

STANDARDISATION OF EPICOTYL GRAFTING IN MANGO

By

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THESIS

submitted in partial fulfilment of
the requirement for the degree

MASTER OF SCIENCE IN HORTICULTURE

Faculty of Agriculture

Kerala Agricultural University

Department of Horticulture

(Pomology & Floriculture and Landscaping)

COLLEGE OF HORTICULTURE

Vellanikkara - Trichur

1985

DECLARATION

I hereby declare that this thesis entitled "Standardisation of epicotyl grafting in mango" is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title of any other university or society.

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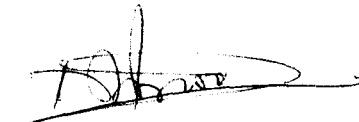


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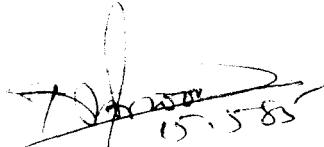


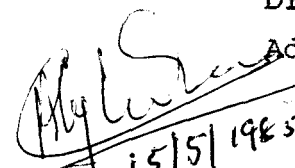
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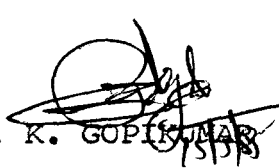
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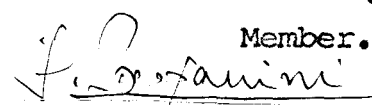
CERTIFICATE

We, the undersigned, members of the advisory committee of Mr. Jombo Ratan, a candidate for the degree of Master of Science in Horticulture agree that the thesis entitled "Standardisation of epicotyl grafting in mango" may be submitted by Mr. Jombo Ratan, in partial fulfilment of the requirement for the degree.


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ACKNOWLEDGEMENT

No word can truly represent my deep sense of gratitude to Dr. M. Aravindakshan, Chairman of my Advisory Committee and Director, Centre for Tree crops and Environmental Horticulture for suggesting a most relevant and an enchanting topic for my research programme. I am highly indebted to him for his constructive criticism flanked by constant encouragement during the course of investigation.

I am greatly indebted to Dr. Abi Cheeran, Member of my Advisory Committee and to Sri. A. Sukumara Varma, Associate Professor, for their valuable suggestions during the course of the study.

Heartfelt thanks are also due to Dr. K. Gopi Kumar, Assistant Professor, member of my Advisory Committee for his ever willing readiness for going through the manuscript timely and critically.

My sincere thanks and gratitude are also due to Professor, P.V. Prabhakaran, Member of my Advisory Committee and also to Sri. V.K.G. Unnithan, Associate Professor for the help rendered by them in statistical analysis.

I wish to express my deep sense of gratitude to Dr. K.V. Bhatt, Scientist, Division of Wood Science, Kerala Forest Research Institute, Peechi for providing facilities and extending a helping hand for the microtomy works.

I wish to record my deep sense of indebtedness to Sri. R.K.Puri, Factory Manager, Darlco Cannings Limited, Trichur for providing mango stones free of Cost and for the keen interest shown by him on fruit technology.

I express my sincere thanks also to Sri. R.P. Srivastava Agronomist and Joint Director of Agriculture, Arunachal Pradesh for his constant encouragement for pursuing master's degree course.

Many thanks are also due to the staff of the District Agricultural Office, Tawang, for their timely release and uninterrupted despatch of my pay during the course of study.

I am extremely grateful to all my friends and the staff of the Department of Pomology and Floriculture for their everwilling co-operation during the entire period of the study.

It is my proud privilege to express my gratitude to Kerala Agricultural University for providing opportunity for undergoing the masters degree programme in the College of Horticulture.

The award of fellowship by the North Eastern Council and the opportunity given by the Agriculture Department Government of Arunachal Pradesh to undergo postgraduate programme are also gratefully acknowledged.


(Jombo Ratan)

TO MY PARENTS AND FAMILY

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INTRODUCTION

INTRODUCTION

Mango is grown in an area of 51,210 ha (Anon, 1982) in this state. Every year there is a large demand for grafts of choice varieties. The traditional method of propagation of mango adopted in this state, namely, approach grafting or inarching is expensive and results in increased cost of planting materials. Cheaper methods of producing grafts have therefore to be standardised in order to reduce the cost of production of grafts.

Veneer grafting and stone grafting are adopted in other states of India on commercial scale. In the states of Maharashtra, U.P. and Gujarat these methods have virtually replaced approach grafting. Neither veneer nor stone grafting is widely adopted on a commercial scale in Kerala mainly due to lack of standardised technology.

Systematic work on standardisation of veneer and stone grafting was initiated in the Department of Pomology, College of Horticulture, in the year 1982 and it was shown for the first time that there existed possibilities of adopting stone grafting on commercial basis in our state. The season of grafting, methods of precuring, and the age of rootstock and scion

were standardised in the earlier experiments. The percentage of success in these experiments was confined to 69.33 per cent. It was, however, felt that there existed possibilities for increasing the percentage of success so as to make stone grafting more acceptable by nurserymen.

The present study is the second in the series on standardisation of cheaper methods of vegetative propagation in mango and was aimed at improving the techniques of stone grafting earlier standardised.

The main objectives of the present studies consisted of the followings:

- (i) to standardise the height of rootstock and length of scion for grafting
- (ii) to study the effect of polythene covering on success of grafts
- (iii) to find out the effect of growth regulators on graft 'take'
- (iv) to arrive at suitable control measures for the fungal infection of grafts

Anatomical studies of graft union were also taken up in order to understand the healing process and to arrive at possible reasons for graft failure.

REVIEW OF LITERATURE

REVIEW OF LITERATURE

A comprehensive review on epicotyl and veneer grafting was made by Dhungana (1984), while reporting the results of investigations on epicotyl and veneer grafting in mango carried out in the Department of Pomology and Floriculture, College of Horticulture, Kerala Agricultural University. The present review has therefore been mainly confined to the literature relevant to the studies carried out on the aspects mentioned in the earlier section.

2.1. Effect of height of rootstock and length of scion Epicotyl grafting

Traub and Auchter (1934) were the first workers to report the details of embryo grafting in mango from Florida. In their trials they used scions of 2 to 5 cm lengths for grafting on newly sprouted mango embryo.

Bhan et al. (1969) coined the term 'stone grafting' or 'epicotyl grafting' at Krishnanagar West Bengal, using newly sprouted mango stones as rootstock and semimature, terminal shoot (10 to 12 cm) as scions and obtained a success of 75 to 80 per cent.

Maity and Biswas (1980) reported from Krishnanagar

West Bengal that epicotyl grafting of mango could be best done with 3 to 4 month old scion materials which were 12 to 15 cm long. They obtained 50 to 96 per cent success during June-July and the grafted plants were ready for planting within six months. According to them defoliated scion shoots always produced higher percentage of successful grafts than undefoliated ones irrespective of cultivars.

Studies carried out in the Department of Pomology and Floriculture, College of Horticulture during 1982-1984 revealed that there was good scope for stone grafting in mango under Kerala condition (Dhungana 1984). He observed that season of operation and defoliation of scion had considerable effect on the overall success. According to him four month old, 10 to 12 cm long scions which were defoliated 10 days prior to grafting when grafted on five day old seedlings during August gave maximum percentage of success (69.33 per cent). It was also found that growth rate of scions, number of leaves, girth of stock and scion were maximum when grafting was done in August.

While comparing the stone grafting and veneer grafting in mango, he concluded that stone grafting was found to be most suitable for Kerala condition from the point of view of the percentage of success. However, time for operation of stone grafting was highly restricted. Veneer grafting had the advantage that it could be done over a longer period of two to three months without wastage of rootstocks.

Detailed studies on epicotyl grafting in cashew were carried out by Nagabhusanam (1982) and he stated that grafting should be done at a height of 5 cm above the cotyledons using cleft method during monsoon. For getting maximum success (80 per cent) the length of scion should be 5 to 6 cm.

Shylaja (1984) conducted studies on epicotyl grafting in cashew under Vellanikkara condition and observed that scion shoots defoliated 10 days before grafting and of 8.6 to 10.5 cm length gave maximum success of 82.60 per cent in August.

Gunjate et al. (1980) from Dapoli (Maharashtra) reported that terminal mature shoots of 10 cm in length were the best scion material for epicotyl grafting in jack fruit. The rootstock in polybag was beheaded

5 to 6 cm above soil surface and a vertical slit of 5 to 6 cm length was made in the centre of the epicotyl. They reported the success of grafting as 50 to 90 per cent during April to June.

Veneer grafting

Mukherjee and Majumdar (1961) obtained 76 to 96 per cent success in veneer grafting in mango with 10 to 15 days defoliated terminal scion shoots of 6 to 12 cm length under Delhi conditions.

Majumdar et al. (1972) observed that success of veneer grafting was not effected by scion length ranging from 2.5 cm to 10 cm long but subsequent growth was greater with longer scion under Calcutta condition.

Ram and Bist (1982) in a study on veneer grafting of mango in Tarai region of U.P. observed that maximum percentage of success (80 per cent) was obtained with scion length of 10 cm.

Splice grafting

Torres (1949) reported that the use of 8 to 10 cm long scion on 3 to 12 months old stock in splice grafting of mango gave better success under Philippine condition.

Dela Rocha (1953) from Lamoãina reported 90 per cent success for splice grafting in mango when grafting was done at a height of 10 cm from ground level. Scions having minimum of three buds were tied with rubber band and sealed with paraffin wax.

Side grafting

Persai (1963) while comparing the side grafting and shield budding of mango under Ehopal conditions observed that 15 cm long scion of bright dark green colour were most suitable for side grafting.

Kanwar and Bajwa (1974) tried side grafting of mango under Ludhiana conditions and found that scion of length 7.5 cm gave maximum success of 90.22 per cent. Grafting could be done successfully from March to October with defoliated scions.

Sahani (1982) while reviewing the vegetative propagation in cashew recommended that side grafting could be done on stock of one to two years old at 15 to 20 cm height from the ground level. Scions should be three to six months old and 15 cm long for getting maximum success.

Bench grafting

Patel and Amin (1976) reported that bench grafting of young mango seedlings could be done successfully at a height of 6 to 7 cm on stock with scions of 4 to 6 mm thickness under Anand condition.

Soft wood grafting

Amin (1978) reported that in mango, seven days defoliated scion shoots of length 10 to 13 cm gave 100 per cent success in soft wood grafting in mango. The method adopted was cleft and polythene tape of 1.2 cm wide, 45 cm long and 200 guage was used as tying materials.

2.2. Effect of covering materials on success of grafts

Rao and Rao (1956) from Mangalore reported that in mango, inarching, covering the graft union with polythene helped in moisture retention which in turn gave better success.

Singh (1960) recommended that in dry places, high humid condition could be created around graft union by keeping a glass jar inverted over scions. A hole in the bottom of the jar was essential. Hole was covered loosely with glass pieces so that respiratory gas could escape.

Pinheiro et al. (1970) reported that covering the graft union with white polythene film for 39 days enhanced the percentage of success. In cleft method they obtained a success of 97.1 per cent followed by whip (88.9 per cent) and tongue method (68.8 per cent) under Brazilian condition.

Teotia and Maurya (1970) in a study on chip budding of mango under Basti (U.P.) condition observed that covering of grafts with white polythene film was superior to black polythene. They concluded that high absorption of heat (radiant energy) by black polythene might have been responsible for poor response compared to white polythene.

