

**STUDIES ON THE EFFECT OF IBA AND P-HYDROXY BENZOIC ACID
ON THE ROOTING OF HARDWOOD AND SEMI-HARDWOOD
CUTTINGS OF PERLETTE GRAPE (Vitis vinifera L.)**

By

MUKESH KUMAR CHAWLA

**Thesis submitted to the Haryana Agricultural University in
partial fulfilment of the requirements for the degree of**

MASTER OF SCIENCE

in

HORTICULTURE

Haryana Agricultural University

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This is to certify that this thesis entitled " Studies on the effect of IBA and p-hydroxy benzoic acid on the rooting of hardwood and semi-hardwood cuttings of Perlette grape" submitted for the degree of M.Sc. in the subject of Horticulture of the Haryana Agricultural University, is a bonafide research work carried out by Sh. Mukesh Kumar Chawla under my supervision and that no part of this thesis has been submitted for any other degree.

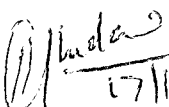
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Ranghust
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Major Advisor

CERTIFICATE-II

This is to certify that this thesis entitled "Studies on the effect of IBA and p-hydroxy benzoic acid on the rooting of hardwood and semi-hardwood cuttings of Perlette grape" submitted by Sh. Mukesh Kumar Chawla to the Haryana Agricultural University in partial fulfilment of the requirements for the degree of M.Sc. in the subject of Horticulture, has been approved by the Student's Advisory Committee after an oral examination of the same, in collaboration with an External Examiner.


17/12/91
Major Advisor


17/12/91
External Examiner


Head of the Department
20/12


23/12
Dean, Postgraduate Studies

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CONTENTS

<u>Chapter</u>		<u>Pages</u>
I	INTRODUCTION	1- 3
II	REVIEW OF LITERATURE	4-16
III	MATERIAL AND METHOD	17-21
IV	EXPERIMENTAL RESULTS	22-56
V	DISCUSSION	57-65
VI	SUMMARY	66-68
	LITERATURE CITED	i - vii
	APPENDIX	

LIST OF TABLES

<u>Table</u>		<u>Page</u>
1-	Initial observations before callusing in hardwood and semi-hardwood cuttings of Perlette grape.	23
2-	Effect of IBA and p-hydroxy benzoic acid treatments on diameter of hardwood cuttings after callusing collected during middle and end of January.	25
3-	Effect of IBA and p-hydroxy benzoic acid treatments on average number of root initials in hardwood cuttings collected during middle and end of January.	27
4-	Effect of IBA and p-hydroxy benzoic acid treatments on status of nitrogen after callusing in hardwood cuttings collected during middle and end of January.	28
5-	Effect of IBA and p-hydroxy benzoic acid treatments on status of carbohydrates in terms of total sugars at the base of hardwood cuttings after callusing collected during middle and end of January.	30
6-	Effect of IBA and p-hydroxy benzoic acid treatments on status of phenols of cuttings after callusing collected during the middle and end of January.	32
7-	Effect of IBA and p-hydroxy benzoic acid treatments on average number of sprouted buds after 35 days of planting of hardwood cuttings collected during the middle and end of January.	35
8-	Effect of IBA and p-hydroxy benzoic acid treatments on average number of leaves after 35 days of planting of hardwood cuttings collected during middle and end of January.	37
9-	Effect of IBA and p-hydroxy benzoic acid treatments on vigour of shoot after 60 days of planting of hardwood cuttings of grape collected during middle and end of January	40
10-	Effect of IBA and p-hydroxy benzoic acid treatments on vigour of shoot after 120 days of planting of hardwood cuttings collected during middle and end of January.	42

TablePage

11-	Effect of IBA and p-hydroxy benzoic acid treatments on average number of roots in hardwood cuttings collected during middle and end of January.	45
12-	Effect of IBA and p-hydroxy benzoic acid treatments on length of largest root in hardwood cuttings collected during middle and end of January.	47
13-	Effect of IBA and p-hydroxy benzoic acid treatments on fresh and dry weight of roots in hardwood cuttings collected during middle and end of January.	49
14-	Effect of IBA and p-hydroxy benzoic acid treatments on changes in constituents of grape cuttings.	51
15-	Effect of IBA and p-hydroxy benzoic acid treatments on growth of semi-hardwood cuttings after 42 days of planting collected in July.	53
16-	Effect of IBA and p-hydroxy benzoic acid treatments on root growth of semi-hardwood cuttings collected in July	56

LIST OF FIGURES

- Fig. 1 Per cent sprouting as effected by time of taking hardwood cuttings.
- Fig. 2 Per cent sprouting as effected by different treatments in January
- Fig. 3 Per cent mortality as effected by different treatments and time of taking hardwood cuttings.
- Fig. 4 Per cent sprouting in semi-hardwood cuttings collected and planted in July.
- Fig. 5 Per cent sprouting as influenced by different treatments in July season.
- Fig. 6 Per cent mortality as influenced by different treatments in July.

CHAPTER-I

INTRODUCTION

Grape is believed to be originated from a region lying between the Black and Caspian Sea. It is grown almost all over the world in the agroecological regions ranging from temperate to tropical climatic conditions. It belongs to family vitaceae. Several varieties are being cultivated for different purposes so as to form an industry of importance. India ranks first in world with regard to average yield of grapes that is 21.5 tonnes/ha (Pandey and Pandey, 1990). Grape cultivation is on increase in parts of U.P. Haryana, Punjab, Rajasthan and other regions of country. In Haryana, grapes cover a considerable area of 5000-6000 acres with a production of 50-60 thousand tonnes (Dhawan and Gupta, 1990). At present grape vines are commercially propagated by cuttings. However, budding and grafting are other methods of propagation but these are performed under certain conditions. Budding is a laborious and time consuming method as raising of such plants take about two years. Thus, the cost of production of such plants is quite high. Since, availability of large number of plants at low cost would be one of the pre-requisite for success of grape orcharding, to achieve this objective, raising plants from cuttings is of paramount importance. So, there is a need to study its propagation by cutting and thus to improve rooting of cuttings. Singh et al. (1971) reported that IBA 500 ppm improved

rooting of cuttings of Perlette vine under Ludhiana conditions while a concentration of 5000 ppm IBA improved the overall rooting of cuttings of early pruned vine of Salt Creek and Dogridge grape (Alley, 1979). Certain phenols are also reported to promote rooting of cutting in other fruit crops. Sadhu et al. (1972) found root promoting effect at p-hydroxy benzoic acid in a number of plants. An effort made by Rakesh (1987) under Haryana conditions resulted in highest rooting percentage and number of roots per cutting when pretreated with IBA in combination with p-hydroxy benzoic acid in lemon. Bose (1972) reported synergism between auxin and phenolic compound in the rooting of cuttings of ornamental shrubs. However, Bartolini (1987) reported that some studies related to rooting of cuttings showed that alone phenolic treatment is better than its combination with exogenously applied auxin. Kling and Meyer (1983) reported that p-hydroxy benzoic acid is negatively interacting with auxins in cuttings of Phaseolus aureus and Acer griesum. So, the role of phenolic compound as rooting co-factor is still controversial.

Saraswat (1973) reported that hard wood cuttings of grape planted during the first week of January resulted better than later planting. Moti and Singh (1968) reported that semi-hard wood cuttings of Bhokri and Gulabi grape taken during July gave 34 per cent rooting.

Literature is lacking with respect to effects of combined application of auxin and phenolic compound in case of grape. So there was need to study the rooting behaviour of hardwood and semi-hard wood cuttings when

treated with indole-phenol complex. Present studies were undertaken with the following objectives :

- 1- To find the optimum time of taking hard wood cuttings.
- 2- To study the rooting success of semi-hard wood cuttings in grapes during July.
- 3- To find the optimum concentration of IBA and p-hydroxy benzoic acid for rooting of cutting.

CHAPTER-II

REVIEW OF LITERATURE

Stem cutting is the cheapest way of raising grape orchard and thus improvement in rooting of cuttings is of paramount importance. But it is influenced by several factors like selection of cuttings, shoot maturity, hormonal changes, callus formation, chemical treatments and environmental factors. The literature related to these factors is reviewed in this chapter.

A. SELECTION OF CUTTINGS

i- Time of taking hardwood cuttings

Hartman and Hansen (1958) reported that fall planted or spring planted cuttings of Mariana 2624 plum rooted equally if treated with IBA; without such treatments, however, only fall collected and planted cuttings rooted appreciably. Moti and Singh (1968) reported that hardwood cuttings of Bhokri and Gulabi grapes taken during January and rooting percentage amounted to 75 and 80 per cent respectively and early January was also reported as better time than July. Sarswat (1973) reported that hardwood cuttings of vines cv. Bhokri and Selection-7 gave the best results when planted during first week of January rather than later. Weaver et al. (1975) under California conditions reported that early collected cuttings of Carignane grape broke sooner than late collected cuttings. But as reported by Basso and Natali (1975) under Arboree conditions (USA) rootstocks cv. Kober 5 BB and SO-4 showed very little effect on the rooting as influenced by time of taking

cuttings (28 October to 22 March). In 140 Ruggeri, however, rooting was reduced in cuttings taken between December and February. Goode et al. (1982) reported that dormant cuttings of muscadina grapevine cvs. Hund and Cowart, rooted best early in the dormant season. Although root quality was poor and percentage rooting was rather low.

ii- Portion of cuttings

Hartman and Hansen (1958) reported that in both plum and quince cuttings taken from basal portion of the shoot tended to root more readily than cuttings made of the terminal sections. This may have been due to somewhat higher food reserves as total sugars, or it could have been due to an accumulation in shoot bases of natural auxins or other root promoting factors. Weaver et. al. (1975) reported that grapevine cuttings collected from apical, middle and basal portions of canes and middle cuttings resulted better than apical portion cuttings. Similar results were obtained by Munoz and Villalobos (1977) in easy to root Vitis vinifera cv. Sultanina and difficult to root Vitis champini cv. Dogridge. Daulta et. al.(1982) reported highest rooting percentage in basal (62-92%) and lowest (48-76%) in apical cuttings of grapes cv. Perlette, Thompson Seedless, Delight and Beauty Seedless.

iii- Diameter and length of cuttings

Goode et al. (1982) reported better rooting in cuttings of large diameter than in case of small diameter cuttings. Singh et al. (1986) reported that thick, thin, short and long internode cuttings of Perlette grape were or were not treated with IBA at 5000 ppm concentration. Untreated thick, short and long internode cuttings gave best sprouting

(66.7 - 73.3%) and rooting (53.3 - 73.3%), the greatest diameter of thickest shoots (7.2 to 7.4 mm). IBA treatments gave poor results in respect of all the indices studied.

B. SHOOT MATURITY AND HORMONAL CHANGES

Munoz and Villalobos (1977) carried out studies with easy to root V. vinifera cv. Sultanina and difficult to root V. champini cv. Dogridge and the results showed that the presence of growth stimulating substances was independent of shoot maturity in Sultanina whereas in Dogridge the presence of growth stimulating substances was dependent on shoot maturity. Studies carried by Nokano et al. (1980) on the rooting of hardwood cuttings of the grapevine cv. Delaware revealed that some GA₃ like activity was detected in bursting buds of intact Delaware vines but very little in basal portion of hardwood cuttings. Cytokinin activity was observed in buds but the chromatographic patterns of active cytokinins in the buds of intact vines differed from those in cuttings. It was suggested that (i) they might indirectly affect rooting by promoting bud break (ii) Auxin activity was lower in well rooted cuttings than in unrooted cuttings (iii) Auxin activity in intact vine decreased from autumn to winter and increased again in spring (iv) ABA activity was high in the vine in autumn and decreased sharply after leaf fall.

Kracke et al. (1981) studied hormonal changes during rooting of hardwood cuttings of grapevine root stocks (easy to root) like Kober 5 BB and explained that root stock contains a very high amount of auxins but very low level of acid GA like and ABA like substances where root stock (hard to root) like 140 Ruggeri contains low level of auxin and high amounts

of GA and ABA like substances, increased slightly in both root stocks until bud burst when they decreased to a low level at root emergence. At the same time GA like compounds increased rapidly in 140 Ruggeri but decreased in Kober 5 BB, ABA like substances in 140 Ruggeri reached a high level, whereas in Kober 5 BB they remained low. Krack et al. (1983) revealed that cuttings of cv. Kober 5 BB (easy to root) and 140 Ruggeri (hard to root), treated with 4000 ppm IBA or 2000 ppm NAA induced marked modifications in the hormonal pattern at different stages of root formation in 140 Ruggeri and to a lesser extent in Kober 5 BB. Auxins doubled the IAA content and induced two peaks during cell multiplication at the cutting base and immediately before root emergence. In general, IBA and NAA promoted a GA degradation so that after callusing the GA level was about half of that in control. NAA induced strong depletion of ABA in Kober 5 BB and to a lesser extent in 140 Ruggeri. Both treatments prevented a rise in ABA content before root development.

C. CALLUS FORMATION

Hartman and Hansen (1958) reported that Mariana 2624 plum rooted best when cuttings were taken in mid November, treated with IBA, stored for six weeks at 60°F until planting in spring whereas in peach hardwood cuttings gave high percentage of root initiation in fall collected cuttings, treated with IBA, then held under moist storage conditions for 6 weeks at 60°F but such cuttings failed to grow when planted. Rooting success was better when cuttings were collected and planted in mid- November and treated with IBA. Mathur (1965) reported that the cuttings of Anab-e-shahi do not form callus and 72 per cent of cuttings of Selection-94 callused after

about 10 days of storage in moist sawdust. The percentage of success in uncallused Anab-e-shahi and callused Selection-94 was 65 and 62 per cent respectively. This indicates that callus formation does not play an important role in rooting of cuttings. The cuttings of Selection-94, which formed callus before planting, rooted better irrespective of fact that whether the callus was scrapped. Singh and Bakshi (1971) reported that the callusing treatment alone significantly reduced subsequent rooting and average number of roots and also tended to impair shoot development. Treatments of both types of cuttings (callused and uncallused) with IBA at 500 ppm improved these characters but higher rates had progressively harmful effects. Callus formation in Perlette cuttings was poor both before and after planting, most of roots emerging around the basal node. Singh and Singh (1973) reported that the rooting performance of hardwood cuttings of Thompson Seedless and Himrod grapevines was not improved by storing the cuttings in wet sand for a month before planting. No callus formation was observed in either stored or directly planted cuttings. Alley et al. (1974) reported that cuttings of Thompson Seedless planted fresh appeared to root as well as cuttings stored in sand bed and planted in March. Goode et al. (1982) found no significant rooting response with precallusing of grape cuttings while a positive effect of callusing in Vitis vinifera and Vitis berlandieri was observed by Hubackova (1987).

