of was estimated by the anthrone method as suggested by Sadashivam and Manickam (7). The total phenolic content was determined using the Folin-Ciocalteu reagent (Singleton and Rossi, 11). The concentration of total phenolics was expressed as the catechol equivalent (mg/g) of the lyophilized sample. Protein content was estimated by using Lowry *et al.* (4) method. The data were presented as an average of three determinants collected for all the different characters studied. The treatment significance was tested by one way ANOVA (Duncan's multiple range test) using the SPSS version 11.0 and p-value at 0.05 (Verma, 14).

**RESULTS AND DISCUSSION**

Significant differences were recorded for number of days taken to bud sprout, sprouting success and numbers of root primordia among the rootstocks

(Table 1). The rootstock 1613 C was the earliest to sprout (8.03 days) at 1000 ppm IBA treatment. It was followed by Dog Ridge (8.07 days), St. George (8.46 days), 41-B (8.63 days), Freedom (8.74 days) and 140-Ru (9.41 days) as compared to the control. Other than Dog Ridge rootstock, 1000 ppm IBA treatment was found to be appropriate for reducing the days taken to bud sprout. The bud sprouting success ranged from 53.00 under control to 94.40% in 2000 ppm IBA treatment. The numbers of root primordia were also significantly differed among the rootstocks and the IBA concentrations. Maximum number of root primordia were recorded in 1613 C and St. George as compared to other rootstocks. In 1613-C, the numbers of root primordia were higher at 1000 ppm concentration (20.78), whereas in St. George it was higher in 750 ppm (19.16). In 140-Ru rootstock, the higher number of root primordia were recorded at 2000 ppm IBA (9.06). The rootstock 41-B imparted more root primordia (14.53) at 1000ppm as against 4.10 in control. Highest number of root primordia (15.83) was also observed in Freedom rootstock at 1500 ppm compared to lowest in control (8.89). This clearly indicates the difference

**Table 1.** Effect different IBA concentration treatments on number of days required for sprouting, sprouting success and number of root primordia in various grape rootstocks.

IBA (ppm)	Dog Ridge	1613 C	St. George	140 RU	41 B	Freedom
<b>a. Days taken for bud sprout</b>						
0 (control)	11.29 ± 0.79 a	10.43 ± 0.49 a	13.04 ± 1.06 a	11.70 ± 0.44 b	10.23 ± 0.72 a	11.48 ± 0.68 a
750	10.18 ± 0.37 b	10.32 ± 0.85 a	12.63 ± 0.73 a	9.87 ± 0.79 c	11.04 ± 0.40 a	10.46 ± 0.49 b
1000	8.07 ± 0.66 c	8.03 ± 0.56 c	8.47 ± 0.40 c	9.41 ± 0.45 c	8.89 ± 0.72 b	8.74 ± 0.71 c
1500	10.28 ± 0.61 b	8.68 ± 0.32 bc	11.14 ± 0.88 b	11.33 ± 0.80 b	8.63 ± 0.51 b	10.80 ± 0.76 ab
2000	9.92 ± 0.48 b	9.44 ± 0.56 b	12.68 ± 0.89 a	13.47 ± 0.91 a	8.91 ± 0.43 b	11.68 ± 0.43 a
<b>b. Sprouting success (%)</b>						
0 (control)	53.00 ± 7.44 c	71.00 ± 6.43 b	64.00 ± 5.80 c	57.00 ± 5.16 c	58.80 ± 6.49 c	58.00 ± 5.25 c
750	67.00 ± 8.74 b	88.42 ± 8.75 a	70.00 ± 4.95 bc	81.25 ± 7.36 b	63.63 ± 3.87 c	85.00 ± 6.01 b
1000	87.00 ± 7.61 a	90.22 ± 7.61 a	70.00 ± 9.82 bc	87.50 ± 9.41 ab	72.72 ± 5.14 b	90.59 ± 6.12 ab
1500	91.94 ± 5.34 a	93.96 ± 4.36 a	80.00 ± 9.43 ab	93.69 ± 5.61 a	95.20 ± 4.61 a	97.20 ± 3.90 a
2000	94.40 ± 4.70 a	96.00 ± 3.07 a	85.00 ± 9.78 a	93.13 ± 6.72 a	96.20 ± 3.90 a	94.40 ± 4.70 ab
<b>c. No. of root primordia</b>						
0 (control)	9.06 ± 0.63 b	19.88 ± 0.95 a	14.56 ± 1.05 c	6.30 ± 0.44 b	4.10 ± 0.24 e	8.89 ± 0.52 c
750	6.75 ± 0.24 c	19.77 ± 1.39 a	19.16 ± 0.70 a	6.40 ± 0.25 b	5.71 ± 0.27 d	12.29 ± 0.58 b
1000	3.82 ± 0.32 e	20.78 ± 1.46 a	14.93 ± 1.18 c	6.08 ± 0.50 bc	14.53 ± 1.18 a	9.90 ± 0.81 c
1500	6.01 ± 0.35 d	20.68 ± 0.77 a	16.71 ± 0.99 b	5.60 ± 0.33 c	8.08 ± 0.57 b	15.83 ± 1.11 a
2000	9.97 ± 0.48 a	20.01 ± 1.63 a	14.58 ± 0.70 c	9.06 ± 0.43 a	6.84 ± 0.25 c	12.16 ± 0.45 b