Jauhari and Singh (1970) compared two types of wrapping materials viz., white and black polythene in mango grafting under Punjab conditions and they stated that use of white polythene as wrapping material during July-August gave better success.

Reyes (1978) while studying the effect of relative humidity on mango grafting under Panama conditions observed that covering the grafts with white polythene films and keeping them under shade gave highest percentage of success of 97 per cent.

Singh and Srivastava (1978) while investigating the effect of storage of bud stick in mango grafting observed that bud sticks stored in white polythene with moss resulted in better success than when they were stored in black polythene.

Singh and Srivastava (1982) also obtained beneficial effect when bud sticks of mango were covered with white polythene film compared to control.

Rao et al. (1957) from Mangalore obtained 70 per cent of 'take' by covering the side grafts of cashew with polythene films of 100 guage. Maximum success was obtained during February and March.

Ascenso and Milheiro (1973) obtained 100 per cent success in cleft grafting of cashew by covering the graft union with polyvinyl film. However, Rao and Putcha Radhakrishnan (1982) under Bapatla (A.P.) observed that in cashew, covering the graft unions in side grafting with polythene was not superior to control. Scions remained green for 20 to 25 days when scion was covered with polythene film. They stated that accumulation of moisture inside the graft led to rotting. When covering was punctured to prevent

excess water accumulation, still it failed.

Nagabhusanam (1982) in his studies on epicotyl grafting of cashew reported better success (60 per cent) when scion and rootstock were capped with narrow polythene bag securing it at the base with rubber band.

Calkins (1932) while top working of avocado in California obtained better success when grafted scion was covered with perforated paper bag. The holes were made when scion elongated and finally it was allowed to grow through opening. Shippy (1930) from Florida observed that in apple, side grafting at air moisture levels below the saturation point inhibited callus formation. The rate of desiccation of cells increased as humidity dropped. Hence, he concluded that humid condition was necessary for rapid proliferation of callus leading to speedy and better healing of graft union. In a study on grafting in apple, Stolbov (1966) found that covering the grafts with plastic film gave 97 per cent success compared to control. Drobota (1967) while top working in apple observed that the grafts covered with polythene films recorded maximum success of 96 per cent and it enhanced bud burst, prevented wind damage, desiccation and protected them against infection of fungus (Schiaphobus sthalidus).

Bringezu and Penning (1969) compared the effect of different coloured polythene sheets for wrapping the graft in apple and found that there was no significant difference between polythene sheets of different colours.

Tamburo et al. (1955) while top working on Guava in Florida found that scion wrapped with perforated polyvinyl film gave 73 per cent success whereas the success was only 50 per cent in unwrapped grafts. Stover (1957) in a study on cleft grafting of grapes noted the highest 'take' when grafts were covered with Sphagnum moss and then with polythene, moreover, the covered grafts required less care than uncovered grafts. Horanszky (1972) reported that vine grafts covered with polyvinyl film gave better take (60 per cent) than those uncovered with film (36 per cent).

Hinriches (1962) recommended the use of aluminium foil covered with polythene sheet to protect the pecan grafts instead of grafting wax or asphaltic tree paints.

Mcfadden (1963) observed that the two liquid emulsions like Dupontis Elvacet 81-900 and Alcas latex 5229 were excellent materials for coating scions

of curtis pecan grafts. These emulsions were found to retain moisture which gave better success.

Hensen and Hartmann (1951) obtained better success (100 per cent) in Walnut grafting by white washing the graft union during very hot weather. They stated that this was achieved because white wash reflected radiant energy which lowered the bark temperature which again was congenial for better graft union.

Fortes et al. (1970) in their studies to find out the effect of covering materials on citrus graft union observed that polythene film covered grafts gave better results.

2.3. Effect of growth regulators on graft 'take'

Gouwentok (1941) reported that in most of the plants, a period of dormancy has to be completed before the cambium could be reactivated by the application of growth regulators. Thus, application of growth substances would be effective in inducing activity if some factors which caused dormancy had been eliminated. Thompson (1945) stated that most consistent response of plants to treatment with relatively high concentration of physiologically active substances was cellular

proliferation. Similarly the degree of response of tissues to such treatments varied with species. Treatment of tissues like meristems which had already proliferated usually caused either inhibition of growth or a distortion of the normal growth pattern.

Went (1945) reported that the application of auxin to stem caused cambial activity over some distance below the point of application. Jacob (1950) stated that differentiation of xylem could be induced by external application of auxins. Mc Quilkin (1950) while working on growth regulators in the healing of tree wounds reported that exogenous application of auxin to the tree wounds or graft union gave no consistent results in healing of the wounds.

According to Wareing (1958), two main groups of regulators viz., GA and IAA influenced the cambial activity to the greatest extent in Pinus sylvestris. He showed that while GA influenced cell division it had no effect on cell differentiation. IAA on other hand stimulated rapid cell differentiation. However, simultaneous effect of the two groups of substances resulted in the appearance of normal cambial activity.

Mansour and Rivals (1970) while veneer grafting several temperate and sub tropical fruit trees observed

that good results were obtained by treating the cut surfaces of stock and scion with 3000 ppm IBA in 50 per cent alcohol followed by stratification in vermiculite at 25 to 27°C.

Wareing and Phillips (1978) observed that rapid multiplication of cambial tissues in xylem and phloem vessels could be induced through the application of IAA along with sucrose. Cambial divisions and differentiation of some new xylem element could be obtained, if GA was applied alone. When IAA and GA were applied together cambial divisions and normal differentiation of xylem and phloem tissues were observed.

Hartmann and Kester (1978) concluded that external application of sugar at 2.5 to 3.5 per cent with IAA or NAA each at 0.5 mg/litre induced xylem and phloem in the callus with cambium in between in most of the crops they studied.

Krishnamoorthy (1981) opined that since auxin promoted cell division of cambium, dipping of stock and scion in auxin solution resulted in early and better graft union.

Mango

Kannan and Rao (1964) while investigating the effect of GA on graft 'take' in mango reported that GA at a concentration of 1000 ppm applied at the time of approach grafting as well as pretreatment of stock and scion resulted earlier separation of grafts (45 days) as compared to control (75 days). There was good callus formation after 15 days of the treatment.

Chaudhury and Basu (1972) stated that the concentration of anti-~~auxin~~ins or inhibitors which inhibited the growth of mango was more in the apex of large leaf and thus the removal of upper half of these apex of leaf resulted better graft union.

Avocado

Samish and Gur (1962) reported that bud take of avocado was improved by the application of IAA at 25 ppm especially when grafting was done on older stock. Dela Rocha et al. (1966) reported that treating the grafted plants with GA at 10, 25, 50 ppm reduced the height of scion growth significantly compared to control. Boswell and Bergh (1979) observed that when NAA was applied at 0.5 per cent concentration prior to growth of the graft, the seedlings died within four months.

Vine

Kordes (1937) observed that percentage of 'take' in vine grafting was more when IBA was applied at 0.01 per cent on both stock and scion. Evanari and Konis (1938) reported that in whip grafts with IAA treatment of the cut surface of stock with IAA resulted in better callus formation and thus shortened the time required for good union.

Dragas and Avramov (1952) obtained beneficial effects with IAA and NAA in the callus formation resulting good graft union in vine grafting.

Stepanova (1969) while investigating the effect of growth stimulant and vitamins on callus formation in vine grafting observed that better graft union was possible with IAA at 0.01 per cent, IBA at 0.005 per cent, NAA at 0.006 per cent, Vit.C at 250 mg/lit.

Guava

Rao and Kaul (1977) while studying the effect of kinetin on veneer grafting of guava in U.P. observed that dipping the scions in kinetin at 150 ppm for 30 minutes was detrimental for graft 'take'.

Citrus

Maity et al. (1959) studied the effect of plant regulators on bud 'take' in grape fruit and found that dipping the bud wood in solution of IBA at 1000 ppm and subsequently in 50 per cent alcohol gave better 'take' than IAA. Wong (1966) observed 95 per cent success in crown veneer grafting of citrus by the application of NAA at 100 ppm during October and IBA 200 ppm during June.

Apple

Molotovskii and Poruckii (1951) observed that extract from scion leaves applied to the cut portion of rootstocks of fruit trees like apple, pear, plum and magnolia resulted in a quicker and slightly better grafts union than growth substances like IAA and nicotinic acid. However, application of growth substance like IAA and nicotinic acid was better than water treatment.

Padfield (1952) obtained good 'take' in apple grafting through the application of bituminous group of compounds. He stated that these compounds maintained a durable permanent cover over the cut surfaces and produced most vigorous scions with no damage to the tissue. The addition of hormone increased callus formation.

Savin (1973) in a study on winter grafting of apple reported that all the growth regulators like IAA, IBA, NAA, MCPA, 24-D and micro elements like Zn, Mo, Mn, B had very little effect on graft 'take' but they stimulated further growth of grafted trees in the mainfield.

Walnut

Hensen and Hartmann (1951) found that external application of auxin did not affect the graft take in walnut. Brierleg (1953) obtained good success (71 per cent) with walnut root grafting with the application of hormodin No.2 powder in tip of scion bud as well as at graft union.

Chestnut

Veitez and Veitez (1983) reported that application of 2 to 10 ppm of IBA enhanced the callus tissue development and thus resulted better graft 'take' in chestnut.

Pear/Quince

Brian and Duron, (1972) in their incompatibility trials in pear and quince grafting observed that

application of IAA at 100 mg/1 + GA at 500 mg/1 to the base of the scion before grafting improved the union but IAA at 500 mg/1 + GA at 100 mg/1 had no such effects.

Stankovic et al. (1963) reported that a mixture of NAA + B + Zn + Fe + Mn + Cu added in grafting wax promoted healing of wounds in apricot, peach and when the growth regulators were applied in apple and pear, the shoot growth was promoted.

2.4. Fungal diseases associated with grafts/trees

Mango

West (1934) reported that Colletotrichum gloeosporioides which attacked tender portions of mango shoot could be easily controlled by the application of Bordeaux mixture at 1 per cent.

Vaheeduddin (1954) from Hyderabad reported that dieback of mango was caused by Fusarium species.

Rath and Mohan (1978) from Orissa reported that dieback of mango was exhibited as withering of tip, twig blight and bark cankers. They reported that this was caused by Botryodiplodia, Rhizoctonia, Colletotrichum, and Sclerotium species.

Singh and Srivastava (1978) reported that fungal disease of mango grafts could be controlled by the application of Blitox, or Benlate at the concentration of 0.2 per cent in scion at the time of veneer grafting of mango under U.P. condition.

Pathak (1980) suggested various precautionary measures such as collection of healthy scion, sterilization of grafting knives, placement of budded trees in dry environment and gradual exposure to full sunlight for the control of dieback disease of mango grafts. Maity and Biswas (1980) while doing epicotyl grafting of mango in Krishnanagar, West Bengal reported that fungal infection could be controlled by dipping scions and seedlings in 0.2 per cent Captan solution for few minutes before grafting.

Kanwar and Jawanda (1983) while topworking mango trees through side grafting used Bordeaux mixture paste as protective measures against fungal infection.

Avocado

Pinheiro and Pinheiro (1971) reported the treatment of avocado scion with captan at the concentration of 3 g/bag and stored in ambient temperature.

Vine grafting

Becker (1966) recommended treatments of vine rootstock and scion with solution of 0.5 per cent Chinosol for at least 12 hours before grafting.

Eifert et al. (1970) observed that infection during callusing the vine grafts in boxes could be restricted by treating scion and stock wood with 0.5 per cent chinosol and it was also found to promote callus formation in the cut portion.