D. BIOCHEMICAL CHANGES DURING ROOTING

Mokashi (1978) reported that cuttings of Thompson Seedless (difficult to root) had lower contents of sugars, phenolic compounds and C:N ratio but higher nitrogen and starch contents, than in the cuttings of Gulabi (easy

to root) when soaked in IBA for 24 hours. Hosoi et al. (1972) reported that nitrogen level was reduced through out the propagational period of hardwood cuttings of grapevine, carbohydrates increased after 40 days, whereas Zarkuá (1974) reported a direct correlation between nitrogen and root regeneration of grapevine cuttings. Iannini et al. (1976) reported that levels of sugars and phenolic compound increased with rooting response of woody scion of grape cv. Merlot with a single bud. Kaundal and Singh (1987) reported that hardwood cuttings of pear under Ludhiana conditions were kept in moist sand for 30 days for callus formation and after that biochemical constituents were estimated at the base of cuttings. Cuttings were treated with 0, 50, 100 and 200 ppm IBA (24 hours soaking) before callus preparation. IBA at 100 ppm gave the maximum rooting of 41.16 per cent and this was correlated with highest contents of total sugars (20.30 mg/g); C/N ratio (33.5) and total phenols (2.28 mg/g). The untreated control cuttings with the lowest rooting success of 20.12 per cent were correlated with maximum starch content (24.20 mg/g), lowest total sugars (10.60 mg/g), total nitrogen (1.48%). C/N ratio (23.5) and total phenols (1.50 mg/g) after 30 days of storage in sand. It is indicative that changes in C/N ratio had a direct bearing with the rooting intensity. It has been reported earlier that high C/N ratio favoured the rhizogenesis (Hyun, 1967; Singh, 1976). Growth regulator treatments lowered the starch, total nitrogen and increased the total sugars, C/N ratio and total phenols. However, a casual relationship between the decreasing level of starch and total nitrogen with the increasing intensity of rhizogenesis could not be established. Similar observations were reported by Kaundal and Bindra (1984) in almond peach hybrid. Earlier studies by Basu et al. (1972) in

mango cuttings revealed that during rhizogenesis decreasing level of nitrogen is indicative that it is being utilized during the process of rhizogenesis.

E. CHEMICAL TREATMENTS

i- IBA (Indole-3-butyric acid)

Mostafa and Hudson (1967) found highly active root promoting material in extracts of basal segments of IBA treated 'Old Home' pear cuttings bases of the cuttings were dipped for 5 seconds in 2000 ppm IBA in 50 per cent ethanol then were placed horizontally in moist peat moss. Maximum rooting of 83.3 per cent was induced by treated cuttings. The untreated cuttings under control gave only 50 per cent rooting. Singh et al. (1971) reported that IBA treatment at 500 ppm before callusing of cuttings by quick dip method improved average number of roots and shoot development but higher rates had progressively harmful effects when applied to Perlette grape cuttings. Peterson (1973) reported that Dogridge grape cuttings dipped quickly in 0.2 per cent IBA rooted well in a sand bed. Alley (1979) reported that IBA before callusing at 5000 ppm concentration by quick dip method improved the overall rooting and number of rooted cuttings of early pruned vines cv. Salt Creek and Dogridge. It improved the number of rooted cuttings of late pruned Dogridge and size of all rooted Salt Creek cuttings. With easy to root cv., IBA improved the number of rooted Ganzin I and Zinfandel cuttings. Williams and Anteliff (1984) reported that 2000 ppm IBA treatment to the base of cutting before callusing is the optimum concentration of rooting of cutting of grape. Kaundal and Singh (1987) reported that IBA treatments by quick dip method at 2000 ppm produced the maximum rooting success of 36.06 per cent which was significantly better than 500 and 1000 ppm concentration in

pear cuttings. Hermann (1968) revealed that 5000 ppm solution with quick dip method gave same rooting success in pear cuttings. In later studies, Sandhu and Singh (1982) found that very little rooting was obtained in different pear cvs. with 500 and 1000 ppm IBA applications. This variation in dose response may be attributed to the difference in type, genetic make up, time of preparation and physiological status of cuttings.

Mokashi (1978) obtained best results by soaking the cuttings of Gulabi cv. of grape in IBA at 250 ppm for 12 hours before planting. Dorokhov (1983) found that cultivar of grape with moderate rooting capacity for rooting required 0.005 per cent IBA for 12 hours for good rooting (80-85%).

Herath (1978) reported highest percentage of rooting of cuttings of grape cv. improved Isabela treated in 50 ppm IBA for 24 hours. Sherer et al. (1985) evaluated root inducing capacity of six growth regulators and best root formation of grape root stock was induced by IBA (0.005-0.01%) before planting. Kaundal and Singh (1987) reported that cuttings of pear with IBA for 24 hours soaking at 100 ppm concentration gave the maximum rooting success of 41.16 per cent and it was recorded to be minimum 20.12 per cent in untreated cuttings. In earlier studies Nanda (1970) obtained 20.00 per cent rooting success whereas Singh (1976) noted 37.00 per cent success in sand pear cuttings with 100 ppm IBA application.

ii- Phenolic compounds

Jones et al. (1976) found that caffeic acid, catechol and pyragallol did not increase rooting of cuttings of apple significantly in comparison to control whereas Sherer et al. (1985) evaluated root inducing capacity of six growth regulators and best root formation of grape root stock was induced by

p-aminobenzoic acid .Gad et al. (1987) reported that rooting of Pelargonium graveolens, P. pettatum and P. zonalo cuttings was promoted by dipping cuttings for 2 hours in 4-chlororesorcinol, a polyphenol oxidase inhibitor, at 2 mm in P. pettatum cuttings, the treatments inhibited flowering during rooting period. Treated cuttings developed more numerous and larger leaves and produced an overall more fresh and dry weight than untreated cuttings. The enhanced vegetative growth was attributed to the better developed root system.

iii- **Auxin-phenol synergism**

Stonier et al. (1970) studied inactivation of certain protectors by polyphenol oxidase in Japanese Morning Glory. The oxidation of catechol to o-quinone is accompanied by a loss of chromophores that absorb ultraviolet light and appearance of reddish brown colour. The data indicated that auxin protector contain o-dihydroxy phenolic groups at their active site that inhibit IAA oxidation as described by some authors. It is suggested that high molecular weight auxin protector and phenolic compounds described by other authors comprise part of a metabolic system concerned with regulation of peroxidase catalysed redox reaction. Basu (1970) obtained synergism of auxin-phenol in rooting of cuttings of ornamental plants irrespective of auxin sparing effect of non-auxinic chemicals (mono or poly phenols) and placement of OH group of non-auxinic chemicals. The level of synergistic effects were found to be dependent on the concentration of the interacting auxins and non-auxinic chemicals. Bose (1972) reported synergism between auxin and phenolic compounds, in the rooting of cuttings of ornamental shrubs. Sadhu et al. (1973) found auxin synergists in the rooting of water apple cuttings.

Debnath (1986) obtained maximum rooting (95%) in the cuttings of lemon treated with ferulic acid at 2000 ppm and then treated with NAA at 5000 ppm.

iv- Auxin-phenol antagonism

Possible role of phenolic compounds in growth and rhizogenesis of cuttings was studied by Turetskaya et al. (1987) and revealed that some phenolic compounds behave as co-factor of IAA, while p-coumaric acid acts as inhibitor of IAA activity. It does not possess protector properties that is unlike other phenolic compounds, it is not IAA protector that does not depress IAA-oxidase activity. At the same time, it is capable of inhibiting synthesis of IAA from L-tryptophan. Zenk and Muller (1963) reported that in vivo destruction of exogenously applied IAA as influenced by naturally occurring phenolic acid. Basu (1970) obtained antagonism irrespective of auxin sparing effect of non-auxinic chemicals. The level of antagonistic effects were found to be dependent on the concentration of the interacting auxins and non-auxinic chemicals. It has been observed that antagonism between auxins and non-auxinic chemicals was always accompanied by increased upward transport of radiocarbon of the labelled auxins. Non-promotion of such transport was associated with synergism by different synergistic chemicals whether an auxin remains fixed at the base by forming a complex with a non-auxinic chemical, which is biologically more active than either of them, is yet to be elucidated as the distribution of C₁₄ into different compounds was not much affected by the non-auxinic chemicals. A critical study is yet required to further elucidate the mechanism of synergism. Kling and Meyer (1983) studied effects of phenolic compounds and IAA on adventitious root

formation in cuttings of Phaseolus aureus and Acer griesum. Out of 21 phenolic compounds tested, p-hydroxy benzoic acid was negatively interacting with IAA activity. Bartolini et al. (1987) reported that p-hydroxy benzoic acid treatment gave more rooting percentage, root number and quantity of roots than when the cuttings of grapes root stock Ruggeri 140 were treated with combined treatment of IBA and p-hydroxy benzoic acid. Most of the tested phenolics have been shown to exert an influence on adventitious root formation, alone or with IBA. The fact that their action was often stimulative of the amount of roots produced, even in the less favourable period, seems to indicate that root elongation is more affected by the treatments than root initiation. Differences in the chemical structure of phenolic acids (length of side chain, number and position of hydroxy groups) involve significantly different effects on adventitious root formation and on the phenolic auxin interaction. The side chain is substantially able to modify the biological action of phenolics; in this sense different behaviour of cinnamic and benzoic acid should be noticed and opposed influence on the rooting of p-coumaric acid and p-hydroxy benzoic acid with respect to presence or absence of IBA treatment should be studied in detail.

v- NAA (Naphthalein Acetic Acid)

Gangwar and Singh (1968) reported that stem cuttings of grape were treated with 50, 100, 200 and 400 ppm NAA by standard immersion method before planting. NAA at concentration of 200 ppm gave 55.5 per cent rooting while its other strengths gave lower rooting (27-33.3%) than that of control (50%). Krack et al. (1983) revealed that NAA 2000 ppm induced marked modification in the hormonal patterns at different stages of root

formation in 140 Ruggeri and to a lesser extent in Kober 5 BB. Auxins doubled the IAA contents and induced two peaks during cell multiplication at the cutting base and immediately before root emergence. In general, NAA promoted a GA degradation so that after callusing the GA level was about half of that in control. NAA induced strong depletion of ABA in Kober 5 BB and to a lesser extent in 140 Ruggeri. Both treatments prevented a rise in ABA content before root development. Sherer et al. (1985) evaluated root inducing capacity of six growth regulators and best root formation was induced by NAA (0.005-0.01%) in grapevine root stock.

vi- Indole Acetic Acid (IAA)

Herath et al. (1978) found that in comparison to IBA, IAA did not improve the rooting of cuttings of grapes. Dorokhov (1983) reported that in easy to root cv. Moldova grape, a one hour dip in 0.01 per cent IAA gave good callusing and rooting but delayed bud burst.

F. ENVIRONMENTAL FACTORS

i- Temperature

Alley et al. (1974) reported that a spring having moderate temperature (30°C) and no hot spells resulted in a better take of cuttings of grapes, stored than one with high temperature spells in early summer (43°C). Fujii and Nokano (1974) reported optimum temperature 21°C for rooting of cutting of grapes in early spring and warm air temperatures further improved rooting. Goussard (1977) reported that bud break was also inhibited at higher temperature and longer exposure (above 55°C for longer than one minute in hot water) of cuttings of Cabernet Sauvignon grapes. Deidda and Agabbio (1978) reported that low temperature increase shooting and rooting of grapevine cuttings especially

in the basal and mid stem cuttings. Hosoi et al. (1979) reported that per cent rooting and number of roots per cutting were highest in cuttings rooted at 25°C regardless of initial weight of cuttings of Delaware vine while root growth and shoot growth were better with rising temperature at an early stage of propagation. The change in the rate of apparent photosynthesis expressed either on a cutting basis or a leaf area basis was related to root development. At later stage of propagation rooting was best at 20°C followed by 25°C and 30°C.

ii- **Light**

Ooshi et al. (1980) reported that cuttings of Delaware grapes grown in total darkness or in darkness from 10-40 days after planting showed reduced root growth, which then increased with increasing exposure to light.

ROOTING SUCCESS OF SEMI-HARDWOOD CUTTINGS OF GRAPES

Literature is lacking about rooting success of semi-hardwood cuttings. Moti and Singh (1968) reported that semi-hardwood cuttings of Bhokri and Gulabi cultivar of grape taken during July gave 34 per cent rooting which was very low from those of cuttings taken during the dormant season.

Jindal and Singh (1989) reported that summer cuttings prepared from benlate sprayed grapevine giving aerial roots had showed 90 percent survival in Beauty-Seedless and 68 percent in Pusa Seedless. However, cuttings of these two varieties without aerial roots (without benlate spray) showed 78 and 50 percent survival respectively.

CHAPTER-III

MATERIAL AND METHODS

The present investigations entitled "Studies on the effect of IBA and p-hydroxy benzoic acid on the rooting of hardwood and semi-hardwood cuttings of Perlette grape" were carried out at the Experimental Orchard of the Department of Horticulture, Haryana Agricultural University, Hisar during the year 1990-91. The details of experiment are as under :

PROPAGATION BY CUTTINGS

Seven thousand and two hundred cuttings (hardwood) and three hundred cuttings (semi-hardwood) of uniform size were taken from well established Perlette vines. Hardwood cuttings were collected after pruning in January at two dates and semi-hardwood cuttings were taken after pruning in July.

- Dates of taking cuttings** : i- 15th and 30th January (hardwood)
ii- 15th July (semi-hardwood)
- Callusing period** : i- 40 days for hardwood cuttings
ii- No callusing for semi-hard wood cuttings.
- Dates of planting cuttings** : i- 25th February and 11th March (hardwood)
ii- 15th July (semi-hardwood)

Number of treatments : Fifteen (both in hardwood and semi-hardwood)

Replications : Four (both in hardwood and semi-hardwood)

Design : Randomized Block Design (Pair-t-test)

Hardwood cuttings

Number of cuttings/replication : Sixty

Number of cuttings/treatment : Two hundred and forty

Number of cuttings/block : Three thousand and six hundred

Semi-hardwood cuttings

Number of cuttings/replication : Five

Number of cuttings/treatment : Twenty

Number of cuttings/block : Three hundred

Treatment combinations

- Control (T_0)
- IBA 800 ppm (T_1)
- IBA 1200 ppm (T_2)
- IBA 1600 ppm (T_3)
- IBA 2000 ppm (T_4)
- p-hydroxy benzoic acid 1000 ppm (T_5)
- p-hydroxy benzoic acid 2000 ppm (T_6)
- IBA 800 ppm + p-hydroxy benzoic acid 1000 ppm (T_7)
- IBA 1200 ppm + p-hydroxy benzoic acid 1000 ppm (T_8)
- IBA 1600 ppm + p-hydroxy benzoic acid 1000 ppm (T_9)
- IBA 2000 ppm + p-hydroxy benzoic acid 1000 ppm (T_{10})
- IBA 800 ppm + p-hydroxy benzoic acid 2000 ppm (T_{11})
- IBA 1200 ppm + p-hydroxy benzoic acid 2000 ppm (T_{12})
- IBA 1600 ppm + p-hydroxy benzoic acid 2000 ppm (T_{13})
- IBA 2000 ppm + p-hydroxy benzoic acid 2000 ppm (T_{14})

Preparations

Solution of concentration mentioned against each treatment was prepared just before giving the treatment. For example, 800 ppm IBA solution was prepared by adding 10 ml alcohol (50%) to 800 mg IBA powder to make the chemical completely soluble and final volume was made with distilled water upto one litre. In cuttings, treatments were applied to the basal one centimeter of cuttings for two minutes and then burried in soil for about 40 days for callusing of cuttings.