Data are the means of three replicates standard deviation. <sup>a</sup>Values followed by different letters in columns differ significantly by DMRT at p = 0.05.

in behavior of different rootstocks to different rooting parameters. The early bud sprout, sprouting success and more numbers of root primordia in some of the rootstocks might be due to the availability of more carbohydrates in the mother vine, which might have broken down into sugars and acted as source of energy required for the sprouting of buds. In addition to the environmental factors, genetic characters, the exogenous supply of hormones and pre-planting treatments, the sprouting and rooting of cuttings depends on the features of endogenous factors (Dvin *et al.*, 3; Sivozaki *et al.*, 9). Although, the rooting process is a combined activity of number of biochemical constituents, several workers have shown that the carbohydrate availability is one of the factors that causes early rooting and bud sprouting (Bartolini *et al.*, 1; Veierskow, 13). The response of different IBA concentrations for days taken to bud sprout indicates that different rootstocks requires specific concentration of IBA. In Dog Ridge rootstock, the IBA concentration in the range of 1500

to 2000 ppm was found to be ideal. Similar results were also obtained in our earlier studies (Somkuwar *et al.*, 12).

In Dog Ridge rootstock, higher leaf carbohydrate content (180.87 mg/g FW) in leaf was recorded in 1000 ppm IBA concentration as compared to minimum in control (71.78 mg/g FW) (Fig. 1). Higher amount of fresh leaf protein (36.31 mg/g) and total phenolics (1.36 mg/g) were recorded at the same concentration. In 1613-C and St. George rootstocks, the carbohydrate content among the different treatments was at par. In 1613-C, higher protein content (43.10 mg/g) and total phenolics (0.95 mg/g) were recorded at 750 ppm IBA as compared to the control. In 140-Ru rootstock, higher amount of leaf carbohydrate (130.0 mg/g) was estimated as compared to minimum in control (58.0 mg/g). The similar trend was also recorded for protein content in these rootstocks. However, the higher total phenol content (2.08 mg/g) in the same rootstock was recorded at 1500 ppm IBA treatment. In 41-B, higher amount of carbohydrate (156.0 mg/g) and 1.92