Neider (1972) observed that Botrytis infection in vine grafts could be controlled by the application of 0.5 per cent chinosol in stock and scion. However, it retarded early growth of grafts but a slight lower concentration rate gave good control as well as better growth.

Chestnut

Jaynes and Hildreth (1965) reported the treatments of scions and nuts in 0.06 per cent 8-hydroxy quinoline sulphate solution as a control measures for fungal infection in seed grafting of chestnut. Jaynes and Messner (1968) observed that treatment of chestnut grafts with heavy suspension of Thiram reduced survival

of grafts by 22 per cent. They found that a mixture of IBA, NAA and Thiram at 0.6 per cent applied at graft union slightly increased their survival.

2.5. Anatomy of graft union

Robert (1949) stated that the most important factors determining the success in grafting was not the nature of the union but the genetically determined incompatibility which brought about all antagonistic interaction between stock and scion.

Bloch (1962) while studying the wound healing in trees stated that healing of graft union could be considered as the healing of wound. Ciz (1969) in a study to evaluate the compatibility of graft union suggested that incompatibility could be diagnosed through anatomical studies after 2 to 3 months of grafting.

Esau (1979) conducted detailed studies on graft union in tree crops and concluded that secondary growth and cambial activity were involved in wound healing resulting in proper union of stock and scion. The establishment of graft union was similar to those processes associated with wound healing. Breakdown products of dead cells on the surface of stock and scion formed a necrotic layer. Intact cells just next to the necrotic layers enlarged, divided and

formed callus tissue which filled the space left between the stock and scion. Eventually the cambia of the two partners became continuous across the callus by a change of callus cells into cambial cells which formed vascular tissues. The callus also arose from phloem ray and immature ray cells.

Fahn (1982) reported that important function of cambium was to form callus in the wound portion. These tissues consisted of a mass of soft parenchyma tissue which rapidly formed on or below the damaged surface. The outer cells of parenchymatous masses became suberized. Below this protective layers, a reorganised cambium produced new vascular tissues. He further stated that union of stock and scion was not only through the cambia but also through wood rays. They also proliferated and took part in union.

Mango

Walter (1932) reported that mango bud was unique in its method of union with stock plant. In his studies he observed that cohesion took place only under the bark not at the sides. The bud was thus found

to be carried upward on a cushion of rapidly dividing cambium cells.

Luthra and Sharma (1946) studied the conductivity and histology of grafted mango shoots in Langra variety. They observed that excessive growth of parenchymatous tissue (callus) between stock and scion and distorting of xylem elements, blocked the conducting vessels. According to them these features seriously hindered lateral transfer of water from stock to scion. Symptoms were less apparent when cut was made only $\frac{1}{2}$ " longitudinal depth.

Anatomical studies showed that mango stem consisted of an outer cuticle and inner epidermis, cortex, endodermis, pericycle, arched shaped fibre, resin canal, phloem, cambium, xylem, unisiriatic medullary rays, bisiriatic medullary rays, pith cell full of granules (Singh, 1969).

Torres (1960) stressed the essentiality of cambial contact in mango splice graft. Shimoya et al. (1970) from anatomical studies of wedge grafting of mango reported presence of fungal mycelium in graft union which appeared to facilitate graft union.

Soule (1971) conducted detailed studies on the anatomy of bud union in mango by chip budding method. He observed the following five important stages in formation of bud union under Florida conditions viz., stage 1 (4 days) wound periderm formation, stage 2 (8 days) callus proliferation from cambium resulted firm attachment of the component, stage 3 (12 days) completion of cambial bridge, stage 4 (36-45 days) differentiation of vascular tissues and completion of healing union, stage 5 (6-8 months) several cylinders of new tissues developed, lateral shifting of scion to align with stock.

Dave and Rao (1982) studied the cambial activity in Mangifera indica and found that radial growth of the tree was continuous as cambium was active all the year round. Tangential divisions in the cambial zone resulted in differentiation of vascular elements. Climatic factors showed no relationship with the cambial activity.

Avocado

Hellatou et al. (1977) observed the nature of union of cleft grafting ⁱⁿ young avocado seedlings and reported that callus growth began within five days and

the union was completed within 11 days. Callus in the secondary cortex was the first to react to the mechanical injury caused by grafting and it established an incipient callus bridge between stock and scion followed by cells in the cambial region and parenchymatous cells in the scion. Scions began to grow in 21 days and attained 15 cm length by 40th day.

Citrus

Mendel (1936) carried out anatomical studies in the bud union in citrus. The six stages described in the healing of citrus T - budding were stage 1 (24 hours) cell division, stage 2 (5 days) callus bridge, stage 3 (10 days) differentiation in the callus of bark flaps and callus of shield, stage 4 (15 days) occurrence of xylem tracheids in the callus of bark flaps and callus of shield, stage 5 (25-30 days) lignification of callus completed in the bark flaps, stage 6 (30-40 days) lignification of callus completed in the bark flaps and in the bark shield.

Callus formation began in all the tissues adjacent to wound. Lignification occurred gradually in the course of differentiation.

Apple

Sass (1933) studied the histology of tongue graft union of apple and found that callus were produced exclusively from the tissue outside the xylem cylinder where grafts were well matched.

Jawanda (1968) in his anatomical studies on incompatibility in apple using crab apple as root stock observed that deposition of parenchyma at the union and distortion of vascular tissues were common features in some of ^{the} incompatibility of apple grafting.

Robitaille and Carlson (1970) reported that compatibility or incompatibility due to anatomical abnormalities in certain species of Malus and Prunus appeared to be the results of secondary influence. They observed that biochemical factors controlled the compatibility or incompatibility of graft union.

Pear

Histochemical tests in union of pear quinch grafts revealed that the lignification of cell walls were mainly responsible for the formation of strong graft union (Buchloh, 1960).

Copes (1969) conducted detailed studies of graft union formation in douglasfir where following four stages were differentiated, stage 1 (2 days) formation of contact layers, stage 2 (10-14 days) callus bridge, stage 3 (10-17 days) periderm formation, stage 4 (17-23 days) cambial layers. Callus bridge had originated from secondary phloem or cortex and the callus lignification began along the cut edges of the union after 19 days and completed across the entire length of union by another 17 days.

MATERIALS AND METHODS

MATERIALS AND METHODS

The studies reported in this thesis is in continuation of the earlier studies conducted in the Department of Pomology and Floriculture during May 1982 to June 1984, with the objective of standardising economic methods of vegetative propagation in mango. Dhungana (1984) had conducted studies on epicotyl and veneer grafting and had concluded that epicotyl grafting was most suitable under Kerala conditions. The season of grafting and the precuring treatments were standardised in the earlier studies.

However, it appeared necessary to improve the graft 'take' and final survival of epicotyl grafting so as to make this method commercially sound.

The present studies on epicotyl grafting were carried out during the period June 1983 to March 1985 with the following objective:

- i) To standardise the height of rootstock and length of scion
- ii) To study the effect of polythene covering on success
- iii) To find out the effect of growth regulators on graft 'take'

- iv) To arrive at a suitable control measure for the infection of grafts.

Detailed anatomical studies of the graft union were also taken up in order to understand the nature of healing of the graft union as well as to find out possible reasons for the failure of certain grafts.

The details of the experiments conducted and the methods adopted are given below:

3.1. Standardisation of height of rootstock and length of scion

These studies were conducted from June to November 1983. The grafting was done in July and August. The scion materials of three different length viz., 5 cm, 6 cm and 8 cm from Neelum variety were used for grafting. The grafting was done on rootstocks of different heights viz., 2-4 cm, 4-6 cm, 6-8 cm and 8-10 cm.

Thus there were altogether 12 treatment combination of three types of scion and four classes of height of rootstock.

Experimental design	-	Completely randomised design
Number of replications	-	3

3.1.1. Raising of rootstocks

Well developed, uniform and healthy stones were collected from Darlco Canning Factory, Trichur as well as from around Vellanikkara and they were sown on raised seed beds of 4m X 1m in partial shade in coconut garden. The stones were sown in lines at a spacing of 2.5 cm X 10 cm. A thin layer of sand was spread evenly on the surface of seed bed. First sowing was done on 10th June 1983 and 1984. Subsequent sowings were made at 15 days intervals upto 20th July 1983 and 1984.

Stones were removed from the nursery beds immediately after sprouting with sharp pointed wood sticks taking utmost care not to injure the taproots (Plate I). Those sprouted seedlings were then transplanted in polythene bags of size 20 cm x 15 cm filled with potting mixture consisting of F.Y.M., sand and soil in 2:1:1.

3.1.2. Selection and preparation of scion sticks

Terminal mature, 3 to 4 months old shoots having dormant apical buds (Plate II) were selected

from disease free healthy mother trees, Neelum variety. Defoliation was done by clipping the portions of petiole leaving a stub behind ten days ahead of the time of grafting (Plate III) as adopted by Dhungana (1984). Selected scions were cut back at appropriate length of 5 cm, 6 cm and 8 cm.

3.1.3. Preparation of rootstock and grafting

The grafting was done when the seedlings were established in poly bags within five to ten days. A transverse cut was made in the epicotyl so as to obtain the different classes of rootstocks viz., 2 to 4, 4 to 6, 6 to 8 and 8 to 10 cm heights, then a vertical downward slit (cleft) of 3 to 4 cm length (Plate IV) was made in the centre of the stock with clean sharp knife. A slanting cut of 3 to 4 cm length was also made at both side of scion (Plate V).

The wedge shaped scions were then inserted into the slits made on the seedlings (Plate VI). The graft joint was then firmly tied with transparent polythene strips of 1.5 cm width, 30 to 40 cm length and 200 guage thickness (Plate VII).

3.1.4. After care of grafts

During the course of graft union, wherever polyembryonic seeds produced additional seedlings

they were removed. So also the rootstock sprouts occasionally noticed were removed.

3.1.5. Observations

5.a. Percentage of sprouting and survival

The observations on days taken for sprouting of scions after grafting were recorded. The scions that remained green whether sprouted or unsprouted, 15 days after grafting was accounted as the initial success. Those scions actually sprouted (Plate VIII) and survived after three months (Plate IX) were accounted as final success as adopted by Dhungana (1984).

The following growth parameters were recorded at fortnightly intervals for a period of three months (from August to November 1983).

5.b. Extension growth of scion

The extension growth of scion was measured from the point where the scion putforth new growth.

5.c. Number of leaves

The number of leaves produced was recorded at fortnightly intervals.

5.d. Girth of stock and scion

A fixed circular mark with black paint was made both on stock and scion at one cm below and above graft union. The girth of stock and scion at these joints was recorded at fortnightly intervals.

3.2. Effect of covering materials on graft 'take'

These studies were conducted from June to November 1984. The treatments comprised of the following:

- 1) Covering the whole graft with transparent polythene bags
- ii) Covering the whole grafts with black polythene bags
- iii) Control (without covering)

The total number of seedlings grafted under each treatment was 480. The grafting was done at a height of 6 to 8 cm by using scions of 8 cm as this combination was found to give maximum success in previous studies:

Number of treatments	:	3
Design	:	Completely randomised design
Replications	:	5

3.2.1. Preparation of stock and scion

The method for raising rootstock and selection of scions was as same as under experiment 1 (3.1.1. and 3.1.2 sections). The grafts were covered by 200 guage transparent or black polythene bags (20 x 15 cm) as the case may be by inserting the bags over the grafts leaving sufficient space at the top of the grafts for facilitating the growth of the scion. The bags were then tied loosely at the base with a rubber band (Plate X). The bags were retained on the grafts till 30 days after grafting.