For planting of cuttings, nursery beds were thoroughly prepared by repeated digging with the help of spade in both the seasons. All sorts of weeds and pables were removed and land was finally levelled and bed was irrigated. Then planting was done after giving final digging and levelling. In nursery beds,, cuttings were planted, little bit slanted to the ground, in rows at one feet apart and cutting to cutting distance was kept 10 cm. It is important to note that the apex of cutting made with a slanting cut should remain above the ground and basal portion of cuttings made with smooth horizontal cut should be burried in ground. At the time of planting of cuttings (generally having 4 to 6 buds), approximately 2/3rd portion of each cutting was burried in soil. Two hundred and forty cuttings of each treatment were planted in nursery beds after treating with their respective concentrations in rows with randomization. The cuttings received uniform cultural practices throughout the experiment. The following observations were recorded :

A. INITIAL OBSERVATIONS

A sample of 15 cuttings was collected before and after callusing

and was used for taking following observations :

- a) Diameter at the base of cutting before and after callusing was recorded with the help of vernier caliper in centimeter .
- b) Average number of root initials after callusing were counted for each treatment.
- c) Initial status and status after callusing of cuttings for nitrogen, carbohydrates and phenols were determined.
 - Nitrogen was estimated by methods of analysis of soil, plant and water as described by Chapman and Pratt (1961).
 - Phenolic content was estimated by modified method of Swain and Hills (1959) as described by Amorium et al. (1977).
 - Carbohydrates in terms of total sugar was estimated by Ferricyanide method, a modification of Hagedorn Jenson, Hanes Technique (1929) as described by Haul and Narain (1931).

B. SHOOT GROWTH PARAMETERS

1. Sprouting

The cuttings under each treatment were examined after 7 days interval. The cuttings were considered to be sprouted when rudimentary leaves appeared atleast in one of the buds. On the basis of total number of cuttings, the cuttings sprouted were counted and percentage was worked out.

2. Average No. of bud sprouted per cutting

Number of buds sprouted per cutting in each treatment were recorded and average of sixty cuttings in each replication as worked out.

3. **Number of leaves per cutting**

Total number of leaves per cutting were counted and average data was presented on the basis of 60 cuttings per replication.

4. **Vigour of shoot after 60 and 120 days**

Vigour of shoot was determined by measuring the length and diameter of longest shoot. Average length and diameter of longest shoot was examined in each treatment.

C. **ROOT GROWTH PARAMETERS**

A sample of five cuttings for each treatment was uprooted and following parameters were recorded :

1. **Number of roots per cutting**

Total number of roots per cutting were counted under each treatment and their average values were calculated.

2. **Length of longest root**

Maximum length of root in each cutting under each treatment was measured with the help of scale and expressed in cm. Their average values were calculated.

3. **Fresh weight of roots**

All the roots from all the collected cuttings in each treatment were removed with the help of fine knife and their fresh weight was recorded immediately and expressed in 'g' per cutting.

4. **Dry weight of roots**

After recording the weight of fresh roots, roots were dried at 60°C to a constant weight in a forced draft oven and expressed as dry wt of roots in 'g' per cutting.

CHAPTER-IV

EXPERIMENTAL RESULTS

The investigations entitled "Studies on the effect of IBA and p-hydroxy benzoic acid on the rooting of hardwood and semi-hardwood cuttings of Perlette grape" were conducted on three different dates that is 15th and 30th January (hardwood) and 15th July (semi-hardwood) at the Experimental Orchard of Haryana Agricultural University, Hisar during 1990-91. The results obtained during the course of studies have been given in this chapter.

EXPERIMENT NO. 1 : Rooting of hardwood cuttings

Data recorded on propagation by hardwood cuttings with the aid of IBA and p-hydroxy benzoic acid in dormant season is presented below:

Initial observations

Initial observations before callusing in hardwood and semi-hardwood cuttings of Perlette vine with respect to diameter, status of nitrogen, carbohydrates, and phenols from the basal parts of cuttings were recorded (Table-1). It is clear from the data that hardwood cuttings taken during mid and end of January had more diameter than semi-hardwood cuttings collected during July. Hardwood cuttings were also rich in nitrogen and total carbohydrates content when determined from the basal part of the cuttings but semi-hardwood cuttings showed slightly more phenolic compounds.

Diameter

Diameter at the base of cutting after callusing was increased by

Table 1 : Initial observations before callusing in hard wood and semi-hardwood cuttings of Perlette grape

Initial observations	<u>Hardwood cuttings</u>		<u>Semi-hardwood cuttings</u>
	MJ	EJ	July
Diameter at the base of cutting (cm)	0.90	0.92	0.88
Nitrogen (%)	0.73	0.72	0.58
Carbohydrates (total sugar, mg/g)	10.10	10.00	8.50
Phenolic compounds(%)	0.24	0.25	0.26

MJ = Middle of January (15.1.1990)

EJ = End of January (30.1.1990)

different treatments in hardwood cutting collected during middle and end of January (Table-2). Average diameter of the cuttings collected during early part of January was more (1.17 cm) in comparison to later cuttings (1.14 cm).

Diameter in early collected cuttings after 40 days of callusing ranged between 0.95 to 1.45 cm while in later cuttings 0.93 to 1.43 cm. The maximum diameter (1.45 cm), in early collected cuttings, was recorded when cuttings were treated with 2000 ppm IBA (T_4) and it was significantly more than other treatments except T_3 , T_5 and T_{10} whereas minimum diameter at the base of cuttings was recorded in control. The treatment T_4 was followed by T_3 which was also significantly better than T_0 , T_1 , T_7 , T_{11} , T_{12} , T_{13} and T_{14} , while the remaining treatments were at par. All the treatments were effective in callus formation over control except T_1 , T_{11} , T_{12} , T_{13} and T_{14} . Data given in the table clearly indicates that higher concentration of p-hydroxy benzoic acid were not effective in enhancing callus formation while lower concentrations were effective over control. In case of late collected cuttings, maximum diameter (1.43 cm) was recorded when cuttings were treated with maximum concentration of IBA (T_4) and almost similar trend was observed as in cuttings collected during middle of January in all the treatments.

It is clear from the data, lowest concentration of IBA and its combination with lower concentration of p-hydroxy benzoic acid did not increase the diameter over control while best results were obtained when cuttings were treated with 2000 ppm IBA. However, higher concentration of p-hydroxy benzoic acid in combination with 2000 ppm IBA, was not effective

Table 2: Diameter (cm) of hardwood cuttings after callusing

Treatments	Hardwood cuttings	
	MJ	EJ
T ₀ Control	0.95	0.93
T ₁ 800 ppm IBA	1.07	1.05
T ₂ 1200 ppm IBA	1.20	1.15
T ₃ 1600 ppm IBA	1.32	1.28
T ₄ 2000 ppm IBA	1.45	1.43
T ₅ 1000 ppm PHBA	1.27	1.23
T ₆ 2000 ppm PHBA	1.22	1.20
T ₇ 800 ppm IBA + 1000 ppm PHBA	1.15	1.13
T ₈ 1200 ppm IBA + 1000 ppm PHBA	1.17	1.15
T ₉ 1600 ppm IBA + 1000 ppm PHBA	1.22	1.20
T ₁₀ 2000 ppm IBA + 1000 ppm PHBA	1.25	1.23
T ₁₁ 800 ppm IBA + 2000 ppm PHBA	1.00	1.00
T ₁₂ 1200 ppm IBA + 2000 ppm PHBA	1.02	1.03
T ₁₃ 1600 ppm IBA + 2000 ppm PHBA	1.07	1.05
T ₁₄ 2000 ppm IBA + 2000 ppm PHBA	1.12	1.07
Mean	1.17	1.14
CD at 5%	0.20	0.21

$t_{cal} = 6.67$

PHBA = p-hydroxy benzoic acid

MJ = Middle of January

EJ = End of January

over control.

Root initials

Number of root initials per cutting were influenced by different treatments in hardwood cuttings taken during middle and end of January (Table-3). Average number of root initials was more in early collected cuttings (5.70) than in late collected cuttings (4.53).

In case of early pruned cuttings number of root initials per cutting varied from 4.0 to 8.0 in various treatments. Maximum number of roots were recorded when cuttings were treated with 2000 ppm IBA which was significantly better than all other treatments except T₃ when cuttings were soaked in 1600 ppm IBA solution. However, T₃ was at par with T₂, T₅, T₆, T₉, T₁₀, T₁₃ and T₁₄ but significantly better than the remaining treatments. In case of late collected cuttings, average number of root initials per cuttings varied from 2.0 to 7.0 in various treatments. Like early cuttings, the maximum number of roots were recorded in cutting treated with 2000 ppm IBA and similar trend was observed in remaining all the treatments.

It is clear from the data that lowest concentration of IBA and its combination with the concentration of p-hydroxy benzoic acid did not increase the number of root initials in case of early collected cuttings but all the treatments increased the average number of roots ^{initials} in cuttings collected late in January.

Nitrogen

Reduction in nitrogen status at the base of cuttings was observed due to various treatments and callusing of hardwood cuttings for 40 days (Table-4). The average level of nitrogen was lower in cuttings collected during mid of



Plate No. 1 : Effect of 2000 ppm IBA (T_4) on number of initial roots (right) of cuttings taken during middle of January, in comparison to control (left)



Plate No. 2 : Effect of 2000 ppm IBA (T_4) on number of initial roots (right) of cuttings taken during end of January, in comparison to control (left)

Table 3 : Effect of various treatments on average number of root initials in hardwood cuttings of Perlette grape

Treatments		Hardwood cuttings	
		MJ	EJ
T ₀	Control	4.00	2.00
T ₁	800 ppm IBA	5.00	4.00
T ₂	1200 ppm IBA	6.00	5.00
T ₃	1600 ppm IBA	7.00	6.00
T ₄	2000 ppm IBA	8.00	7.00
T ₅	1000 ppm PHBA	6.50	5.00
T ₆	2000 ppm PHBA	6.00	4.00
T ₇	800 ppm IBA + 1000 ppm PHBA	5.00	4.00
T ₈	1200 ppm IBA + 1000 ppm PHBA	5.00	4.00
T ₉	1600 ppm IBA + 1000 ppm PHBA	6.00	5.00
T ₁₀	2000 ppm IBA + 1000 ppm PHBA	6.00	5.00
T ₁₁	800 ppm IBA + 2000 ppm PHBA	4.00	3.00
T ₁₂	1200 ppm IBA + 2000 ppm PHBA	5.00	4.00
T ₁₃	1600 ppm IBA + 2000 ppm PHBA	6.00	5.00
T ₁₄	2000 ppm IBA + 2000 ppm PHBA	6.00	5.00
Mean		5.70	4.53
CD at 5%		1.39	1.00

$t_{cal} = 13.00$

PHBA= p-hydroxy benzoic acid

MJ = Middle of January

EJ = End of January

Table 4 : Status of nitrogen (%) after callusing in hardwood cuttings of Perlette grapes

Treatments	Hardwood cuttings	
	MJ	EJ
T ₀ Control	0.70	0.72
T ₁ 800 ppm IBA	0.65	0.65
T ₂ 1200 ppm IBA	0.58	0.63
T ₃ 1600 ppm IBA	0.54	0.55
T ₄ 2000 ppm IBA	0.51	0.52
T ₅ 1000 ppm PHBA	0.68	0.69
T ₆ 2000 ppm PHBA	0.69	0.69
T ₇ 800 ppm IBA + 1000 ppm PHBA	0.66	0.67
T ₈ 1200 ppm IBA + 1000 ppm PHBA	0.65	0.66
T ₉ 1600 ppm IBA + 1000 ppm PHBA	0.64	0.65
T ₁₀ 2000 ppm IBA + 1000 ppm PHBA	0.63	0.64
T ₁₁ 800 ppm IBA + 2000 ppm PHBA	0.67	0.68
T ₁₂ 1200 ppm IBA + 2000 ppm PHBA	0.66	0.67
T ₁₃ 1600 ppm IBA + 2000 ppm PHBA	0.65	0.65
T ₁₄ 2000 ppm IBA + 2000 ppm PHBA	0.64	0.65
Mean	0.64	0.65
CD at 5%	0.20	0.21

T_{cal} = 5.00

PHBA = p-hydroxy benzoic acid

MJ = Middle of January

EJ = End of January

January (0.64%) than in late January collected cuttings (0.65%). The nitrogen contents at the base of cuttings collected early varied from 0.51 to 0.71 per cent. Early collected cuttings showed that the level of nitrogen was lowest in case when cuttings were dipped in 2000 ppm IBA (T_4) which was significantly lower than all other treatments and preceded by T_3 (1600 ppm IBA) which is again significantly lower than other treatments. All the treatments showed significant decline in nitrogen level of cuttings in comparison to control (T_0) except the treatments of alone p-hydroxy benzoic acid. Cuttings taken during the end of January with respect to level of nitrogen also followed similar trend as in early collected cuttings and the range of nitrogen varied from 0.52 to 0.72 per cent. All the treatments resulted in significant decline in the level of nitrogen at the base of cuttings in comparison to control.

It is clear from the data that highest concentration of IBA resulted in lowest level of nitrogen at the base of cutting.

Carbohydrates

It is clear from the data (Table-5) that carbohydrates content in terms of total sugar at the base of cuttings taken after callusing was more when cuttings were collected during the middle of January (16.32 mg/g) than end of January (13.24 mg/g).