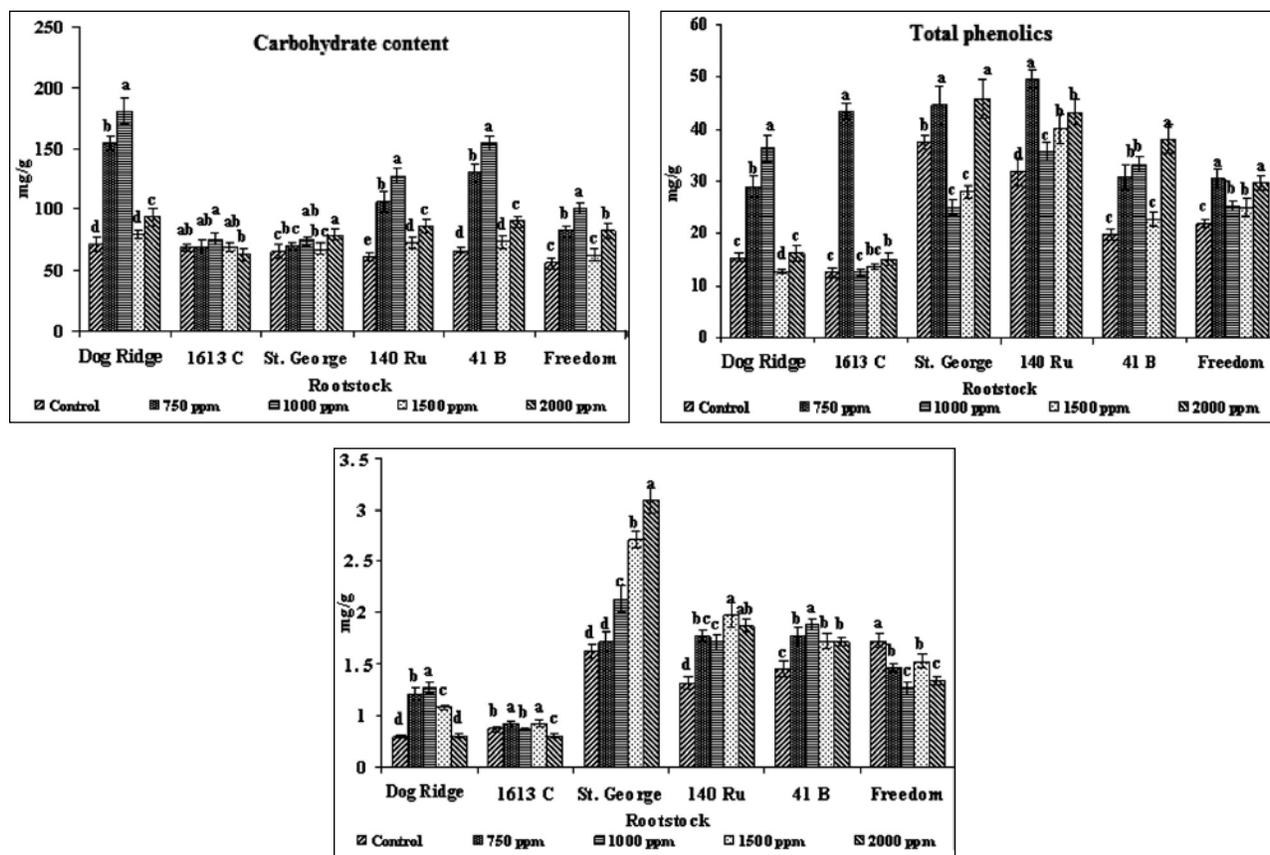


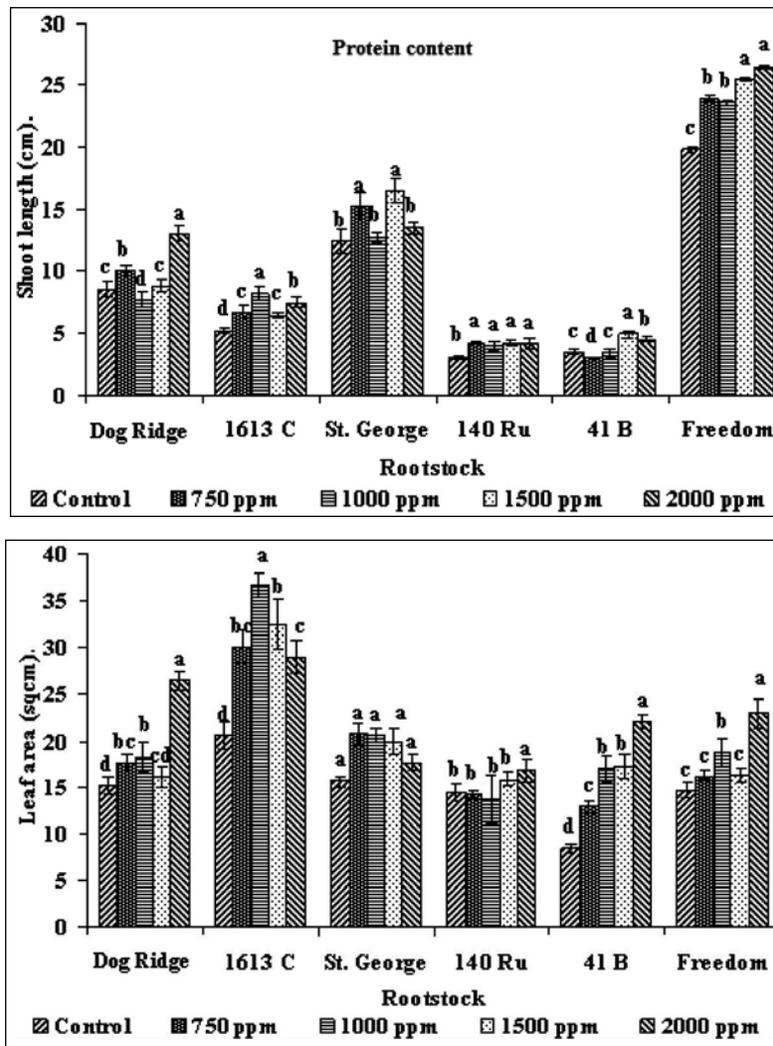
Fig. 1. Effect of different IBA concentration treatments on total carbohydrates, phenolics and protein contents of leaves after 120 days of planting in various rootstocks.