3.2.2. Observations

The observations on the percentage of sprouting and survival of grafts, extension growth of scion and rootstock growth and number of leaves produced were recorded as in previous experiment.

3.3. Effect of different growth regulators on success of grafts

Two growth regulators viz., IAA and GA each at concentrations of 100 and 250 ppm were used for this

study. Grafting was done during July-August 1984. Fifty grafts were prepared under each treatment and each treatment was replicated four times. The details of the treatments are given below:

Treatments

T1	IAA 100 ppm
T2	IAA 250 ppm
T3	GA 100 ppm
T4	GA 250 ppm
T5	Control (water spray)

Total number of treatments	:	5
Experimental design	:	Completely randomised design
Number of replications	:	4

3.3.1. Raising of rootstocks, selection of scions, scion preparation, grafting operation and observations taken were similar as earlier experiments.

3.4. Effect of fungicides on final survival of grafts

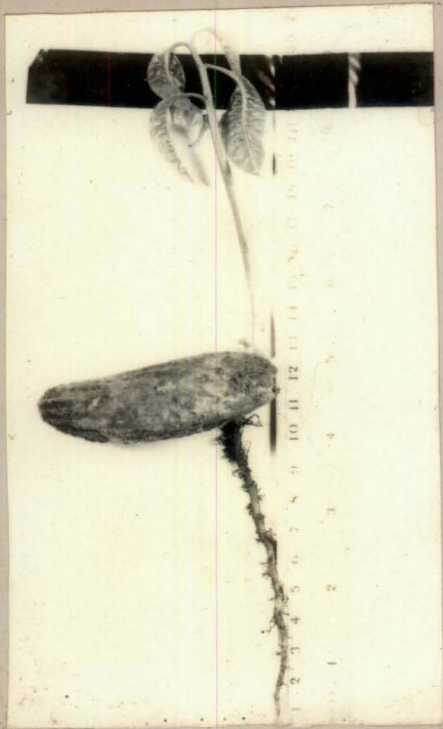
For this study, three fungicides viz., Bordeaux mixture at 1 per cent, Thiram at 0.2 per cent and Caftafol at 0.2 per cent were used. Grafting operation

Plate I. Five days old seedlings for epicotyl grafting

Plate II. Terminal matured, 3-4 months old scion twig

Plate III. Defoliation of scion shoots with intact petioles

Plate IV. Preparation of stock at the height of 6 cm for
epicotyl grafting



I



II



III



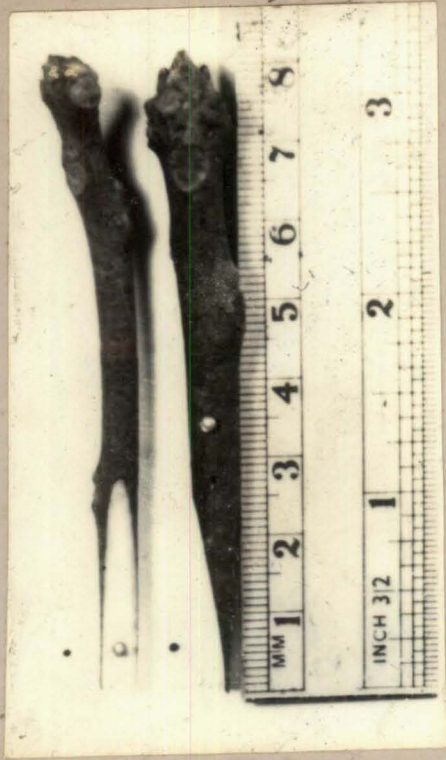
IV

Plate V. Preparation of scion for epicotyl grafting.

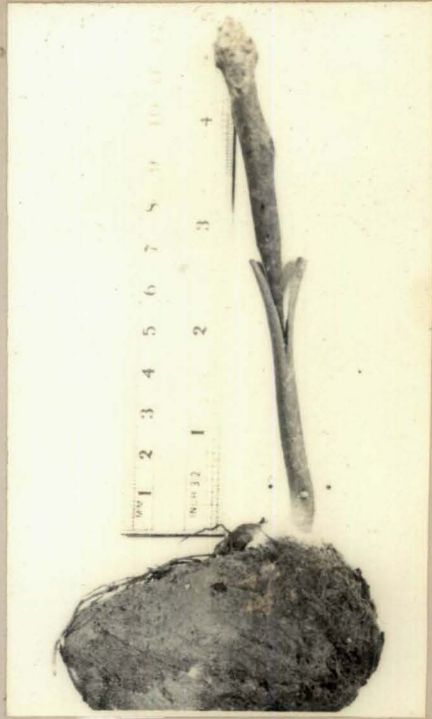
Plate VI. Insertion of scion into stock

Plate VII. Tying of graft joint with polythene strip

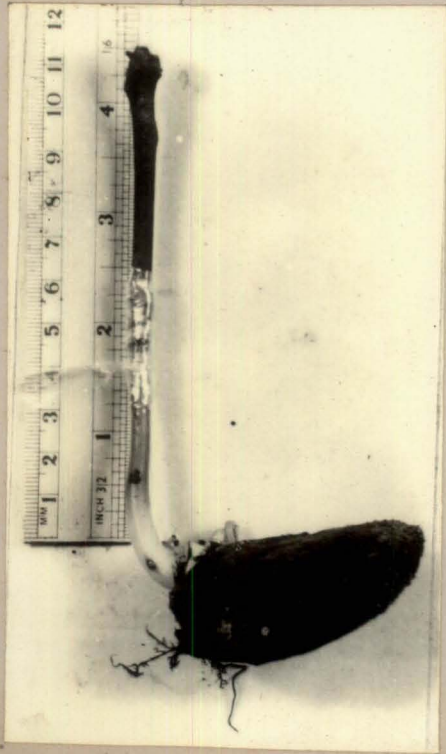
Plate VIII. Just sprouted grafts after 15 days of grafting



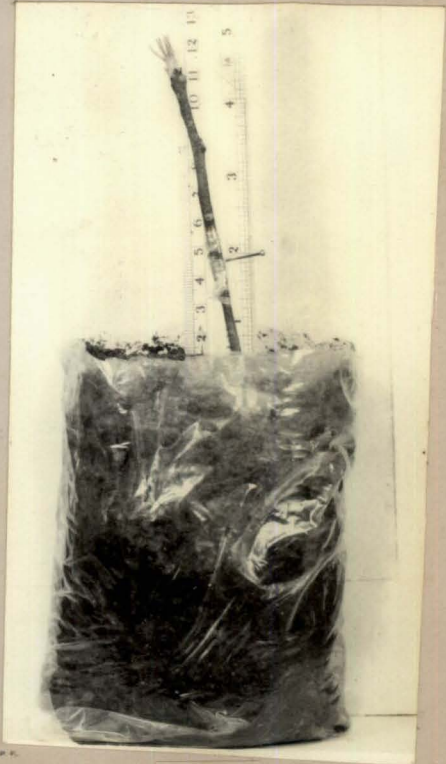
V



VI



VII



VIII

Plate IX. A successful graft after 90 days

Plate X. Covering of grafts with transparent plythene bag



IX



X

was done during August 1984. Fifty grafts were prepared under each treatment and each treatment was replicated four times.

Treatments

T1	Bordeaux mixture 1 per cent
T2	Thiram 0.2 per cent
T3	Caftafol 0.2 per cent
T4	Water spray

Total number of treatments	:	4
Design	:	Completely randomised design
Number of replications	:	4

3.4.1. Observations

The scions that remained green after 30 days of grafting were accounted as initial success as suggested by Patel and Amin (1976).

The first application of the above fungicides was made one month after grafting and subsequently at an interval of 15 days till 90 days. Observations were recorded as in previous experiments.

3.5. Anatomical studies of graft union

Grafts were separately prepared for anatomical studies in August 1984. Samples were collected for

subsequent anatomical studies as detailed below:

3.5.1. Collection and storage of specimens

Representative samples from the graft union were collected at intervals of 24 hrs 5 days, 15 days, 45 days and 90 days after grafting. Samples were also taken from those grafts which showed sign of shrinking or drying from 5th day onwards. In certain cases the scions did not sprout but remained green even after 60 days. Samples from such materials were also taken. The collected samples were then processed immediately as per the following procedure.

3.5.2. Processing

FAA (850 ml of 70 per cent alcohol + 100ml of 40 per cent formaldehyde + 50 ml glacial acetic acid) solution was used for killing as well as fixing. Specimens were kept in FAA solution for minimum period of 72 hours. They were then removed by using a sterilized forceps and washed in running water for $\frac{1}{2}$ hours and later with glass distilled water for further sectioning.

Uniform thin section were taken at a thickness of 25 μ (micron) using "Reichert sliding microtome" as per standard microtomy for hardwood (Cutler, 1978).

The detailed schedule followed for clearing and staining of sections are furnished below:

sections
 ↓
 50% alcohol (2 minutes)
 ↓
 distilled water (2 minutes)
 ↓
 sodium hypochlorite solution (5 minutes)
 ↓
 safranin (2 hrs)
 ↓
 50% Alcohol + 2 to 3 drops of conc. Hcl (2 minutes)
 ↓
 xylene (5 minutes)
 ↓
 mounted on slides with Canada balsam
 ↓
 ** microscopic examination
 ↓
 ** photomicrography

** Microscopic examination

The slides were examined through "olympus binocular research microscope" with 10X/5X objective and 15X eyepiece.

** Photomicrography

Photomicrograph of selected sections were taken using

a photomicrography system (Olympus PM-6) ORWO black and white negative film of 125 ASA was used for taking photomicrograph of the selected sections.

3.6. Statistical analysis

Significant differences among the treatments with regard to the number of sprouting and survival of grafts were detected by employing the chi-square test as described in Panse and Sukhatme (1978).

The test criterion is given below

$$\chi^2 = \frac{1}{n_1 n_2} \sum \frac{(an_2 - a'n_1)^2}{a+a'}$$

Where χ^2 = Chi - square

a = number of grafts sprouted or survived for each treatment

a' = number of grafts unsprouted or not survived for each treatment

n₁ = number of grafts sprouted or survived for all the treatments

n₂ = number of grafts unsprouted or not survived for all the treatments.

The degrees of freedom for chi-square is (K - 1) where K is the number of treatments.

Pairwise comparison of treatments were made using the chi-square test of independence (after applying Yate's correction for continuity).

The relevant test criterion is given by

$$\chi^2 = \frac{(|ad - bc| - n/2)^2 n}{(a+b)(a+c)(b+d)(c+d)}$$

Where n = a+b+c+d

a and c = number of grafts sprouted or survived in the two treatment

b and d = number of grafts unsprouted or not survived in the two treatment with one degree of freedom.

Growth parameters

Effect of the treatments on the difference with respect to quantitative characters viz., extension growth of scion, number of leaves, girth of stock, girth of scion was tested using standard methods of analysis of variance. Wherever treatments effects were found to be significant pairwise comparisons were made by calculating the critical difference (C.D) using the standard formula.

RESULTS

RESULTS

The present investigations were carried out in the Department of Pomology and Floriculture, College of Horticulture during the years 1983-85 with an objective to standardise the suitable height of rootstock, length of scion, effect of polythene covering and growth regulators on graft 'take' in epicotyl grafting of mango. The results of the studies are summarised below:

4.1.a. Effect of height of rootstock and length of scion on sprouting and final survival of grafts

The observations on the percentage of sprouting and final survival of epicotyl grafts are presented in Table 1. The results indicated that the length of scion and height of rootstock had profound effect on initial success as well as final survival of the grafts. The highest percentage of success of 87.50 per cent and survival of 72.5 per cent were obtained when scions of 8 cm length were grafted on rootstock at 6 to 8 cm height (Fig.1.).