The carbohydrate content in cuttings taken during the middle of January ranged between 10.15 to 21.51 mg/g in various treatments. Maximum level of carbohydrates (21.51 mg/g) was found in cuttings treated with 2000 ppm IBA (T_4) and it was significantly more than any other treatment except T_3 treatment (1600 ppm IBA) which was again significantly better than remaining treatments except T_{10} treatment when cuttings were treated with a combination

Table 5 : Status of carbohydrates in terms of total sugars at the base of cutting after callusing (mg/g)

Treatments		Hardwood cuttings	
		MJ	EJ
T ₀	Control	10.15	10.00
T ₁	800 ppm IBA	18.25	17.30
T ₂	1200 ppm IBA	20.63	20.02
T ₃	1600 ppm IBA	21.28	20.50
T ₄	2000 ppm IBA	21.51	21.40
T ₅	1000 ppm PHBA	10.63	10.30
T ₆	2000 ppm PHBA	10.58	10.20
T ₇	800 ppm IBA + 1000 ppm PHBA	14.61	14.00
T ₈	1200 ppm IBA + 1000 ppm PHBA	16.89	16.40
T ₉	1600 ppm IBA + 100 ppm PHBA	19.60	18.60
T ₁₀	2000 ppm IBA + 1000 ppm PHBA	20.90	20.10
T ₁₁	800 ppm IBA + 2000 ppm PHBA	12.50	12.51
T ₁₂	1200 ppm IBA + 2000 ppm PHBA	13.18	13.00
T ₁₃	1600 ppm IBA + 2000 ppm PHBA	16.85	15.00
T ₁₄	2000 ppm IBA + 2000 ppm PHBA	18.25	16.20
Mean		16.32	13.24
CD at 5%		0.58	0.30

$t_{cal} = 4.60$

PHBA = p-hydroxy benzoic acid

MJ = Middle of January

EJ = End of January

of 2000 ppm IBA and 1000 ppm p-hydroxy benzoic acid. It is also clear that both the concentration of p-hydroxy benzoic acid alone were not effective in improving the carbohydrates level at the base of cuttings over the control. However, p-hydroxy benzoic acid in combination with IBA concentration showed negative effect and this effect was more pronounced than higher concentration of p-hydroxy benzoic acid was combined with lower concentration of IBA than with higher concentration of IBA. Like early cuttings maximum carbohydrates were found (21.40 mg/g) in late cuttings treated with 2000 ppm IBA, though almost similar trend was observed in both the cases.

Phenols

The total phenolic contents recorded after callusing was found increased (Table-6). Phenols at the base of cuttings taken after callusing were less when cuttings were collected during middle of January (0.86%) than in cuttings taken late in January (0.88%). The range of total phenols at the base of cuttings collected during middle of January varied from 0.25 to 1.64 per cent. The maximum concentration of phenols (1.64%) in early collected cuttings was found when treated with 2000 ppm IBA in combination with 2000 ppm p-hydroxy benzoic acid (T_{14}) and it was significantly higher than the untreated (0.25%) and the cuttings treated with lowest concentration of IBA but it was at par with remaining treatments. The treatment T_{14} was followed by T_{13} which is at par with all the treatments except control. The minimum phenols were found in control. There was increasing trend with increasing concentration of IBA, p-hydroxy benzoic acid and their combinations. The range of phenols in late pruned cuttings varied from 0.27 to 1.66 per cent.

Table 6 : Status of phenols (%) of cuttings after callusing

Treatments		Hardwood cuttings	
		MJ	EJ
T ₀	Control	0.25	0.27
T ₁	800 ppm IBA	0.34	0.37
T ₂	1200 ppm IBA	0.46	0.47
T ₃	1600 ppm IBA	0.54	0.54
T ₄	2000 ppm IBA	0.55	0.55
T ₅	1000 ppm PHBA	0.57	0.59
T ₆	2000 ppm PHBA	0.82	0.84
T ₇	800 ppm IBA + 1000 ppm PHBA	0.84	0.87
T ₈	1200 ppm IBA + 1000 ppm PHBA	0.91	0.93
T ₉	1600 ppm IBA + 1000 ppm PHBA	0.96	0.98
T ₁₀	2000 ppm IBA + 1000 ppm PHBA	1.01	1.02
T ₁₁	800 ppm IBA + 2000 ppm PHBA	1.19	1.22
T ₁₂	1200 ppm IBA + 2000 ppm PHBA	1.25	1.27
T ₁₃	1600 ppm IBA + 2000 ppm PHBA	1.52	1.55
T ₁₄	2000 ppm IBA + 2000 ppm PHBA	1.64	1.66
Mean		0.86	0.88
CD at 5%		1.18	1.16

t_{cal} = 4.60

MJ = Middle of January

PHBA = p-hydroxy benzoic acid

EJ = End of January

The treatment of highest concentration of IBA in combination with highest concentration of p-hydroxy benzoic acid (T_{14}) resulted in highest phenols at the base of cuttings. The trend was almost similar as in early pruned vine cuttings.

It is clear from the data that highest concentration of IBA (2000 ppm) in combination with highest concentration of p-hydroxy benzoic acid (2000 ppm) increased the phenolic status of cuttings significantly. However, the level of phenols was also found increased even when cuttings were treated with alone IBA, p-hydroxy benzoic acid and IBA combination with lowest concentration of p-hydroxy benzoic acid though insignificant.

Sprouting of cuttings

Data recorded on per cent sprouting of cuttings as influenced by time of taking cuttings and various root promotive treatments is presented in Fig.1 and Fig. 2.

Effect of time of taking cuttings

The results given in Fig.1 revealed that per cent sprouting increased upto 35 days of planting and the cuttings taken during mid January showed significantly better per cent sprouting (68%) than the cuttings collected during end of January (52%). Early collection of cuttings was significantly superior to late collection of hardwood cuttings.

Effect of IBA and p-hydroxy benzoic acid

All the treatments differed when compared with control in both the timings of taking cuttings that is on 15th and 30th January (Fig.2). The sprouting of cuttings was influenced by different treatments during both the timings of taking cuttings. With an increase in IBA concentration, there was

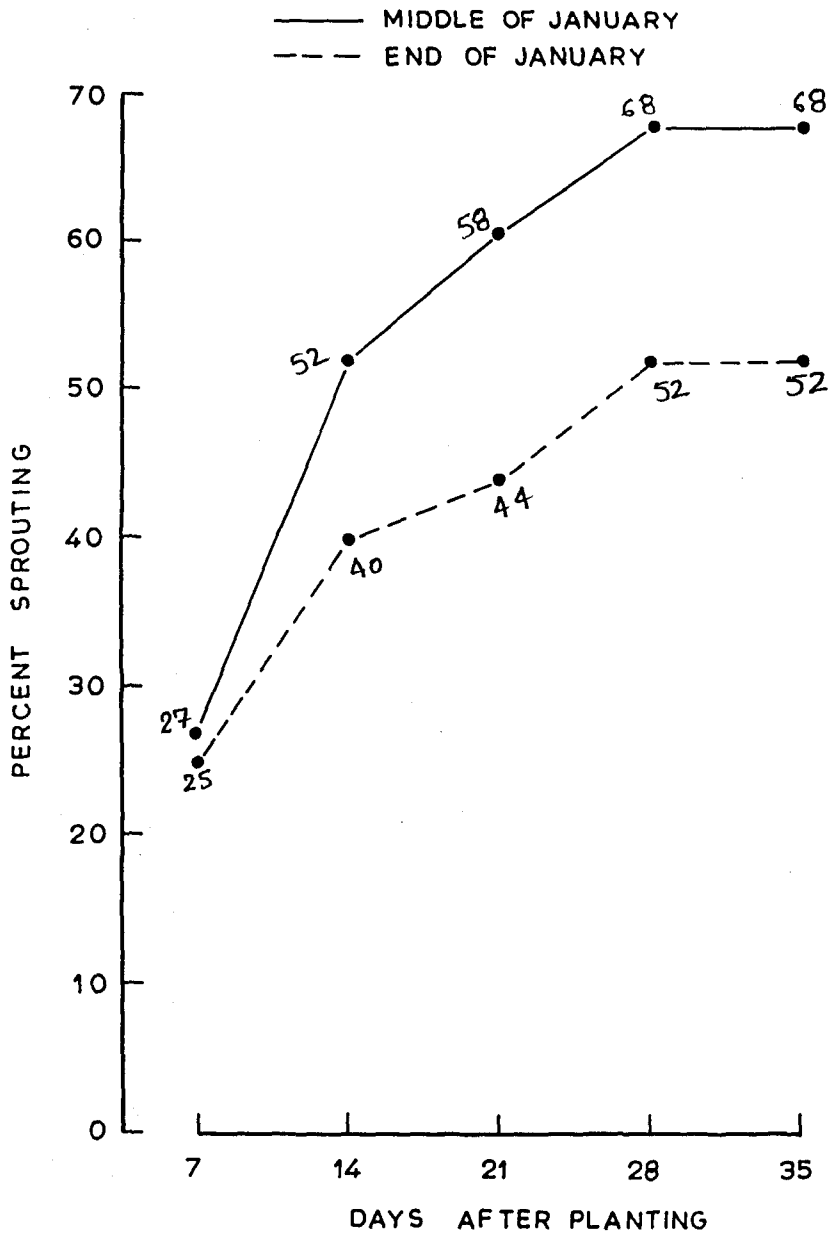


Fig.1 PERCENT SPROUTING AS EFFECTED BY TIME OF TAKING CUTTING

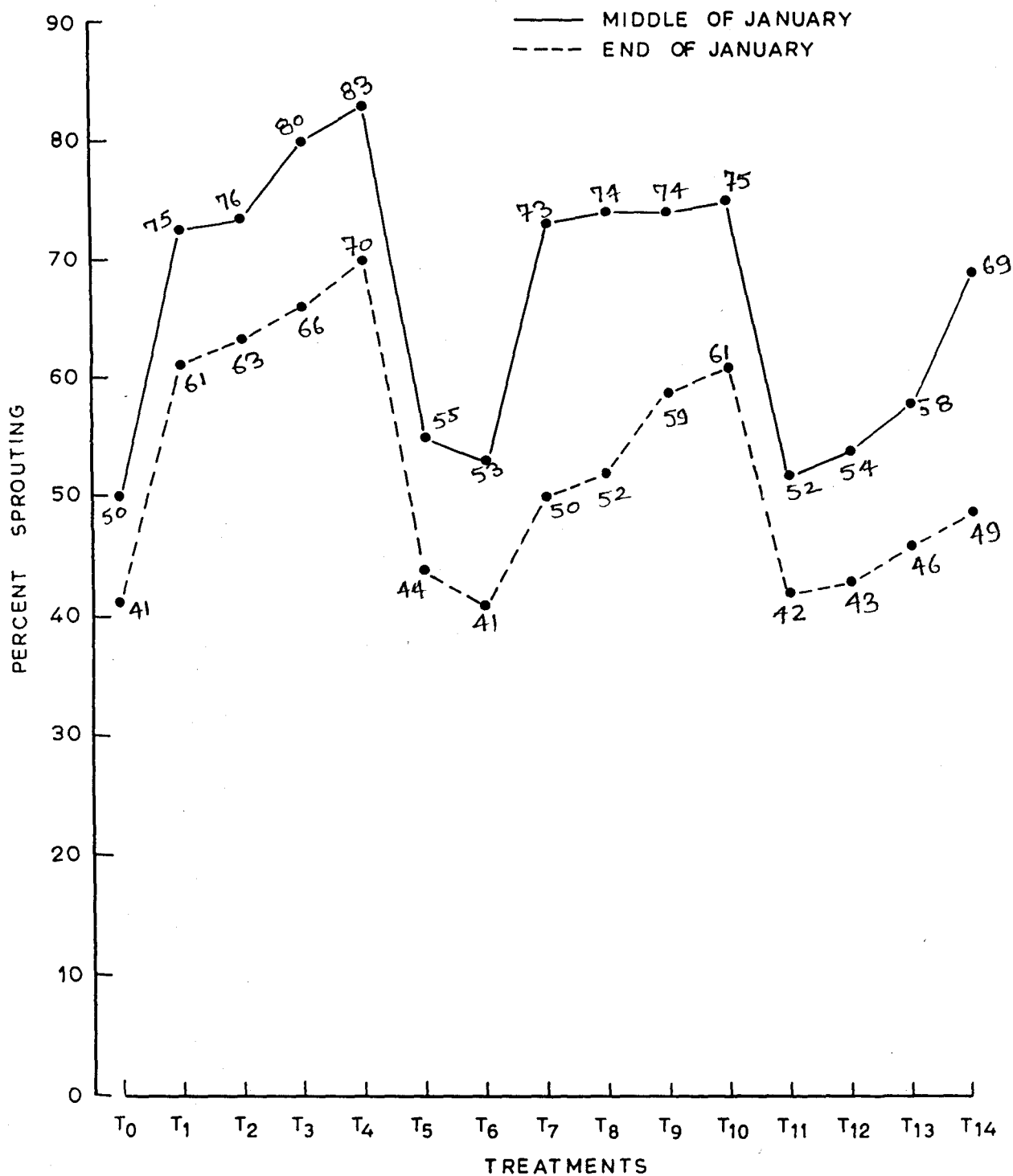


Fig.2. PERCENT SPROUTING AS EFFECTED BY DIFFERENT TREATMENTS

increase in per cent sprouting of cuttings. Addition of p-hydroxy benzoic acid, however, decreased the sprouting of cuttings, in both the timings of taking cuttings but were still better than control.

The perusal of the data clearly states that IBA in combination with lower concentration of p-hydroxy benzoic acid proved better with respect to percentage of cuttings sprouted than its combination with higher concentration of p-hydroxy benzoic acid and alone p-hydroxy benzoic acid treatments. The maximum sprouting in early collected cuttings (83%) and in late collected cuttings (70%) was obtained with highest concentration of IBA.

Sprouting of buds

Average number of sprouted buds and leaves were recorded after 35 days of planting of hardwood cuttings.

Buds

Sprouting of buds was influenced by different treatments (Table-7). Average number of sprouted buds per cutting was more in early collected cuttings (0.95) than in late collected cuttings (0.89) and it ranged from 0.69 to 1.32 in early collected cuttings. Maximum number of sprouted buds was recorded (1.32) in cuttings treated with 2000 ppm IBA (T_4) and T_4 treatment was followed by T_3 treatment (1600 ppm IBA). The number was significantly more than T_2 and T_3 treatments. All the treatments increased the average number of sprouted buds over control except the treatments of highest concentration of p-hydroxy benzoic acid and all the concentrations of IBA in combination with highest concentration of p-hydroxy benzoic acid. The range of average number of buds in cuttings taken during late January varied from 0.66 to 1.24. The maximum concentration of IBA resulted in

Table 7 : Average number of sprouted buds after 35 days of planting of hardwood cuttings

Treatments	Hardwood cuttings	
	MJ	EJ
T ₀ Control	0.69	0.66
T ₁ 800 ppm IBA	1.03	0.90
T ₂ 1200 ppm IBA	1.28	1.20
T ₃ 1600 ppm IBA	1.31	1.21
T ₄ 2000 ppm IBA	1.32	1.24
T ₅ 1000 ppm PHBA	0.92	0.82
T ₆ 2000 ppm PHBA	0.79	0.77
T ₇ 800 ppm IBA + 1000 ppm PHBA	0.94	0.90
T ₈ 1200 ppm IBA + 1000 ppm PHBA	0.97	0.95
T ₉ 1600 ppm IBA + 1000 ppm PHBA	1.00	0.97
T ₁₀ 2000 ppm IBA + 1000 ppm PHBA	1.01	0.99
T ₁₁ 800 ppm IBA + 2000 ppm PHBA	0.69	0.65
T ₁₂ 1200 ppm IBA + 2000 ppm PHBA	0.70	0.68
T ₁₃ 1600 ppm IBA + 2000 ppm PHBA	0.72	0.69
T ₁₄ 2000 ppm IBA + 2000 ppm PHBA	0.82	0.76
Mean	0.95	0.89
CD at 5%	0.15	0.11

$t_{cal} = 8.33$

MJ = Middle of January

PHBA = p-hydroxy benzoic acid

EJ = End of January

maximum sprouting of buds. Almost similar trend was observed like in early collected cuttings.