#Data columns represent means of three replicates with standard deviation as error bars. Different letters as data labels indicate significant difference between the means by DMRT at  $p < 0.05$ .

mg/g of total phenolics were recorded at 1500 ppm concentration. However, higher protein content of 39.50 mg/g was recorded at 2000 ppm compared to lowest in control (20.0 mg/g). Among the different IBA treatments, higher carbohydrate content was recorded at 1000 ppm compared to control. However, total protein was more at 750 ppm. The total phenol content in this rootstock was less under high rate of percent success. The quantity increasing with increase in IBA concentration in this rootstock may explain that 1000 ppm IBA concentration is ideal to obtain better rooting in 140-Ru rootstock. The variation in response among rootstocks for different IBA concentration might also be due to the genotypic variation among the rootstocks representing different species. From the study it was

observed that the higher indigenous carbohydrate was associated with the higher rooting. However, the role of phenols accumulated in cuttings during rooting seems to be also important (Shivozaki *et al.*, 9). Among the rootstocks, 140-Ru and 41-B had very poor shoot growth. Application of IBA externally to the cuttings may help to induce rooting and changing the enzymatic activities affecting phenolic levels allowing favorable endogenous hormone balance (Heloir *et al.*, 5). Ribnicky *et al.* (6) also reported that the applied auxins induce modification in their own metabolism, mostly by conjugation and in other hormones such as cytokinins.

The shoot length, root length and leaf area recorded 120 days after planting (Fig. 2), revealed



**Fig. 2.** Effect of different IBA concentrations on shoot length, root length and leaf area after 120 day of planting in various grape rootstocks.

\*Data columns represent means of three replicates with standard deviation as error bars. Different letters as data labels indicate significant difference between the means by DMRT at  $p < 0.05$ .

significant differences among rootstocks and the IBA treatments. Higher shoot length was recorded in Dog Ridge, Freedom and St. George rootstocks at 2000 ppm IBA. Maximum shoot length in Freedom was recorded in 1500-2000 ppm IBA. Among the different IBA treatments, higher root length was recorded in the early sprouting treatments. This might be due to the early appearance of root primordial along with the bud sprouts in different rootstocks. As the shoot growth increased during the growth period, the root growth also increased. Among all the rootstocks, Dog Ridge recorded higher root length at all the IBA concentration treatments and was followed by Freedom. This is possible while shoot growth is continuous; the root length remains same. Leaf area registered significant differences within rootstocks and the IBA concentration treatments. Among the different rootstocks, 1613-C exhibited higher leaf area at all IBA treatments and was followed by St. George and Freedom. Among the different IBA concentration treatments, the differences were significant for shoot length, which is generally observed under field conditions. The variation in shoot growth among the rootstocks was also evident. Under the control treatment, the rooting inhibitors might have not been leached out resulting in reduced rooting.

In conclusion, the results confirmed the importance of exogenous IBA applications and its concentration for rooting in grape rootstocks. The required concentration of IBA for rooting was rootstock specific.

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Received : June, 2014; Revised : February, 2015;  
Accepted : April, 2015