The pairwise comparison by chi-square test also showed the superiority of this treatment over others (Appendix I). The minimum percentage of sprouting

Table 1. Effect of height of rootstock and length of scion on sprouting and survival of grafts

Height of rootstock	Length of scions	Total number of grafts	Sprouting		Survival	
			No.	Percentage	No.	Percentage
2-4	5	120	18	15	9	7.50
	6	120	25	20.83	17	14.16
	8	120	51	42.50	43	35.83
4-6	5	120	48	40.00	37	30.83
	6	120	56	46.66	46	40.00
	8	120	91	75.83	85	70.83
6-8	5	120	55	45.83	47	39.16
	6	120	62	51.66	55	45.83
	8	120	105	87.50	95	72.50
8-10	5	120	25	20.83	12	10.00
	6	120	31	25.83	19	15.83
	8	120	56	46.66	41	34.16

Value of chi-square

61.57**

294.05**

** Significant at 1% level.

Table 2. Effect of height of rootstock on sprouting and survival of grafts

Height of rootstock in cm	Total number of grafts	Sprouting percentage	Survival percentage
2-4	360	26.11	19.16
4-6	360	56.16	47.22
6-8	360	61.66	54.72
8-10	360	31.11	20.00

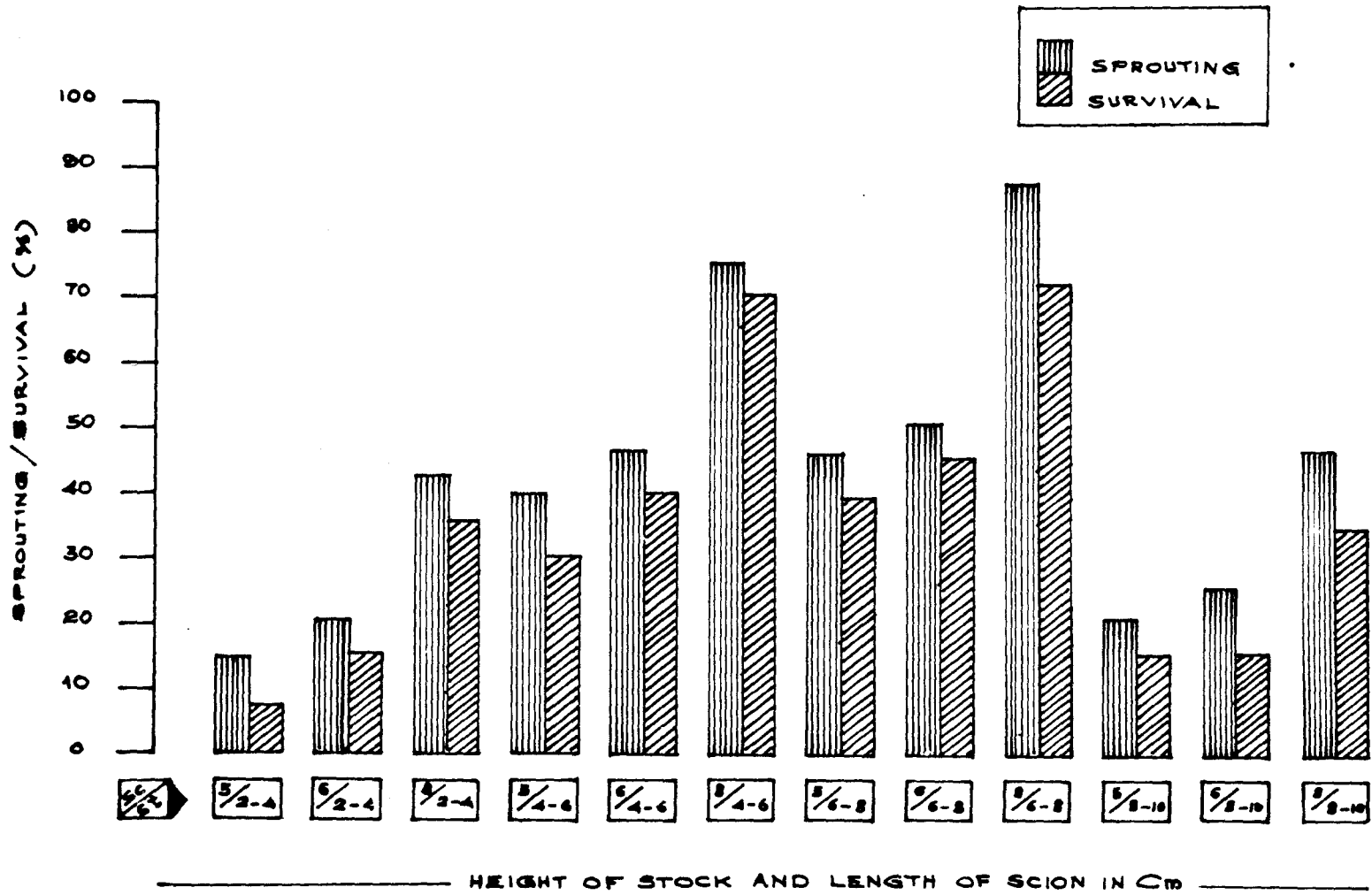
Value of chi-square

126.45**

127.93**

** Significant at 1% level.

FIG. 1. EFFECT OF HEIGHT OF STOCK AND LENGTH OF SCION ON SPROUTING AND SURVIVAL OF GRAFTS



(15 per cent) and survival of 7.5 per cent was recorded when scions of 5 cm length were grafted on rootstock at 2 to 4 cm height.

The results of the statistical analysis of the aggregate data on the effect of four classes of rootstock on percentage of success pooled over the length of scion are given in Table 2. It will be seen from the Table that the height of rootstock affected significantly the percentage of sprouting and survival of grafts. The highest percentage of sprouting of 61.66 per cent and survival of 54.72 per cent was observed for rootstock of 6 to 8 cm height while the minimum sprouting (26.11 per cent) and survival of 19.16 per cent was recorded for rootstock of 2 to 4 cm height (Appendix III). The difference in sprouting and survival was not significant when grafting was done on rootstock at a height of 2 to 4 cm and 8 to 10 cm, It is also evident that rootstock at 4 to 6 cm height ranked next to 6 to 8 cm height with respect to sprouting (56.16 per cent) and survival of 47.22 per cent (Fig.2.).

The results of the statistical analysis of the aggregate data on the effect of scion length on percentage of sprouting pooled over the four classes of

rootstock are presented in Table 3. It will be seen that the scion of 8 cm gave maximum success and survival of 63.12 per cent and 55.00 per cent respectively. Pairwise comparisons for the treatments also revealed the significant superiority of scion length of 8 cm over the other length of scion (Fig.3.).

4.1.b. Extension growth of scion

The observations on the extension growth of scion at monthly intervals in the different rootstock scion combinations are furnished in Table 4.

Analysis of variance of the data revealed that there was no significant difference between the treatment combinations with regard to growth of scion during 30, 60 and 90 days of observations. However, it was observed that scions of 6 cm length grafted on rootstock of 4 to 6 cm height gave maximum extension growth of scion throughout the period of study for 3 months.

The results of the analysis of the aggregate data on the effect of different height of rootstock on extension growth of scion pooled over the different length of scion are presented in Table 5. The results of the statistical analysis indicated that the

Table 3. Effect of lengths of scion on sprouting and survival of grafts

Length of scions in cm	Total number of grafts	Sprouting percentage	Survival percentage
5	480	30.41	21.87
6	480	36.25	28.95
8	480	63.12	55.00
Value of chi-square		126.45**	127.93**
** Significant at 1% level.			

Table 4. Effect of height of rootstock and length of scions on growth of scion at 30 days intervals

Height of rootstocks in cm	Length of scion in cm	Mean extension growth of scion in cm		
		30 days	60 days	90 days
2-4	5	6.55	8.24	9.01
	6	7.33	8.33	9.65
	8	6.61	7.33	8.68
4-6	5	6.85	7.63	8.26
	6	9.31	9.63	11.16
	8	7.51	8.43	8.68
6-8	5	10.16	10.66	10.83
	6	9.91	10.33	10.33
	8	7.16	8.13	9.01
8-10	5	8.98	10.21	10.55
	6	6.46	7.53	7.75
	8	8.86	8.86	9.11
F test		NS	NS	NS
C.D.		-	-	-
SEm ±		0.63	0.58	0.55

FIG. 2. EFFECT OF SCION LENGTH ON SPROUTING AND SURVIVAL OF GRAFTS

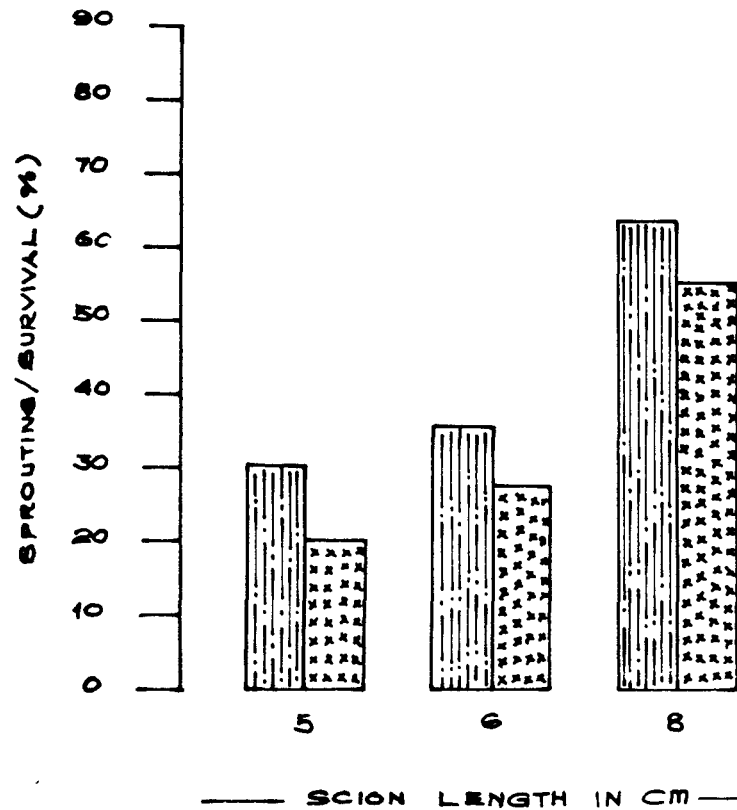
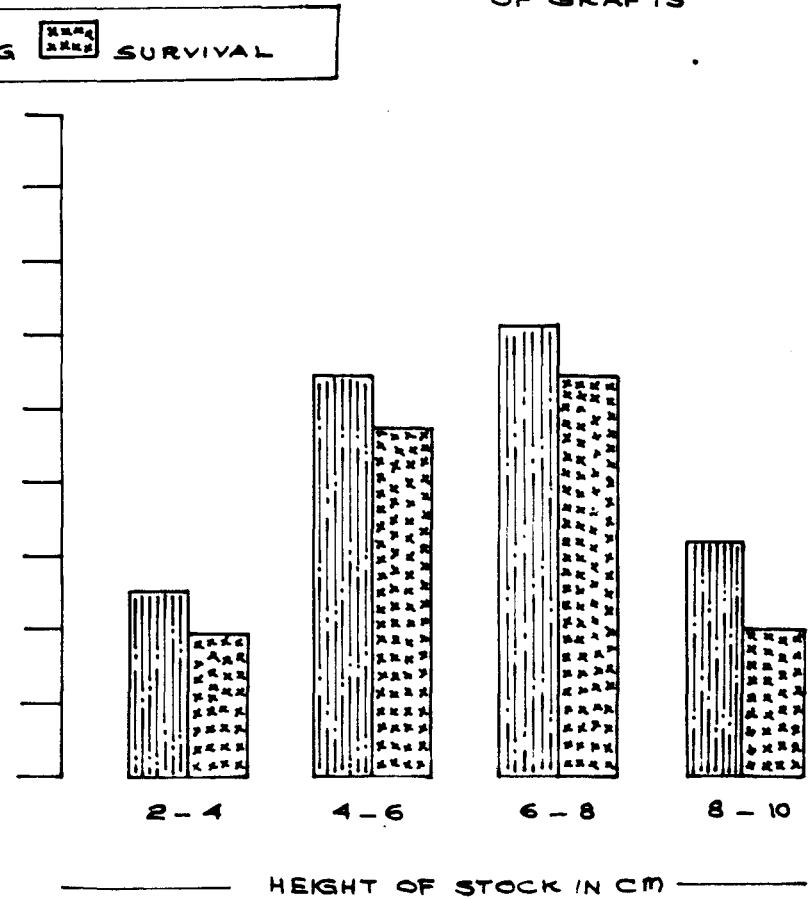


FIG. 3. EFFECT OF STOCK HEIGHT ON SPROUTING AND SURVIVAL OF GRAFTS



treatments were on par with regard to the extension growth of scion.