It is clear from the data that IBA alone treatments are most effective in increasing the average number of sprouted buds in both the timings of taking cuttings. IBA in combination with highest concentration of p-hydroxy benzoic acid did not increase the sprouting of buds. However, lowest concentration of p-hydroxy benzoic acid and its combination with IBA showed some positive effect. Alone p-hydroxy benzoic acid treatment increased the average number of buds sprouted over control in hardwood cuttings taken early and late in January.

Leaves

The number of leaves per cutting was increased by various treatments (Table-8). The average number of leaves was more when cuttings were collected in middle of January (11.5) than late January (10.2). The number was ranged from 4.10 to 15.00 in cuttings taken during middle of January. The maximum number of leaves per cutting (15.00) was obtained where cuttings were treated with 2000 ppm IBA (T_4) and the number was significantly more than the remaining treatments. The treatment T_4 was followed by T_3 (1600 ppm IBA) which gave significantly more number of leaves than the remaining treatments. All the treatments resulted in an increase in the number of leaves in comparison to control. The average number of leaves in cuttings collected in the end of January varied from 3.40 to 12.90 and maximum number of leaves were obtained from cuttings treated with 2000 ppm IBA. The trend was almost similar as in early pruned vine cuttings.

It is clear from the data that IBA alone treatments were most

Table 8 : Average number of leaves after 35 days of planting of hardwood cuttings

Treatments	Hardwood cuttings	
	MJ	EJ
T ₀ Control	4.10	3.40
T ₁ 800 ppm IBA	13.50	12.30
T ₂ 1200 ppm IBA	13.70	12.40
T ₃ 1600 ppm IBA	14.60	12.50
T ₄ 2000 ppm IBA	15.00	12.90
T ₅ 1000 ppm PHBA	10.20	9.80
T ₆ 2000 ppm PHBA	9.80	8.30
T ₇ 800 ppm IBA + 1000 ppm PHBA	12.10	10.90
T ₈ 1200 ppm IBA + 1000 ppm PHBA	12.70	11.20
T ₉ 1600 ppm IBA + 1000 ppm PHBA	12.90	11.30
T ₁₀ 2000 ppm IBA + 1000 ppm PHBA	13.00	11.40
T ₁₁ 800 ppm IBA + 2000 ppm PHBA	9.90	8.50
T ₁₂ 1200 ppm IBA + 2000 ppm PHBA	10.10	9.70
T ₁₃ 1600 ppm IBA + 2000 ppm PHBA	10.30	9.90
T ₁₄ 2000 ppm IBA + 2000 ppm PHBA	10.60	10.30
Mean	11.50	10.20
CD at 5%	0.14	0.12

$t_{cal} = 5.90$

MJ = Middle of January

EJ = End of January

PHBA = p-hydroxy benzoic acid

effective. However, phenolic compound treatments alone and their combination with IBA also showed positive effect but IBA with lower concentration of p-hydroxy benzoic acid gave more number of leaves than its combination with highest concentration of p-hydroxy benzoic acid and alone p-hydroxy benzoic acid treatments.

Mortality

Per cent mortality recorded after 60 days of planting of hardwood cuttings at both the timings, as influenced by different treatments is depicted in Fig.3.

The perusal of the data revealed that percentage of mortality decreased with the increasing concentration of IBA in both the timings of taking cuttings. With the addition of p-hydroxy benzoic acid to various concentrations of IBA, there was ~~no further~~ ⁱⁿ decrease in per cent mortality cuttings. However, alone p-hydroxy benzoic acid treatment and their combination with IBA reduced the per cent mortality over control. The maximum mortality (20, 25%) of cuttings occurred whereas minimum mortality (8, 13%) was observed when cuttings were treated with 2000 ppm IBA (T₄).

It is clear from the data that cuttings taken during the middle of January showed lesser mortality in comparison to the cuttings taken in the end of January. Application of even lower concentrations (1200 ppm and 1600 ppm) of IBA during early cuttings were also better than higher concentration of IBA 2000 ppm when applied late.

Vigour of shoots

Vigour of sprouts was determined by measuring the diameter and length of longest shoot after 60 and 120 days of planting of hardwood cuttings.

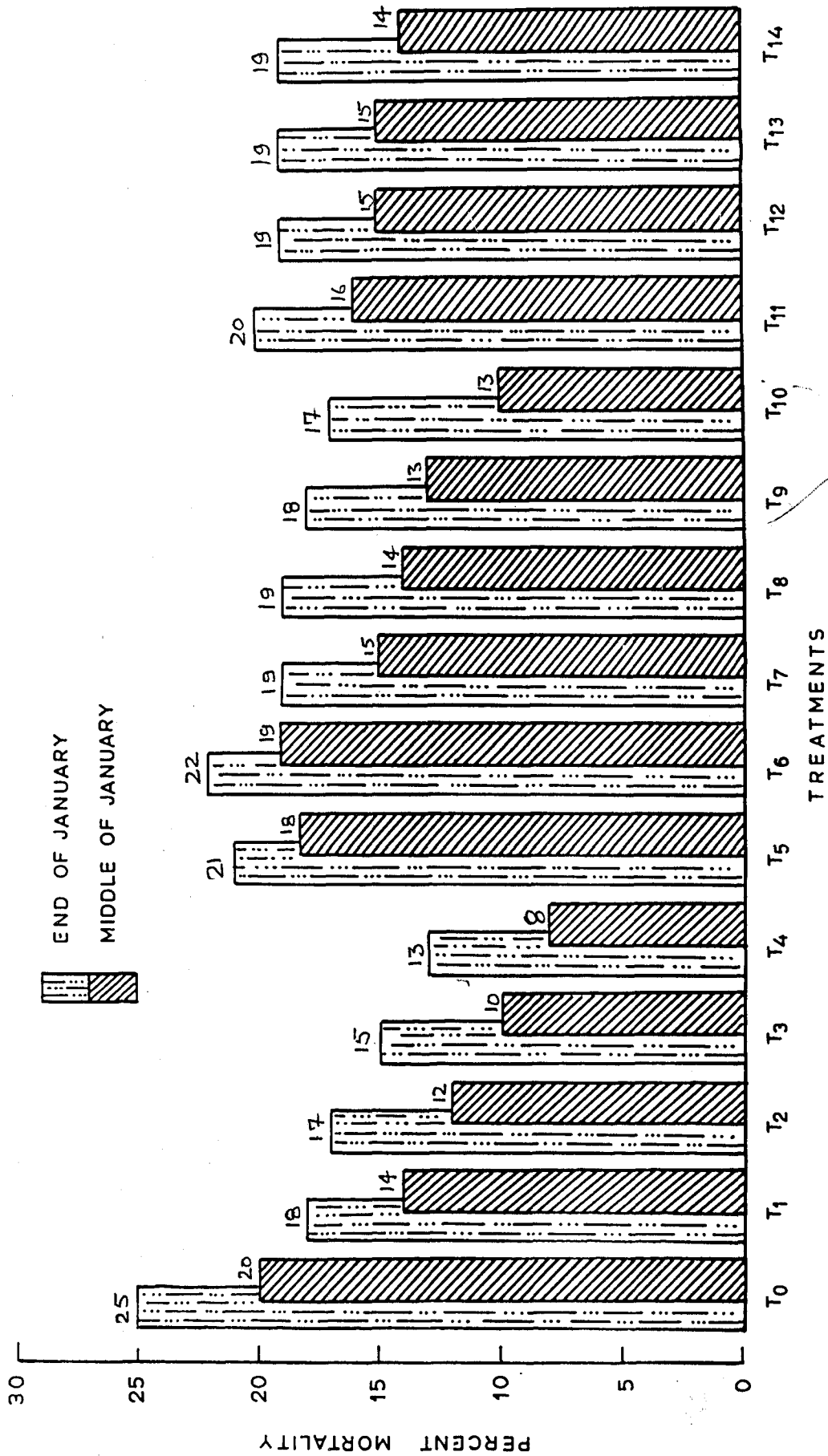


Fig. 3. PERCENT MORTALITY AS INFLUENCED BY DIFFERENT TREATMENTS

In general more vigorous shoots were obtained in cuttings taken during mid January (dia. 0.23 cms, length 26.31 cms) in comparison to the shoot obtained from late collected cuttings (dia.0.22 cms, length 23.12 cms).

After 60 days

The perusal of the data given in Table-9 showed that average diameter of shoot produced from cuttings taken during middle of January (0.23 cm) was more than that of shoot obtained from cuttings taken in the end of January (0.22 cm). The range of diameter of shoot in case of early collected cuttings varied from 0.20 to 0.28 cm as influenced by different treatments. Maximum diameter (0.28 cm) of shoot was obtained from the cuttings treated with 2000 ppm IBA (T_4) when pruning was done during early dormant season, and it was significantly more than the diameter resulted by other treatments. The treatment T_4 was followed by T_3 and T_5 that is when cuttings were dipped in 1600 ppm IBA and 1000 ppm p-hydroxy benzoic acid respectively. All the treatments gave more diameter at shoot over control. In case of cuttings taken during the end of January, the diameter of shoot ranged between 0.20 to 0.25 cm. Maximum diameter (0.25 cm) of shoot was obtained from cuttings treated with 2000 ppm IBA (T_4). Almost similar trend was observed as in case of early pruned vine cuttings.

Average length of shoot produced from cuttings taken during mid January (26.31 cm) was more than that of shoot obtained from cuttings taken during late January (23.12 cm) as shown in Table-9. The range of length of shoot in case of early collected cuttings varied from 14.40 to 35.70 cm as influenced by different treatments. Maximum length of shoot (35.70 cm) was obtained from the cuttings treated with 2000 ppm IBA (T_4)

Table 9 : Vigour of shoot after 60 days of planting of hardwood cuttings of grape

Treatments		Hardwood cuttings			
		D(cm)	MJ L(cm)	EJ D(cm)	L(cm)
T ₀	Control	0.20	14.40	0.20	14.30
T ₁	800 ppm IBA	0.23	29.50	0.21	22.50
T ₂	1200 ppm IBA	0.24	31.10	0.22	29.67
T ₃	1600 ppm IBA	0.25	32.10	0.23	30.33
T ₄	2000 ppm IBA	0.28	35.70	0.25	32.07
T ₅	1000 ppm PHBA	0.25	20.00	0.23	18.00
T ₆	2000 ppm PHBA	0.22	19.70	0.22	17.67
T ₇	800 ppm IBA +1000 ppm PHBA	0.22	25.20	0.21	18.67
T ₈	1200 ppm IBA+1000 ppm PHBA	0.23	28.70	0.21	22.67
T ₉	1600 ppm IBA+1000 ppm PHBA	0.23	28.80	0.22	26.33
T ₁₀	2000 ppm IBA+1000 ppm PHBA	0.24	29.00	0.23	27.00
T ₁₁	800 ppm IBA+2000 ppm PHBA	0.22	24.10	0.20	18.33
T ₁₂	1200 ppm IBA+2000 ppm PHBA	0.22	24.20	0.20	21.33
T ₁₃	1600 ppm IBA+2000 ppm PHBA	0.23	25.20	0.21	23.00
T ₁₄	2000 ppm IBA+2000 ppm PHBA	0.23	27.00	0.22	24.67
Mean		0.23	26.31	0.22	23.12
CD at 5% level		0.01	1.20	0.01	4.62

t_{cal} =6.67 for D (Diameter) MJ = Middle of January

t_{cal} =6.42 for L (Length) EJ = End of January

PHBA =p-hydroxy benzoic acid

when pruning was done during early dormant season and it was significantly more than the diameter resulted by other treatments. The treatment T_4 was followed by T_3 (1600 ppm IBA), T_3 in turn is at par with T_2 (1200 ppm IBA) and significantly better than other treatments. All the treatments gave more length over control. In case of cuttings taken during end of January, the length of shoot ranged between 14.30 and 32.07 cm. Maximum length of shoot (32.07 cm) was obtained from cuttings treated with 2000 ppm IBA (T_4). Almost similar trend was observed as in case of early pruned vine cuttings.

It is clear from the data that alone IBA treatments, particularly higher concentration of IBA, increased the vigour of shoot. Lower concentration of p-hydroxy benzoic acid in combination with various concentrations of IBA, however, was more effective in increasing the vigour of shoot, in comparison to IBA combination with highest concentration of p-hydroxy benzoic acid and alone p-hydroxy benzoic acid. Alone p-hydroxy benzoic acid treatments also increased the vigour of shoot obtained from cuttings collected in middle and end of January.

After 120 days

The perusal of the data given in Table-10 showed that average diameter of shoot produced from cuttings taken during middle of January (0.36 cm) was more than that of shoot obtained from cuttings taken in the end of January (0.34 cm). The range of diameter of shoot in case of early collected cuttings varied from 0.34 cm to 0.39 cm as influenced by different treatments. Maximum diameter (0.39 cm) of shoot was obtained from cuttings treated with 2000 ppm IBA (T_4) when pruning was done during early dormant season, and it

Table 10 : Vigour of shoot after 120 days of planting of hardwood cuttings

Treatments	Hardwood cuttings			
	MJ		EJ	
	D(cm)	L(cm)	(D(cm)	L(cm)
T ₀ Control	0.34	55.30	0.33	49.00
T ₁ 800 ppm IBA	0.35	75.80	0.34	71.70
T ₂ 1200 ppm IBA	0.36	77.57	0.35	76.20
T ₃ 1600 ppm IBA	0.38	83.80	0.36	80.20
T ₄ 2000 ppm IBA	0.39	88.30	0.38	82.00
T ₅ 1000 ppm PHBA	0.36	73.30	0.34	74.20
T ₆ 2000 ppm PHBA	0.35	66.00	0.33	61.30
T ₇ 800 ppm IBA + 1000 ppm PHBA	0.35	71.00	0.34	55.70
T ₈ 1200 ppm IBA + 1000 ppm PHBA	0.36	72.30	0.34	61.00
T ₉ 1600 ppm IBA + 1000 ppm PHBA	0.36	73.30	0.35	62.70
T ₁₀ 2000 ppm IBA + 1000 ppm PHBA	0.36	74.00	0.35	67.50
T ₁₁ 800 ppm IBA + 2000 ppm PHBA	0.33	56.30	0.32	50.00
T ₁₂ 1200 ppm IBA + 2000 ppm PHBA	0.35	60.40	0.33	51.40
T ₁₃ 1600 ppm IBA + 2000 ppm PHBA	0.36	62.00	0.34	53.00
T ₁₄ 2000 ppm IBA + 2000 ppm PHBA	0.36	66.00	0.34	57.30
Mean	0.36	65.98	0.34	63.55
CD at 5%	0.02	3.10	0.02	0.80

t_{cal} = 6.67 for D (Diameter)

MJ = Middle of January

t_{cal} = 6.92 for L (Length)

EJ = End of January

PHBA= p-hydroxy benzoic acid

was significantly more than the diameter resulted by other treatments except T_3 (1600 ppm IBA). No other treatment gave better response over control except T_3 and T_4 . In case of cuttings taken during end of January, the diameter shoot ranged from 0.33 to 0.38 cm. Maximum diameter (0.38 cm) of shoot was obtained from cuttings treated with 2000 ppm IBA (T_4) and almost similar trend was observed as in early pruned vine cuttings.