The results of the three types of scion length with respect to extension growth of scion are furnished in Table 6. The statistical analysis showed no significant difference between different treatments with regard to extension growth of scion.

4.1.c. Number of leaves

The data furnished in Table 7 show the effect of different heights of stock and scion length on number of leaves produced. The statistical analysis indicated that the difference in the number of leaves produced in various treatments was not significant.

The results of aggregate data on the effect of height of stock on number of leaves produced pooled over the different length of scion are tabulated in Table 8. Here also the difference between different treatments was not statistically significant. However, rootstock of 8 to 10 cm height showed a tendency for producing maximum number of leaves throughout the entire period of study.

The results of the analysis of the aggregate data on the effect of scion length on number of leaves

Table 5. Effect of height of rootstock on growth of scion at 30 days intervals

Height of rootstock in cm	Mean extension growth of scion in cm		
	30 days	60 days	90 days
2-4	6.83	7.97	9.11
4-6	7.89	8.56	8.37
6-8	9.08	9.71	10.06
8-10	8.10	8.87	9.13
F test	NS	NS	NS
C.D.	-	-	-
SEm \pm	0.67	0.70	0.90

Table 6. Effect of length of scion on extension growth of scion at 30 days intervals

Length of scion in cm	Mean extension growth of scion in cm		
	30 days	60 days	90 days
5	8.13	9.19	9.67
6	8.25	8.95	8.97
8	7.54	8.19	8.87
F test	NS	NS	NS
C.D.	-	-	-
SEm \pm	0.58	0.61	0.78

Table 7. Effect of height of rootstock and length of scion on number of leaves at 30 days intervals

Height of rootstock in cm	Length of scion in cm	Mean number of leaves		
		30 days	60 days	90 days
2-4	5	8.13	9.33	9.33
	6	7.14	10.33	11.83
	8	11.66	12.83	13.66
4-6	5	7.16	7.16	7.66
	6	11.50	12.33	14.83
	8	11.66	13.83	15.00
6-8	5	7.33	12.83	13.50
	6	11.33	11.33	11.33
	8	12.00	14.66	17.66
8-10	5	11.16	15.83	18.16
	6	11.26	13.50	14.83
	8	14.00	14.83	14.83
	F test	NS	NS	NS
	C.D.	-	-	-
	SEm \pm	0.66	0.72	0.75

Table 8. Effect of height of rootstock on number of leaves at 30 days intervals

Height of rootstock in cm	Mean number of leaves			
	30 days	60 days	90 days	
2-4	8.87	10.83	11.61	
4-6	10.11	11.11	12.50	
6-8	12.05	12.94	14.16	
8-10	12.36	14.72	15.94	
	F test	NS	NS	NS
	C.D.	-	-	-
	SE m \pm	0.63	0.85	1.03



produced pooled over the different height of rootstocks are given in Table 9. The statistical analysis for the treatment difference showed that the length of scion did not effect the number of leaves produced although 8 cm length of scion produced slightly more number of leaves at the close observations (15.29 cm).

4.1.d. Girth of stock

The girth increment of stocks in different stock scion combinations are presented in Table 10. It was observed that there was a uniform trend in the increase in girth of stocks when 8 cm scion length was grafted on rootstock of 6 to 8 cm height though the effect was not statistically significant.

The results of the analysis of the aggregate data on the effect of rootstock height on girth of stock pooled over the different length of scion are presented in Table 11. Analysis of variance of the data revealed that the stock girth is free from the effect of rootstock height.

Table 12 presents the results of analysis of the aggregate data on the effect of length of scion on girth of stock pooled over the height of different rootstock at 30, 60, 90 days intervals. The difference

Table 9. Effect of length of scion on number of leaves at 30 days intervals

Length of scion in cm	Mean number of leaves		
	30 days	60 days	90 days
5	9.99	11.29	12.16
6	10.35	11.07	13.20
8	12.20	14.02	15.29
F test	NS	NS	NS
C.D.	-	-	-
SEm \pm	0.54	0.73	0.89

Table 10. Effect of height of rootstock and length of scion on girth of stock at 30 days intervals

Height of root stock in cm	Length of scion in cm	Mean girth of stock in cm		
		30 days	60 days	90 days
2-4	5	2.13	2.21	2.41
	6	2.18	2.26	2.35
	8	2.63	2.51	2.53
4-6	5	2.31	2.36	2.53
	6	2.25	2.36	2.51
	8	2.30	2.51	2.53
6-8	5	2.15	2.31	2.38
	6	2.00	2.06	2.20
	8	2.00	2.18	2.30
8-10	5	2.03	2.16	2.25
	6	2.00	2.18	2.33
	8	2.00	2.21	2.35
F test		NS	NS	NS
C.D.		-	-	-
SEm \pm		0.34	0.35	0.27

Table 11. Effect of height of rootstock on girth of stock at 30 days intervals

Height of rootstock in cm	Mean girth of stock in cm		
	30 days	60 days	90 days
2-4	2.28	2.40	2.50
4-6	2.28	2.41	2.52
6-8	2.05	2.18	2.26
8-10	2.01	2.18	2.31
F test	NS	NS	NS
C.D.	-	-	-
SEm \pm	0.03	0.06	0.05

Table 12. Effect of length of scion on girth of stock at 30 days intervals

Length of scions in cm	Mean girth of stock in cm		
	30 days	60 days	90 days
5	2.15	2.26	2.37
6	2.10	2.22	2.35
8	2.20	2.41	2.48
F test	NS	NS	NS
C.D.	-	-	-
SE m \pm	0.03	0.05	0.04

in the effect was not significant.

4.1.e. Girth of scion

The data presented in Table 13 show the girth of scion as affected by height of rootstock and length of scion at intervals of 30 days for a period of 3 months. Analysis of the data showed that the height of rootstock and length of scion had no significant effect on girth of scion.

The statistical analysis of the aggregate data on the effect of height of rootstock on girth of scion pooled over different length of scion are furnished in Table 14 also indicated that height of rootstock had no pronounced effect on girth of scion during the entire period of observations.

The results of analysis of the aggregate data on the effect of length of scion on the girth of scion pooled over the different height of rootstock are furnished in Table 15. The results indicated that the length of scion had no positive effect on the girth of scion during the course of investigations.

Table 13. Effect of height of rootstock and length of scion on girth of scions at 30 days intervals

Height of rootstock in cm	Length of scion in cm	Mean girth of scion in cm		
		30 days	60 days	90 days
2-4	5	1.78	1.88	1.98
	6	1.80	1.88	1.95
	8	2.00	2.15	2.21
4-6	5	2.19	2.36	2.41
	6	2.08	2.20	2.28
	8	2.03	2.21	2.25
6-8	5	2.03	2.08	2.23
	6	1.95	2.01	2.18
	8	1.91	2.05	2.18
8-10	5	1.92	2.00	2.08
	6	1.96	2.10	2.25
	8	2.00	2.18	2.26
	F test	NS	NS	NS
	C.D.	-	-	-
	SEm \pm	0.24	0.27	0.29

Table 14. Effect of height of rootstock on girth of scion at 30 days intervals

Height of rootstock in cm	Mean girth of scion in cm			
	30 days	60 days	90 days	
2-4	1.86	1.97	2.05	
4-6	2.10	2.26	2.31	
6-8	1.96	2.05	2.20	
8-10	1.96	2.09	2.20	
	F test	NS	NS	NS
	C. D.	-	-	-
	SEm \pm	0.04	0.05	0.04

4.2.a. Effect of covering materials on sprouting and final survival of grafts

The observations on the effect of covering materials on initial success and final survival of grafts are presented in Table 16.

The results indicated that covering materials had profound effect on initial success as well as a final survival of grafts. A maximum of 95.83 per cent of sprouting was recorded when grafts were covered with transparent polythene bags. The survival was also maximum (94.58 per cent) for this treatment. Covering the grafts with black polythene gave the lowest percentage of sprouting (20.20 per cent) and survival of 18.75 per cent. The effect of covering materials on initial success and final survival of grafts is depicted in (Fig.4.).

The pairwise comparison of the data using chi-square analysis indicated that covering the grafts with transparent polythene bag gave better sprouting as well as survival compared to black and control (Appendix VI).

Table 15. Effect of length of scions on girth of scions at 30 days intervals

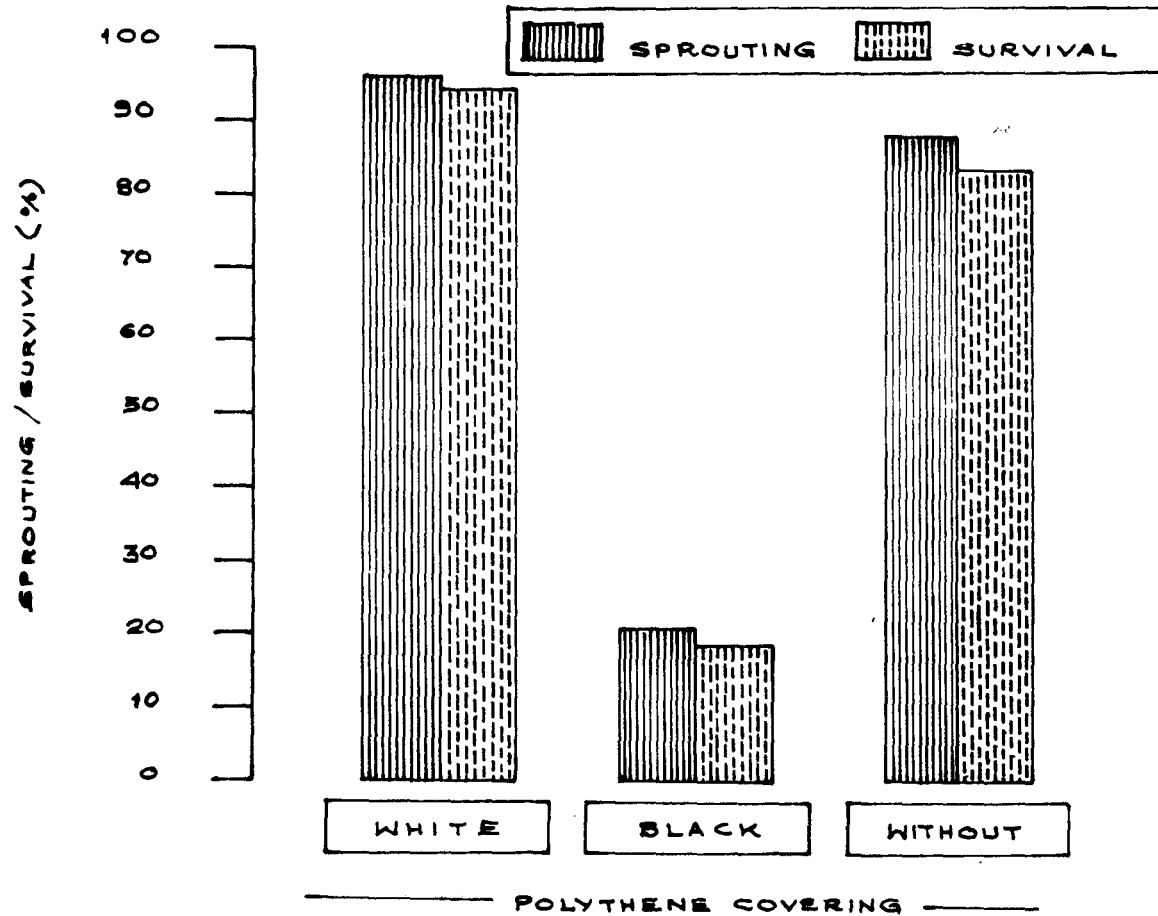
Length of scions in cm	Mean girth of scions in cm		
	30 days	60 days	90 days
5	1.98	2.08	2.17
6	1.95	2.05	2.16
8	1.98	2.15	2.22
F test	NS	NS	NS
C.D.	-	-	-
SEm \pm	0.03	0.04	0.04

Table 16. Effect of covering materials on sprouting and final survival of grafts

Types of covering materials	Total number of grafts	Sprouting Percentage	Survival Percentage
Control	480	88.54	83.54
Transparent Polythene	480	95.83	94.58
Black Polythene	480	20.20	18.75
Chi-square value		31.246**	714.27**

** Significant at 1% level.

FIG. 4. EFFECT OF POLYTHENE COVERING ON SPROUTING AND SURVIVAL OF GRAFTS



4.2.b. Extension growth of scion

The observations recorded on extension growth of scion at monthly intervals are tabulated in Table 17. The effect of covering materials on extension growth of scion was analysed statistically adopting analysis of variance technique and it was observed that covering materials had no significant effect on extension growth of scion. However, the extension growth of scion seemed to be slightly higher in control treatment where no covering material was used.

4.2.c. Number of leaves

The observations on the number of leaves produced at monthly intervals are tabulated in Table 18. The results of the statistical analysis of the data indicated that the covering material had no significant effect on the number of leaves produced. However, the control was found to be slightly better in this respect.

4.2.d. Girth of stock

The observations on the girth of stock as influenced by the covering materials are presented in Table 19. Although results of the statistical

Table 17. Effect of covering material on extension growth of scion at 30 days intervals

Type of covering materials	Mean extension growth of scion in cm		
	30 days	60 days	90 days
Control	6.80	7.88	9.06
Transparent Polythene	5.25	5.60	5.94
Black Polythene	5.16	6.32	7.04
F test	NS	NS	NS
C.D.	-	-	-
SEm \pm	0.31	0.36	0.49

Table 18. Effect of covering materials on number of leaves at 30 days intervals

Types of covering materials	Mean number of leaves		
	30 days	60 days	90 days
Control	9.11	11.61	15.20
Transparent Polythene	8.23	8.82	10.80
Black Polythene	8.41	10.45	13.50
F test	NS	NS	NS
C.D.	-	-	-
SEm \pm	0.54	0.92	1.10

Table 19. Effect of covering materials on girth of stock at 30 days intervals

Types of covering materials	Mean girth of stock in cm		
	30 days	60 days	90 days
Control	2.01	2.12	2.27
Transparent polythene	2.02	2.13	2.31
Black polythene	1.85	1.96	2.11
F test	NS	NS	NS
C.D.	-	-	-
SEm \pm	0.02	0.04	0.04

analysis indicated no significant difference between the treatments, the girth of stock was found to be slightly higher in control (2.11 cm) where no covering materials were used.

4.2.e. Girth of scion

The data on effect of covering material on scion girth are furnished in Table 20. Statistical analysis of the data indicated that the girth of scion was not influenced significantly by covering material.

4.3.a. Effect of growth regulators on sprouting and survival of grafts

The observations on the effect of different plant growth regulators on sprouting and survival of grafts are presented in Table 21.

The results of the chi-square analysis indicated high significant differences between the treatments (Fig.5.). However, the pairwise comparison between treatments revealed that IAA 100 ppm, GA 250 ppm and control were on par (Appendix VIII). The high significant difference was found to be due to GA treatment which was giving minimum percentage of sprouting (77 per cent).

Table 20. Effect of covering material on girth of scion at 30 days intervals

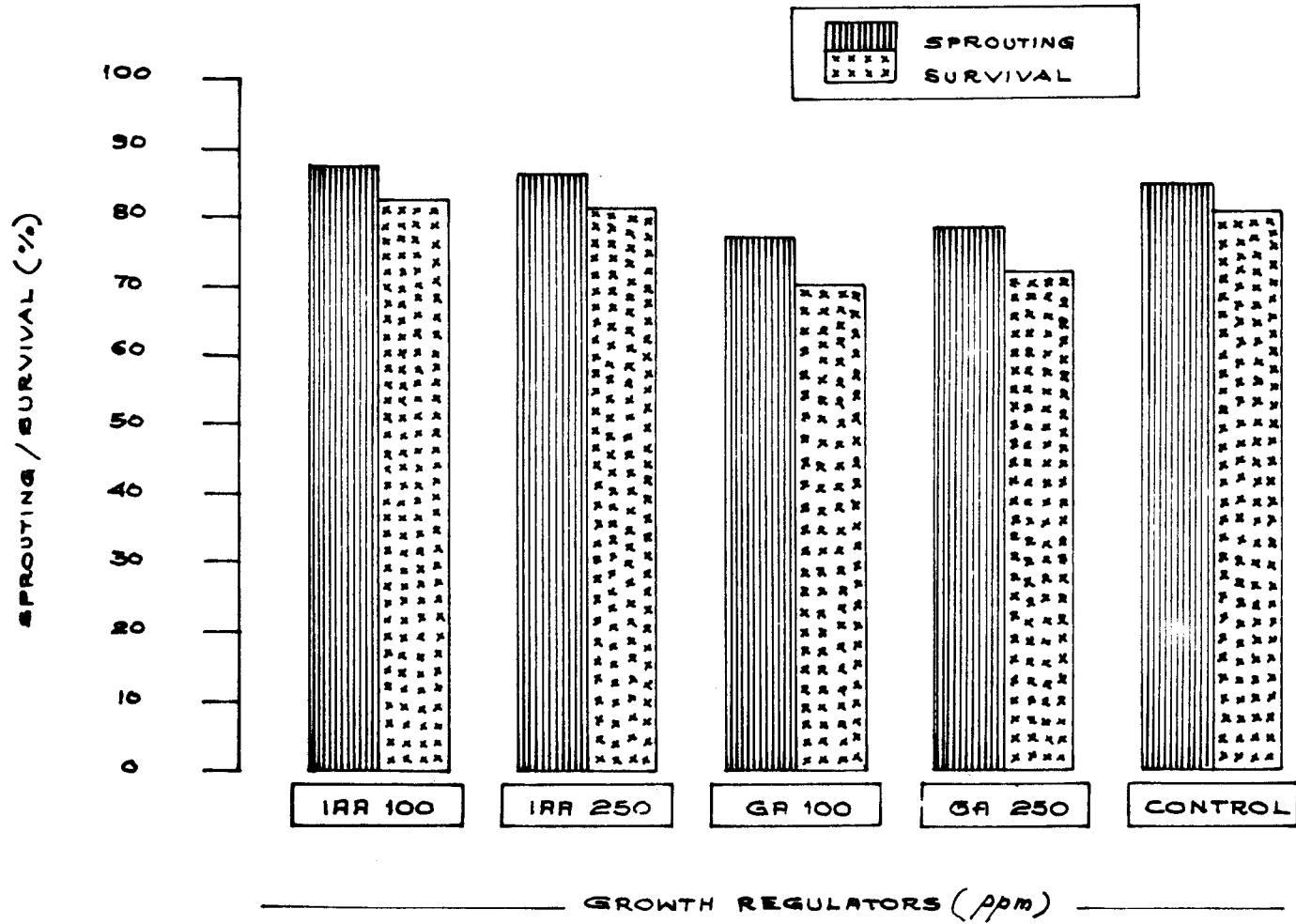
Types of covering materials	Mean girth of scion in cm		
	30 days	60 days	90 days
Control	1.82	1.99	2.11
Transparent polythene	1.87	1.98	2.11
Black polythene	1.69	1.77	1.85
F test	NS	NS	NS
C.D.	-	-	-
SE m \pm	0.04	0.03	0.03

Table 21. Effect of growth regulators on sprouting and final survival of grafts

Growth regulators in ppm	Total No. of grafts	Sprouting percentage	Survival percentage
Control	200	85.0	80.5
IAA 100	200	87.5	81.5
IAA 250	200	86.5	81.5
GA 100	200	77.00	70.0
GA 250	200	78.5	72.0
Chi-square		13.03**	310.70**

** Significant at 1% level.

FIG. 5. EFFECT OF GROWTH REGULATORS ON SPROUTING AND SURVIVAL OF GRAFTS



Further, Pairwise comparison showed no significant difference between the two treatments of GA viz., GA at 100 ppm and 250 ppm.

4.3.b. Extension growth of scion

The data on the effect of different plant growth regulators on extension growth of scion are tabulated in Table 22. The results of the statistical analysis revealed that growth regulators tried had no significant effect on the growth of scion. However, maximum growth was noticed in IAA 100 ppm over the others.

4.3.c. Number of leaves

The data on the effect of growth regulators on number of leaves produced are presented in Table 23. It is evident from the table that the growth regulators did not affect significantly the number of leaves produced. It was observed that all the treatments including control were on par with regard to number of leaves produced.

4.3.d. Girth of stock

The data on the effect of growth regulators on girth of stock are presented in Table 24.

Table 22. Effect of growth regulators on extension growth of scion at 30 days intervals

Growth regulators in ppm	Mean extension growth of scion in cm		
	30 days	60 days	90days
Control	6.37	7.36	8.77
IAA 100	7.00	7.70	9.13
IAA 250	5.90	6.83	8.18
GA 100	7.02	7.56	8.65
GA 250	5.96	6.20	7.58
F test	NS	NS	NS
C.D.	-	-	-
SEM \pm	0.70	0.64	0.95

Table 23. Effect of growth regulators on number of leaves at 30 days intervals

Growth regulators in ppm	Mean number of leaves		
	30 days	60 days	90 days
Control	11.25	14.75	19.50
IAA 100	12.25	15.25	20.00
IAA 250	12.00	15.15	20.00
GA 100	11.25	16.00	20.75
GA 250	12.00	17.75	20.25
F test	NS	NS	NS
C.D.	-	-	-
SE m \pm	0.64	0.99	2.67

Table 24. Effect of growth regulators on girth of stock at 30 days intervals

Growth regulators in ppm	Mean girth of stock in cm		
	30 days	60 days	90 days
Control	1.92	2.05	2.37
IAA 100	1.97	2.25	2.52
IAA 250	1.97	2.07	2.22
GA 100	1.96	2.09	2.24
GA 250	1.90	2.12	2.32
F test	NS	NS	NS
C.D.	-	-	-
SEM \pm	0.06	0.29	0.12

From the analysis of data it could be stated that treatment of growth regulators had no significant effect on the girth of stock.