The data given in Table-10 showed that average length of shoot produced from cuttings taken during middle of January (65.98 cm) was more than that of shoot obtained from cuttings taken during late January (63.55 cm). The range of length of shoot in case of early collected cuttings varied from 55.30 cm to 88.30 cm as influenced by different treatments. Maximum length of shoot (88.30 cm) was obtained from the cuttings treated with 2000 ppm IBA (T_4) when pruning was done during early dormant season and it was significantly better than the diameter resulted by other treatments. The treatment T_4 was followed by T_3 (1600 ppm IBA) which is again significantly better than remaining treatments. All the treatments gave more length over control. In case of cuttings collected during end of January, the length of shoot ranged between 49.00 to 82.00. Maximum length of shoot (82.0 cm) was obtained from cuttings treated with 2000 ppm IBA (T_4). Almost similar trend was observed as in case of early pruned vine cuttings.

It is clear from the data that IBA alone treatments are most effective in increasing the vigour of shoot. Diameter of shoot was not increased by combined treatments while length of shoot was found increased by all the treatments. Diameter of shoot obtained from cuttings collected during middle January was at par with control when cuttings were treated with lowest

concentration of p-hydroxy benzoic acid but highest concentration of p-hydroxy benzoic acid, the diameter of shoot was negatively effected. In late collected cuttings, both the concentrations of p-hydroxy benzoic acid alone showed negative effect with respect to diameter of shoot. Length of shoot obtained from cuttings taken in middle of January and end of January was increased by all the treatments over control, while diameter of shoot was not increased by combined treatments in the cuttings taken after early and late pruning in January both.

Root growth of cuttings

Data on root growth of hardwood cuttings with respect to average number of roots, length of longest root, fresh and dry root weights was recorded in the coming season after one year of both the cases. that is cuttings taken during middle and end of January.

Number of roots

Number of roots per cutting was increased by various treatments (Table-11). Average number of roots was more in cuttings collected during middle of January (12.82) than in late January (9.33). Average number of roots varied from 8.25 to 17.50 by different treatments in early collected cuttings. The maximum number of roots were recorded when cuttings were treated with 2000 ppm IBA (T_4) which was significantly more than other treatments except T_3 when cuttings were dipped in 1600 ppm IBA solution. However, T_3 was at par with T_2 but gave significantly more number of roots than other treatments. Rooting capacity of cuttings was made better by all treatments than untreated control except the alone p-hydroxy benzoic acid

Table 11 : Average number of roots in hardwood cuttings of grapes

Treatments		Hardwood cuttings	
		MJ	EJ
T ₀	Control	8.25	6.00
T ₁	800 ppm IBA	13.75	9.00
T ₂	1200 ppm IBA	15.25	10.50
T ₃	1600 ppm IBA	16.50	12.25
T ₄	2000 ppm IBA	17.50	15.00
T ₅	1000 ppm PHBA	9.50	6.50
T ₆	2000 ppm PHBA	9.00	6.00
T ₇	800 ppm IBA + 1000 ppm PHBA	12.50	7.25
T ₈	1200 ppm IBA + 1000 ppm PHBA	13.00	8.75
T ₉	1600 ppm IBA + 1000 ppm PHBA	14.00	10.50
T ₁₀	2000 ppm IBA + 1000 ppm PHBA	14.50	12.00
T ₁₁	800 ppm IBA + 2000 ppm PHBA	11.25	7.50
T ₁₂	1200 ppm IBA + 2000 ppm PHBA	11.75	7.50
T ₁₃	1600 ppm IBA + 2000 ppm PHBA	12.00	9.50
T ₁₄	2000 ppm IBA + 2000 ppm PHBA	13.50	11.50
Mean		12.82	9.33
CD at 5%		1.32	0.97

$t_{cal} = 12.89$

PHBA = p-hydroxy benzoic acid

MJ = Middle of January

EJ = End of January

treatments. Cuttings collected late in January showed the range of average number of roots 6 to 15. Maximum number of roots (15) were recorded when cuttings were treated with 2000 ppm IBA (T_4). Almost similar trend was noticed as in case of early collected cuttings.

It is clear from the data that highest concentration of IBA was most effective in increasing the average number of roots. However, IBA in combination with p-hydroxy benzoic acid also increased the number of roots but over control only. Alone p-hydroxy benzoic acid treatments did not increase the number of roots per cutting. There was a reduction in the positive effect of IBA in combination with p-hydroxy benzoic acid on average number of roots though this reduction was more pronounced when high concentration of p-hydroxy benzoic acid was used.

Length of the longest root

There was increase in length as affected by various treatments (Table-12) when cuttings were taken in middle and end of January. Average length of root was more in cuttings taken during middle of January (19.04 cm) than in cuttings taken during end of January (17.95 cm). Length of longest root varied from 13.75 to 20.21 cm in cuttings taken during middle of January. The maximum length was obtained in cuttings treated with 2000 ppm IBA in combination with 2000 ppm p-hydroxy benzoic acid (T_{14}), followed by T_3 which was at par with T_{11} and T_{12} but produced significantly more length of roots in comparison to remaining treatments. All the treatments were better than control. The length in late collected cuttings ranged between 12.00 to 20.10 cm. The maximum length of root was obtained from cuttings treated with 2000 ppm IBA in combination with 2000 ppm p-hydroxy benzoic

Table 12 : Length of largest root (cm) in hardwood cuttings

Treatments	Hardwood cuttings	
	MJ	EJ
T ₀ Control	13.75	12.00
T ₁ 800 ppm IBA	18.85	16.00
T ₂ 1200 ppm IBA	18.95	16.50
T ₃ 1600 ppm IBA	19.02	17.00
T ₄ 2000 ppm IBA	19.03	17.20
T ₅ 1000 ppm PHBA	19.07	18.00
T ₆ 2000 ppm PHBA	19.32	18.20
T ₇ 800 ppm IBA + 1000 ppm PHBA	19.35	18.40
T ₈ 1200 ppm IBA + 1000 ppm PHBA	19.42	18.70
T ₉ 1600 ppm IBA + 1000 ppm PHBA	19.50	18.90
T ₁₀ 2000 ppm IBA + 1000 ppm PHBA	19.54	19.00
T ₁₁ 800 ppm IBA + 2000 ppm PHBA	19.75	19.50
T ₁₂ 1200 ppm IBA + 2000 ppm PHBA	19.80	19.70
T ₁₃ 1600 ppm IBA + 2000 ppm PHBA	20.10	20.00
T ₁₄ 2000 ppm IBA + 2000 ppm PHBA	20.21	20.10
Mean	19.04	17.95
CD at 5%	0.37	0.80

$t_{cal} = 4.78$

PHBA = p-hydroxy benzoic acid

MJ = Middle of January

EJ = End of January

acid. Almost similar trend was observed as in early collected cuttings. It is clear from data that various concentrations of IBA in combination with highest concentration of p-hydroxy benzoic acid were more effective in increasing length of root in comparison to alone IBA treatments, p-hydroxy benzoic acid treatments and treatments of combination of IBA with lower concentration of p-hydroxy benzoic acid. Results showed that phenolic contents are responsible for increasing length of roots.

Fresh and dry root weights

Fresh and dry roots weights were found increased by various treatments (Table-13) in hardwood cuttings taken during middle and end of January. Fresh and dry root weights (13.95 and 12.32 g respectively) were more in cuttings taken during middle of January in comparison to the cuttings taken during end of January (8.60 and 6.90 g respectively).

Fresh and dry root weights in cuttings taken during middle of January were ranged between 9.60 to 17.20 g and 4.4 to 12.0 g respectively. Maximum fresh weight (17.20 g) and dry weight (12.0 g) of roots were obtained from cuttings treated with 2000 ppm IBA (T_4) which was significantly better than remaining treatments except T_3 (1600 ppm IBA) to which it was at par with respect to fresh weight of roots. All the treatments provided more fresh and dry root weight in comparison to untreated control. Fresh and dry root weight in cuttings taken during end of January ranged between 7.00 to 16.50 g and 3.20 to 9.50 g respectively. Almost similar trend was obtained as in early collected cuttings and all the treatments gave more fresh and dry root weights in comparison to untreated control.

Table 13: Fresh and dry weight of roots in hardwood cuttings (g) of Perlette grapes

Treatments	Fresh weight of roots per plant (g)		Dry weight of roots per plant (g)	
	MJ	EJ	MJ	EJ
T ₀ Control	9.60	7.00	4.40	3.20
T ₁ 800 ppm IBA	14.00	12.00	8.70	6.50
T ₂ 1200 ppm IBA	15.90	13.00	10.20	8.50
T ₃ 1600 ppm IBA	16.60	16.00	11.30	8.75
T ₄ 2000 ppm IBA	17.20	16.50	12.00	9.50
T ₅ 1000 ppm PHBA	11.40	10.50	6.10	5.25
T ₆ 2000 ppm PHBA	11.10	10.00	5.90	5.00
T ₇ 800 ppm IBA + 1000 ppm PHBA	13.50	11.75	8.00	6.00
T ₈ 1200 ppm IBA + 1000 ppm PHBA	14.50	12.25	9.10	7.50
T ₉ 1600 ppm IBA + 1000 ppm PHBA	14.80	14.00	9.50	8.25
T ₁₀ 2000 ppm IBA + 1000 ppm PHBA	15.80	15.00	9.70	8.75
T ₁₁ 800 ppm IBA + 2000 ppm PHBA	13.20	10.50	7.90	6.00
T ₁₂ 1200 ppm IBA + 2000 ppm PHBA	13.50	10.75	8.30	6.20
T ₁₃ 1600 ppm IBA + 2000 ppm PHBA	13.80	12.50	8.50	6.50
T ₁₄ 2000 ppm IBA + 2000 ppm PHBA	14.40	13.00	9.20	7.40
Mean	13.95	12.32	8.60	6.90
CD at 5%	0.80	0.78	0.20	0.51

$t_{cal} = 11.33$

MJ = Middle of January

PHBA = p-hydroxy benzoic acid

EJ = End of January

It is clear from the data that fresh and dry root weights of cuttings taken during middle and end of January were found increased by all the treatments over control. Alone IBA treatments are most effective followed by the treatments of IBA in combination with lower concentration of p-hydroxy benzoic acid than treatments of IBA combined with highest concentration of p-hydroxy benzoic acid and finally the treatments of p-hydroxy benzoic acid alone. Higher concentration of p-hydroxy benzoic acid alone or in combination with IBA showed inhibiting effects of IBA alone.

Rooting and changes in biochemical constituents

Changes in biochemical constituents with respect to average number of roots of hardwood cuttings of grapes (Table-14) showed that number of roots per cutting was negatively correlated with nitrogen, positively correlated with carbohydrates. C/N ratio and negatively but negligibly correlated with phenolic status of base of cutting. The maximum number of roots per cutting (17.50) was correlated with minimum level of nitrogen (0.51%), maximum level of carbohydrates (21.51 mg/g) and C/N ratio (42.18) and optimum level of phenols (0.55%). However, further increase in concentration of phenols was negatively correlated with comparative decrease in the average number of roots. The minimum number of roots per cutting (8.25) was correlated with maximum level of nitrogen (0.70%), minimum level of carbohydrates (10.15 mg/g), C/N ratio (14.50) and phenols (0.25%) at the base of cutting.

EXPERIMENT NO. 2 : Rooting of semi-hardwood cuttings

Data recorded on propagation by semi-hardwood cuttings with the aid of IBA and p-hydroxy benzoic acid in July season is presented below :

Table 14 : Changes in biochemical constituents of grape cuttings

Tr.	1 No. of roots	2 Nitrogen(%)	3 Carbohydrates (total sugar, mg/g)	4 C/N ratio	5 Phenols(%)
T ₀	8.25	0.70	10.15	14.50	0.25
T ₁	13.75	0.65	18.25	28.07	0.34
T ₂	15.25	0.58	20.63	35.57	0.46
T ₃	16.50	0.54	21.28	39.41	0.54
T ₄	17.50	0.51	21.51	42.18	0.55
T ₅	9.50	0.68	10.63	15.63	0.57
T ₆	9.00	0.69	10.58	15.33	0.82
T ₇	12.50	0.66	14.61	22.14	0.84
T ₈	13.00	0.65	16.89	25.98	0.91
T ₉	14.00	0.64	19.60	30.63	0.96
T ₁₀	14.50	0.63	20.90	33.17	1.01
T ₁₁	11.25	0.67	12.50	18.66	1.19
T ₁₂	11.75	0.66	13.18	19.97	1.25
T ₁₃	12.00	0.65	16.85	25.92	1.52
T ₁₄	13.50	0.64	18.25	28.52	1.64

$$r_{1,2} = -0.92$$

$$r_{1,3} = +0.95$$

$$r_{1,4} = +0.98$$

$$r_{1,5} = -0.10$$

Sprouting of cuttings

Data recorded on per cent sprouting as influenced by time of planting of cuttings that is July and various root promotive treatments.

Effect of time of planting

Data for per cent sprouting was recorded at weekly interval in July season upto 42 days. Sprouting capacity of cuttings went upto 40 per cent after 42 days of planting of cuttings and depicted in Fig.4.

Effect of treatments

Maximum percentage of sprouting was shown by cuttings treated with 2000 ppm IBA (T_4) as depicted in Fig.5 and remaining three concentrations of IBA showed 40 per cent sprouting. All other treatments combinations did not increase the percentage of cuttings sprouted except T_{10} where highest concentration of IBA was combined with lowest concentration of p-hydroxy benzoic acid. It is clear from data that alone IBA treatments are most effective in increasing the per cent sprouting; IBA in combination with highest concentration of p-hydroxy benzoic acid did not increase the percentage of sprouting over control.

Sprout production

Data was recorded on average number of buds and leaves as influenced by various treatments (Table-15) after 42 days of planting of semi-hardwood cuttings.

Buds

Average number of buds sprouted per cutting were not increased by different treatments except the treatments of higher concentration of IBA (1600 ppm and 2000 ppm IBA) and range varied from 0.2 to 0.5.

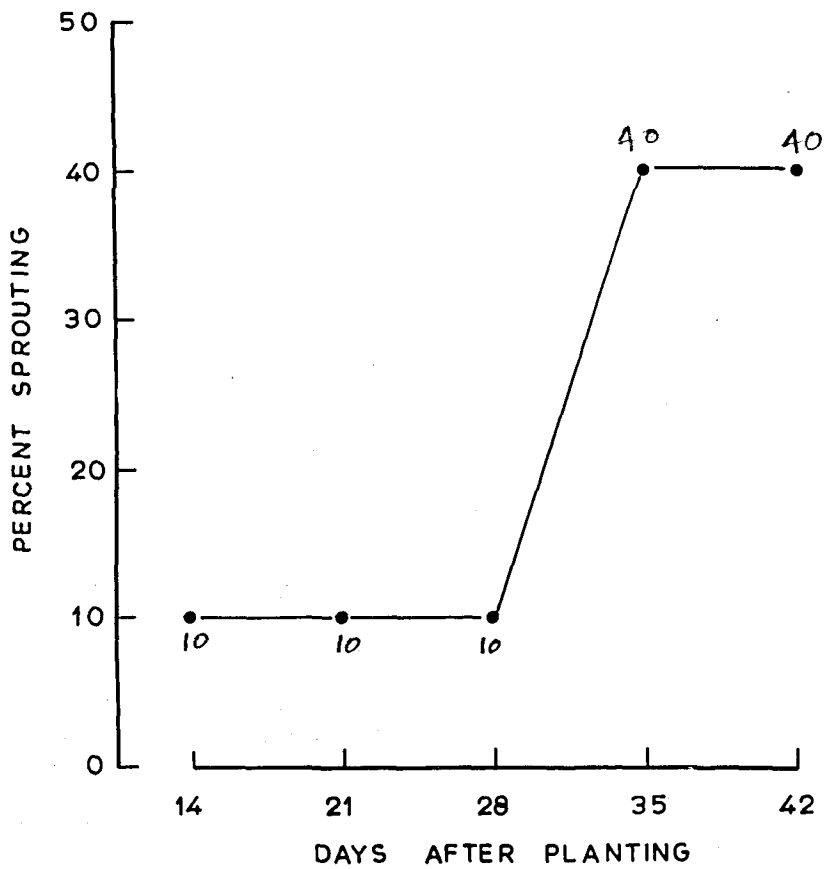


Fig. 4. PERCENT SPROUTING IN JULY PLANTED CUTTINGS

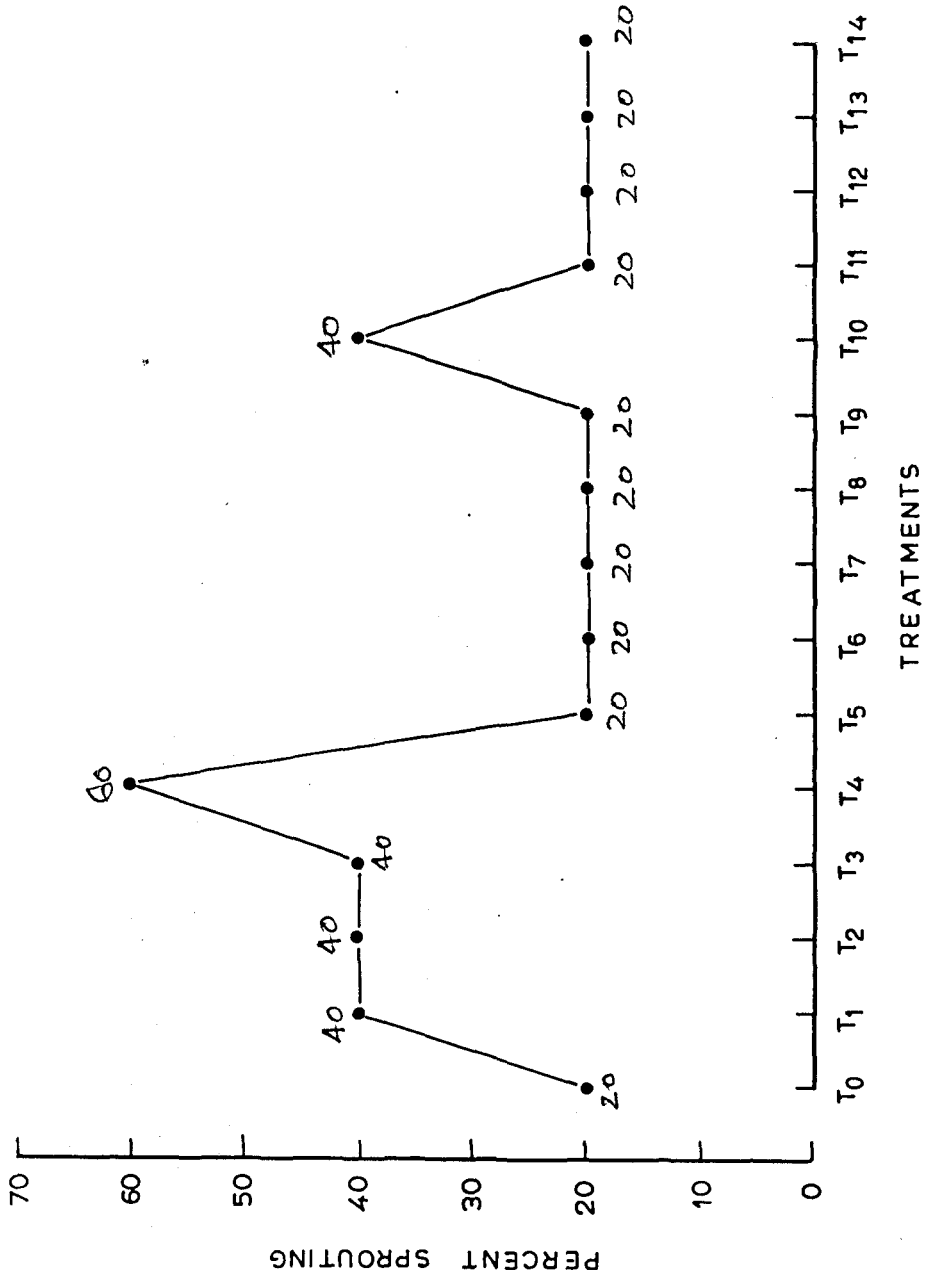


Fig. 5. PERCENT SPROUTING AS INFLUENCED BY DIFFERENT TREATMENTS IN JULY

Table 15 : Growth of semi-hardwood cuttings of Perlette grapes after 42 days of planting

Treatments		Average No. of buds sprouted	Average No. of leaves
T ₀	Control	0.20	1.00
T ₁	800 ppm IBA	0.20	1.37
T ₂	1200 ppm IBA	0.20	1.52
T ₃	1600 ppm IBA	0.50	1.60
T ₄	2000 ppm IBA	0.50	2.42
T ₅	1000 ppm PHBA	0.20	1.20
T ₆	2000 ppm PHBA	0.20	1.03
T ₇	800 ppm IBA+ 1000 ppm PHBA	0.20	1.03
T ₈	1200 ppm IBA + 1000 ppm PHBA	0.20	1.10
T ₉	1600 ppm IBA + 1000 ppm PHBA	0.20	1.40
T ₁₀	2000 ppm IBA + 1000 ppm PHBA	0.30	1.60
T ₁₁	800 ppm IBA + 2000 ppm PHBA	0.20	0.97
T ₁₂	1200 ppm IBA + 2000 ppm PHBA	0.20	1.07
T ₁₃	1600 ppm IBA + 2000 ppm PHBA	0.20	1.25
T ₁₄	2000 ppm IBA + 2000 ppm PHBA	0.20	1.35
Mean		0.25	1.33
CD at 5%		0.10	0.12

Leaves

Average number of leaves ranged between 1.00 to 2.42 as influenced by different treatments. Maximum number of leaves were obtained with treatments of 2000 ppm IBA (T_4) which gave significantly more number of leaves than other treatments. T_4 was followed by T_3 which was again significantly better than other treatments except T_2 . All the treatments are better than control except the treatments of higher concentration of p-hydroxy benzoic acid alone and also the lower concentration of IBA in combination with p-hydroxy benzoic acid.

It is clear from the data that alone IBA treatments of higher concentration were effective in increasing the average number of sprouted bud and average number of leaves. However, combinations of higher concentration of IBA and p-hydroxy benzoic acid increased the average number of leaves while lower concentration of IBA (800 ppm and 1600 ppm) in combination with higher concentration of p-hydroxy benzoic acid did not increase the number of leaves over control. However, there was no growth in semi-hardwood cuttings of Perlette grape. Though cuttings remained viable and the emerging leaves were in tact till the end of growing season.

Mortality

Percentage of mortality was recorded after 60 days of planting of semi-hardwood cuttings (Fig.6). Minimum mortality (40%) was recorded in cuttings treated with 2000 ppm IBA (T_4) whereas maximum mortality (80%) was recorded in untreated control (T_0). Remaining treatment combinations showed 60 per cent mortality.

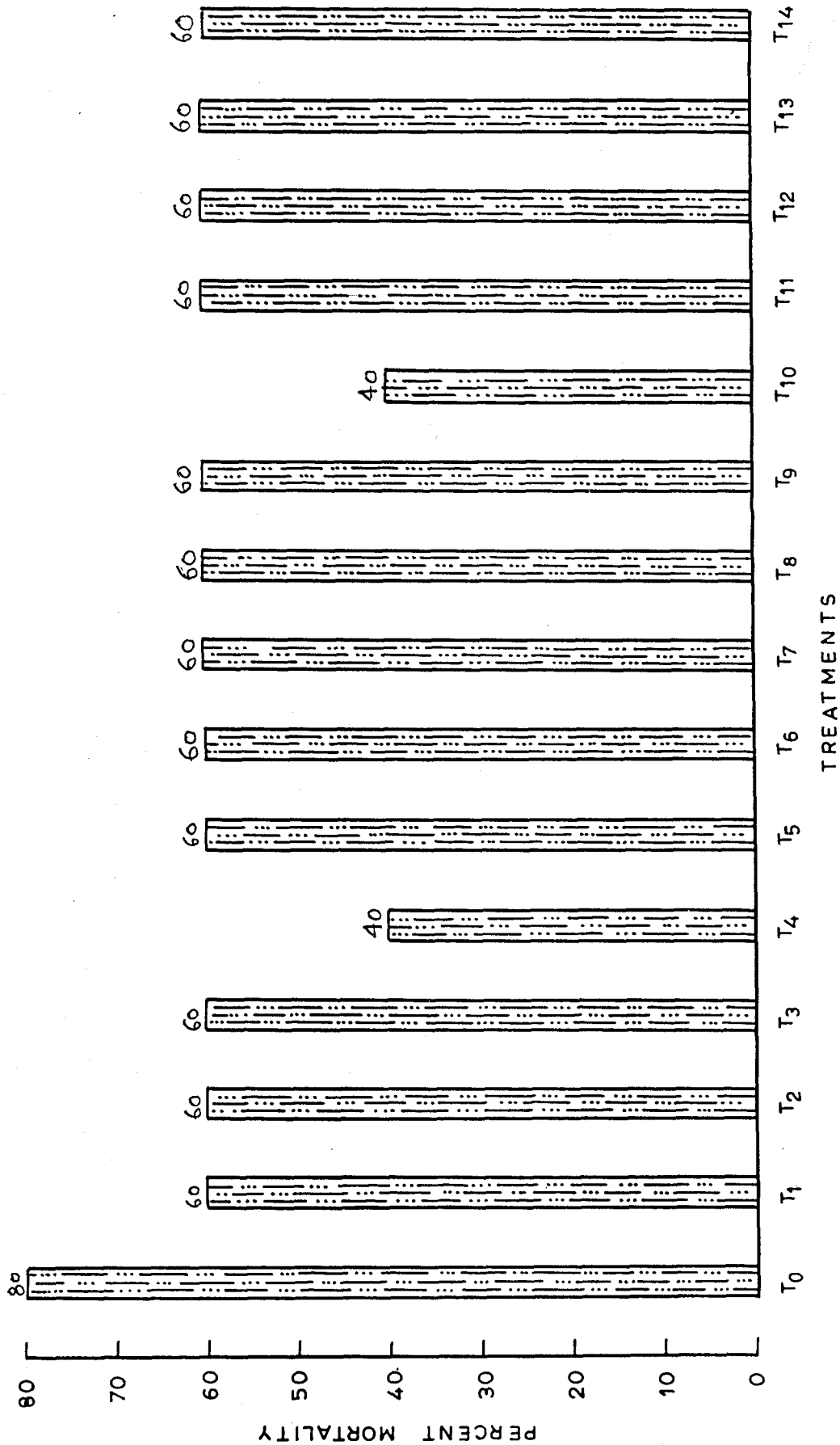


Fig. 6. PERCENT MORTALITY AS INFLUENCED BY DIFFERENT TREATMENTS IN JULY

Root growth

Very little rooting success was recorded in semi-hardwood cuttings (Table-16). Only higher concentrations of IBA and their combinations with lower concentration of p-hydroxy benzoic acid showed rooting response but poorly. Cuttings also remained green for one year though no rooting was observed at the end of the experiment. It is clear from the data that chances of survival of semi-hardwood cuttings of Perlette grape during July are very less.

Note: It has been statistically proved by applying pair t-test that mid January collected cuttings are superior to late January collected cuttings. For every parameter t-cal value is given below each table.

Table 16 : Root growth of semi-hardwood cuttings of Perlette grapes

Treatments	No. of root per plant	Length of largest root(cm)	Root fresh weight (cm)	Dry weight of roots (cm)
T ₀ Control	-	-	-	-
T ₁ 800 ppm IBA	-	-	-	-
T ₂ 1200 ppm IBA	2.00	2.00	1.00	0.50
T ₃ 1600 ppm IBA	3.00	2.00	1.50	1.20
T ₄ 2000 ppm IBA	5.00	2.50	2.00	1.60
T ₅ 1000 ppm PHBA	-	-	-	-
T ₆ 2000 ppm PHBA	-	-	-	-
T ₇ 800 ppm IBA + 1000 ppm PHBA	-	-	-	-
T ₈ 1200 ppm IBA + 1000 ppm PHBA	-	-	-	-
T ₉ 1600 ppm IBA + 1000 ppm PHBA	2.00	1.00	0.50	0.20
T ₁₀ 2000 ppm IBA + 1000 ppm PHBA	2.00	1.00	1.00	0.30
T ₁₁ 800 ppm IBA + 2000 ppm PHBA	-	-	-	-
T ₁₂ 1200 ppm IBA + 2000 ppm PHBA	-	-	-	-
T ₁₃ 1600 ppm IBA + 2000 ppm PHBA	-	-	-	-
T ₁₄ 2000 ppm IBA + 2000 ppm PHBA	-	-	-	-
Mean	0.93	0.56	0.40	0.25
CD at 5%	-	-	-	-

CHAPTER-V

DISCUSSION

The present investigations entitled " Studies on the effect of IBA and p-hydroxy benzoic acid on the rooting of hardwood and semi-hardwood cuttings of Perlette grape" were conducted to find out the rooting success of hardwood and semi-hardwood cuttings with the aid of IBA and p-hydroxy benzoic acid in two seasons that is January and July. Results of these studies have been discussed in succeeding paragraphs.

Hardwood cuttings taken during middle and end of January have more diameter than semi-hardwood cuttings collected during July. It is due to better development and maturity of shoots as hardwood cuttings were collected at the end of growing season while semi-hardwood cuttings were collected during growing conditions. Hardwood cuttings were also comparatively rich in nitrogen and total carbohydrates content but semi-hardwood cuttings have showed slightly higher level of phenolic contents. It is further supported by the fact that higher level of carbohydrates and nitrogen in hardwood cuttings indicates that January collected cuttings are in better physiological conditions than July season collected stem cuttings having immature wood.

Diameter at the base of hardwood cuttings taken on 15th and 30th January after callusing was recorded. The diameter of the cuttings collected early in dormant season was more than in case of cuttings collected late. Possibly it is due to more callusing activities at the base of cuttings

because of favourable environmental conditions. The best results were obtained with 2000 ppm IBA followed by its combination with lower concentration of p-hydroxy benzoic acid. However, 2000 ppm IBA in combination with 2000 ppm p-hydroxy benzoic acid have not increased the diameter. The better response of IBA with respect to increase in diameter is due to its property of cell division and callus formation as reported by Hartman and Kester (1962). The non-additive effect of p-hydroxy benzoic acid may be the existing sufficient amount of phenolics in the cuttings of grape.

The nitrogen contents of cuttings after callusing was lower in early collected cuttings than in late collected cuttings. This lowest level of nitrogen content was recorded in cuttings treated with 2000 ppm IBA, which also resulted in maximum callusing as indicated by increase in diameter at the base of cuttings and average number of root initials. So, it is clear that for better root initiation lower level of nitrogen and higher concentration of IBA is most effective. This decrease in nitrogen level is due to its utilisation during callusing and root initials development. This is further supported by the findings of Basu et al. (1972) in mango cuttings and revealed that decreasing level of nitrogen is indicative that it is being utilised during the process of rhizogenesis.

The carbohydrates contents of cuttings after callusing was higher in early collected cuttings than in late collected cuttings due to the obvious reason of favourable environmental conditions for accumulation of carbohydrates during early dormant season when the growth of vines was completely checked. The highest level of carbohydrates was recorded in cutting treated with 2000 ppm IBA which also resulted in maximum callusing at the base of cuttings. However, p-hydroxy benzoic acid was not effective in increasing the carbohydrates level,

rather it affected carbohydrates when it was applied in combination with IBA. It seems that phenolic treatments may not be favourable for accumulation and diversion of carbohydrates towards the base of cuttings. IBA application to the base of cuttings increased the carbohydrates level of cuttings. It is due to the property of IBA starting metabolisation of sugars and nitrogenous substances from the buds and stem which ultimately helps in initiation of root primordia in cuttings. Hartman and Kester (1962) reported that IBA prevents co-agulation of protein and carbohydrate at the cut portion, activates cambial cells and permit root emergence, and increases RNA formation during root primordia formation which leads to protein synthesis and results in cell wall extension. So, first root initiation cells are dependent on auxins.

The phenolic contents of cuttings after callusing were slightly less in early collected cuttings in comparison to late collected cuttings. This may be due to variable physiological maturity of shoots increasing concentration of IBA from 800 ppm to 2000 ppm increased the phenolic status of cuttings and brought it to the optimum level (0.55%) at which maximum root initials were obtained. The treatments of p-hydroxy benzoic acid alone and in combination with IBA further increased the phenolic contents of cuttings resulting in supra-optimal concentration of phenols in cuttings and hence number of root initials were reduced at these concentrations when compared with 2000 ppm IBA treatments. Increased level of phenolics in cuttings treated with IBA was also reported by Kaundal and Malkiat Singh (1987) in pear cuttings and revealed that IBA is responsible for the release of phenolic content present in buds towards the base of cutting.

The average number of root initials are more in early collected cuttings in comparison to late collected cuttings. It seems that physiological conditions of cuttings during mid-January are more conducive to root initiation than during late January, which may be responsible for differences in root initiation. Similar observations were recorded by Saraswat (1973) in Bhokri and Selection-7 varieties of grape and Weaver et al. (1975) in Carignane grape. It is interesting to note that lower concentration of IBA and its combination with p-hydroxy benzoic acid have not increased the average number of root initials in early collected cuttings but all the treatments have increased the average number of root initials in cuttings collected late in January, though best results were obtained with 2000 ppm IBA. This may be due to ecological adjustment.

Foregoing discussions reveals that various treatments and callusing increasing the carbohydrates level and decreased nitrogen level while phenolic contents were increased. The correlation of above parameters indicates the various treatments which increases C/N ratio and maintains optimum phenolic level ultimately results in better development of root initials at the base of cutting.

Examination of the data reveals that per cent sprouting of buds, number of leaves and vigour of shoot was more in early collected cuttings in comparison to late collected cuttings. Better growth in early collected cuttings is obviously due to better development of root initials as stated earlier. The differential response of different dates of planting or collecting cuttings may be the mainly influence of prevailing climatic conditions and their effects on carbohydrates reserves. Similarly, Moti and Singh (1968) and Saraswat (1973) reported that early collected cuttings were superior to late collected

cuttings with respect to shoot growth parameters. The best results with respect to per cent sprouting, number of leaves and vigour of shoot were obtained with 2000 ppm IBA treatment followed by 1600 ppm IBA treatment. Hartman and Hansen (1958) reported more vegetative growth with IBA application to plum cuttings. Application of p-hydroxy benzoic acid to the cuttings did not increase the above growth parameters though it was better than control. The results of treatment combinations of p-hydroxy benzoic acid and IBA were also significantly poorer than IBA alone. This non-additive effect of p-hydroxy benzoic acid may be due to higher inherent status of phenols in cuttings or the higher amounts of phenolics are not required for growth and establishment of grape cuttings. However, positive effect of IBA in combination with p-hydroxy benzoic acid was reported by Rakesh (1987) with respect to shoot parameters in lemon.

The mortality of cuttings taken during January and planted after 40 days of callusing was less in comparison to cuttings taken late in January due to the obvious reason of better root initials which resulted in more shoot growth. The positive effect of callusing may be explained by the fact that cuttings planted in nursery developed better shoot whereas in non-callused or cuttings with no root initials stopped their growth in nursery after leafing out and died due to lack of root development. Hartman and Hansen (1958) and Saraswat (1973) reported better establishment of cuttings by precallusing. The perusal of the data also revealed that cuttings treated with 2000 ppm IBA showed minimum mortality in both the timings of taking cuttings. It is interesting to note that due to favourable conditions early taking of cuttings gave equally good performance even if lower

concentration of IBA (1200 ppm and 1600 ppm) whereas higher concentration of IBA (2000 ppm) was required during late taking and planting of cuttings. Garner (1958) observed that environment of planting influences the performance of cuttings.

The average number of roots, length of longest root and fresh and dry root weight were more in case of early collected cuttings than in late collected cuttings. This may be due to the better callusing and better biochemical and physiological conditions for rooting during early collection and planting of cuttings. Better callusing is indicative of larger diameter of cutting collected early. Similarly Goode et al. (1982) reported that diameter cuttings rooted better than small diameter cuttings. The other reason for better rooting in early collected and planted cuttings may be the more favourable environmental conditions like temperature (17°C) than late collected cuttings (24°C). Studies of Alley et al. (1974) in Dogridge grape; Goussard (1977) in Cabernet savignon grapes and Hosoi et al. (1979) in Delaware grape supported the present findings that cuttings under moderate temperature (15-20°C) conditions rooted better than cuttings with higher temperature conditions (30°C and above).

The treatment of IBA 2000 ppm had the maximum number of roots and fresh and dry root weights as compared to other concentrations of IBA, p-hydroxy benzoic acid and their combinations. Similar beneficial effects of 2000 ppm IBA with respect to increase in the quantity of rooting were also reported in stem cuttings of lemon by Arora and Yamdagni (1985). The inhibition or non-promotion of average number of roots and fresh and dry root weight by the p-hydroxy benzoic acid treatments alone

and by combined treatments of IBA and p-hydroxy benzoic acid may be due to the higher inherent value of phenols in the cuttings which further increased to undesirable levels of phenols due to the application of p-hydroxy benzoic acid. Present studies conform to the findings of Bartolini et al. (1987) who reported that p-hydroxy benzoic acid in combination with IBA caused decrease in per cent rooting and quantity of roots in comparison to treatments of IBA and p-hydroxy benzoic acid alone in 140 Ruggeri grape cuttings. It seems that differences in chemical structure of phenolic acids (length of side chain, and the number and position of hydroxy groups) involve significantly different effects on adventitious root formation, and on phenolic/auxin interaction, but none of the above chemical feature alone can be seen as a determinant of causing the biological action of phenolics whose activity appears instead determined by its structure as a whole. Another reason for this inhibition of IBA by p-hydroxy benzoic acid was given by Turetskaya et al. (1987) by beans that some phenolics like p-coumaric acid do not have the auxin sparing action from IAA oxidase enzyme. The findings of Basu (1970) in Phaseolus vulgaris conforming to the present findings further explained that antagonism between auxins and non-auxinic chemicals is always accompanied by increased upward transport of radiocarbon of labelled auxins. Non-promotion of such transport is associated with synergism by different synergistic chemicals. The root length was maximum when cuttings were treated with highest concentration of IBA in combination with highest concentration of p-hydroxy benzoic acid contrary to other growth parameters. This may be due to the reason that IBA even alone increases the phenolic status of cuttings as discussed earlier and with the addition of p-hydroxy

benzoic acid, there is further increase in phenolic status of cuttings which was not so effective in root initiation while found effective in root elongation. Obviously the lesser number of roots per cutting resulted in more vigour as measured by the average length of root. Similarly Bartolini et al. (1987) reported the phenols cause root elongation more effectively even in less favourable conditions, than root initiation in 140 Ruggeri grape cuttings.

Levels of carbohydrates, phenolic contents, nitrogen and C/N ratio in hardwood cuttings were co-related with average number of roots. Average number of roots were positively correlated with nitrogen and negligibly negative correlation was observed with total phenols. It has been reported that high C/N ratio favoured rhizogenesis in pear (Hyun, 1976; Singh, 1976). Similar correlations were noted with respect to carbohydrates and nitrogen in almond peach hybrid (Kaundal and Bindra, 1984).

It is clear from the results obtained during studies that performance of semi-hardwood cuttings collected during July was poor in all respects of growth. However, IBA treatment was effective to some extent as the cuttings sprouted and remained green with 2 or 3 leaves attached without making any further growth whereas without IBA, cuttings failed to survive. The main reason of failure in semi-hardwood cuttings may be the immaturity of wood, lower C/N ratio and growing conditions.

It is concluded from the discussion that hardwood cuttings performed better than semi-hardwood cuttings. The hardwood cuttings collected early in the dormant season were superior to late collected cuttings with respect to all the growth parameters. The various treatments and callusing increased

the carbohydrates level and decreased the nitrogen level while phenolic contents were increased. The best results were obtained from cuttings treated with 2000 ppm IBA as it was most effective in increasing C/N ratio and overall growth of rooted cuttings.

CHAPTER-VI

SUMMARY

The present investigations " Studies on the effect of IBA and p-hydroxy benzoic acid on the rooting of hardwood and semi-hardwood cuttings of Perlette grape" were carried out at the Experimental Orchard of Haryana Agricultural University, Hisar during the year 1990-91. Hardwood cuttings were collected during January and semi-hardwood cuttings were taken during July. The results of the experiments are summarised in the following paragraphs :

1. Hardwood cuttings collected during January performed better than semi-hardwood cuttings taken during July.
2. The diameter of basal portion of hardwood cuttings taken during middle and end of January was more than the semi-hardwood cuttings taken during July.
3. Initial status of nitrogen and carbohydrates in hardwood cuttings was more than that of semi-hardwood cuttings. But semi-hardwood cuttings showed slightly higher level of phenols than hardwood cuttings.
4. After callusing, the diameter of cuttings, number of root initials and the level of carbohydrates was more in cuttings collected during middle of January than in cuttings collected late in January. But the level of nitrogen and phenols was lower in early collected



cuttings.

5. The maximum diameter of cuttings, number of roots initials, level of carbohydrates in cuttings was obtained from cuttings treated with 2000 ppm IBA but at this concentration the level of nitrogen at the base of cutting was minimum in both the timings of taking cuttings.
6. Early collected cuttings proved superior to late collected cuttings with respect to per cent sprouting, average number of leaves and sprouted buds and vigour of shoot.
7. The treatment with 2000 ppm IBA resulted in highest per cent sprouting, average number of sprouted buds and leaves and most vigorous shoots in early and late collected cuttings.
8. The per cent mortality was less in early collected cuttings in comparison to late collected cuttings. Application of even lower concentration of IBA during early planting of cuttings was as effective as 2000 ppm IBA applied during late in the season.
9. Early collected cuttings were superior to late collected cuttings with respect to average number of roots, fresh and dry root weight and length of roots.
10. Maximum number of roots and maximum fresh and dry root weights were obtained from cuttings treated with 2000 ppm IBA in both the timings of taking cuttings whereas length of roots was maximum in cuttings treated with 2000 ppm IBA in combination with 2000 ppm p-hydroxy benzoic acid.
11. Average number of roots was positively correlated with total sugars

and high C/N ratio and negligibly negative correlation was observed with total phenols. The positive effect of IBA on biochemical constituents, shoot and root growth parameters was decreased with the addition of increasing concentrations of p-hydroxy benzoic acid.

12. Rooting potential, sprouting capacity and survival ability was very low in semi-hardwood cuttings collected during July.

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APPENDIX- I

Mean monthly meteorological data for temperature and relative humidity recorded during the planting seasons of grape cv. Perlette cuttings at H.A.U. Farm observatory, Hisar.

January to December, 1990

Dates	Max. Temp. (°C)	Min. Temp. (°C)	Relative humidity (%) M/E
15 January (Date of taking cuttings)	18.3	2.5	93/34
30 January (Date of taking cuttings)	24.1	5.0	88/30
25 February (Date of Planting)	25.6	13.2	93/67
11 March (Date of Planting)	29.4	10.8	95/34
Mean of temp. of 40 days of callusing from 15 Jan. to 25 Feb.	19.0	3.5	90/30
Mean of temp. of 40 days of callusing from 30 Jan. to 11 March	25.0	6.5	82/26
15 April	26.4	10.5	90/35
15 May	42.8	22.2	54/22
15 June	42.2	26.5	55/33
15 July (Date of of taking cutting)	37.5	26.5	47/16
15 August	35.3	22.4	57/20
15 September	34.9	22.4	82/37
15 October	34.3	19.3	63/27
15 November	35.5	14.9	29/11
15 December	18.0	3.3	32/12