4.3.e. Girth of scion

The observations recorded on girth of scion influenced by various treatments are presented in Table 25.

From the results presented in the Table it is evident that the highest scion girth (2.44 cm) was recorded with IAA 100 ppm while lowest scion girth (2.25 cm) was recorded with IAA 250 ppm at the end of 90th days of observation.

4.4.a. Effect of fungicides on final survival of grafts

A maximum intensity of dieback disease caused by fungal pathogen, colletotrichum gloeosporioides (penzig) sacc was observed from 2nd week of August onward to October end. The observations on the effect of fungicides on the final survival of grafts are given in Table 26. The results of the chi-square analysis indicated that all the fungicides sprayed controlled the die-back disease to some extent and gave a better survival. There was significant difference between various fungicides tried. Spraying of Bordeaux

Table 25. Effect of growth regulators on girth of scion at 30 days intervals

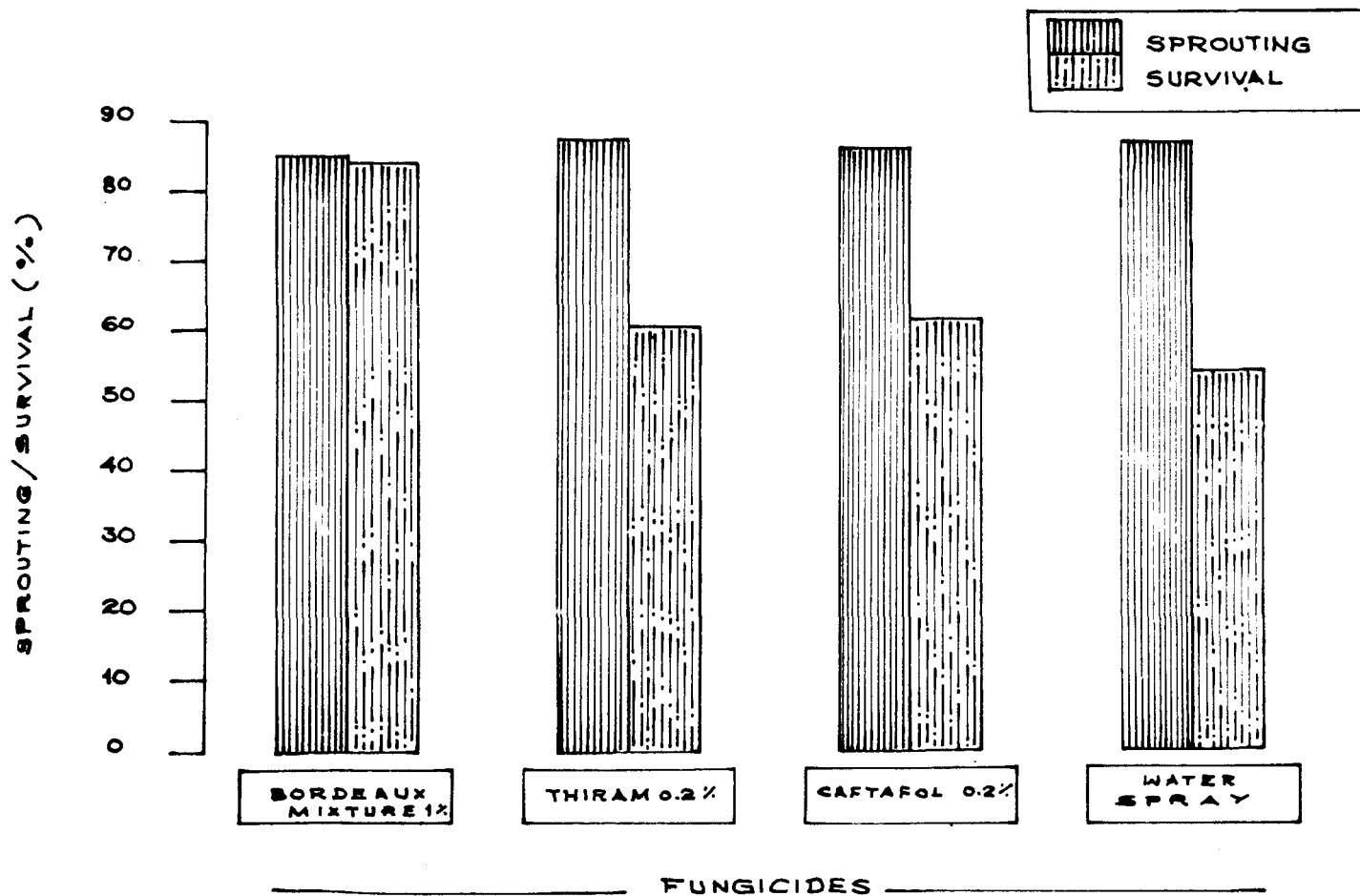
Growth regulators in ppm	Mean girth of scion in cm		
	30 days	60 days	90 days
Control	1.90	2.02	2.27
IAA 100	2.01	2.19	2.44
IAA 250	1.87	2.00	2.25
GA 100	1.90	2.03	2.27
GA 250	1.80	2.02	2.27
F test	NS	NS	NS
C.D.	-	-	-
SEm + ‡	0.05	0.10	0.73

Table 26. Effect of fungicides on final survival of grafts

Fungicides	Total number of grafts	Sprouting percentage	Survival percentage
Control	200	87	54.0
Bordeaux mixture @ 1%	200	85	84.0
Thiram (75 W.P.) @ 0.2%	200	87.5	60.5
Caftafol @ 0.2%	200	86	61.0
Chi-square value		0.59 NS	45.58**

** Significant at 1% level.

FIG. 6. EFFECT OF FUNGICIDES ON FINAL SURVIVAL OF GRAFTS



mixture at a concentration of 1 per cent was found to be very effective in giving the highest survival percentage of 84 followed by caftafol at 0.2 per cent (61.0 per cent) and Thiram at 0.2 per cent (60.5 per cent). The minimum survival percentage of 54 was recorded for control where only water was sprayed (Fig.6.).

Pairwise comparison of treatments also indicated that grafts treated with Bordeaux mixture at 1 per cent survival was better than all other treatments (Appendix X).

4.4.b. Extension growth of scion

A perusal of the data presented in Table 27 indicated that the fungicides had no significant effect on extension growth of scion at the end of experiment.

4.4.c. Number of leaves

Effect of fungicidal treatments on number of leaves produced for a period of three months are presented in Table 28.

Results of the statistical analysis indicated that there was no significant difference between the treatments.

Table 27. Effect of fungicides on extension growth of scion at 30 days intervals

Fungicides	Mean extension growth of scion in cm		
	30 days	60 days	90 days
Control	6.81	7.70	8.60
Bordeaux Mixture @ 1%	6.80	8.27	8.97
Thiram (75 W.P.) @ 0.2%	6.70	7.50	8.82
Coftafol @ 0.2%	6.74	7.63	8.53
F test	NS	NS	NS
C.D.	-	-	-
SEm \pm	0.40	0.68	0.59

Table 28. Effect of fungicides on number of leaves at 30 days intervals

Fungicides	Mean number of leaves		
	30 days	60 days	90 days
Control	11.00	13.00	14.75
Bordeaux mixture @ 1%	11.62	14.00	15.90
Thiram (75 W.P.) @ 0.2%	12.25	14.50	15.75
Caftafol @ 0.2%	11.25	13.75	13.75
F test	NS	NS	NS
C.D.	-	-	-
SEm \pm	0.92	1.52	1.59

Table 29. Effect of fungicides on girth of stock at 30 days intervals

Fungicides	Mean girth of stock in cm		
	30 days	60 days	90 days
Control	2.02	2.17	2.32
Bordeaux mixture @ 1%	2.07	2.30	2.52
Thiram (75 W.P.) @ 0.2%	2.02	2.22	2.37
Caftafol @ 0.2%	2.00	2.12	2.30
F test	NS	NS	NS
C.D.	-	-	-
SEm <u>+</u>	0.06	0.10	0.07

Table 30. Effect of fungicides on girth of scion at 30 days intervals

Fungicides	Mean girth of scion in cm		
	30 days	60 days	90 days
Control	1.90	2.05	2.30
Bordeaux mixture @ 1%	1.95	2.17	2.30
Thiram (75 W.P.) @ 0.2%	1.95	2.12	2.27
Caftafol @ 0.2%	1.97	2.10	2.27
F test	NS	NS	NS
C.D.	-	-	-
SEm <u>+</u>	0.05	0.09	0.07

4.4.d. Girth of stock

The data on the girth of stock as affected by various fungicides and water spray are tabulated in Table 29.

The analysis of the data indicated that fungicide had no significant effect on girth of stock.

4.4.e. Girth of scion

The observations on the girth of scion at a period of three months are given in Table 30. Here also all the treatments including control were on par with regard to their effect on the girth of scion.

5.5. Anatomical studies of the graft union

Anatomical studies were conducted with the objective to find out the possible reasons of graft failures. The different stages of development during different periods of observation are summarised below.

Formation of graft union

The following four main stages could be distinguished in the formation of the epicotyl graft union of mango.

Stage 1 (Pre callus - 24 hours)

Wound periderm was present as thick dark brown coating on external walls of exposed cells. There

was no sign of callus proliferation for a period of 24 hours after grafting (Plate XI). The scion would fall if the polythene wrap was removed.

Stage 2 (Callus - 5 days)

Callus formation started after 5 days of grafting as the scion was firmly attached to the stock. Wound periderm on cut surfaces was thicker and darker in colour than at stage 1 and was ruptured in many spots (Plate XII). Callus was proliferated from stock sides only. Living cells of mainly wood rays in the most recently formed xylem, cambial layers and phloem rays were involved, occasionally cells in the pith also produced callus. The original cut was easily traced where stock and scion tissues were necrotic or inactive.

Stage 3 (Cambial bridge - 15 days)

A cambial bridge across the union was noted 15 days after grafting (Plate XIII). Cambial layers extended circumferentially straight into the callus in the stock side but were strongly arched on the scion side (Plate XIV). Most of the callus proliferation originated from stock tissue particularly adjacent to the cambium. Growth was less active in scion side and confined to the tissues near cambium.

Stage 4 (healed union - 90 days)

Differentiation of apparently normal secondary xylem and secondary phloem from cambial layers in the callus was observed in unions examined 90 days after grafting (Plate XV). Closer views of this complete union have been shown in Plate XVI.

In these study two types of failure were observed viz., shrinking and dying of scions within 5 to 7 days after grafting and included scions remained green for more than 60 days but not sprouted.

The first type of graft failure was observed where there was no sign of callus proliferation even after 7 days of grafting (Plate XVII). Further, wound periderm seemed to be much more thicker than the successful grafts (Plate XVIII).

In the second type of grafts failures, there existed a wide gap between stock and scion (Plate XIX), although in some points, stock and scion were weakly connected. Closer views (Plate XX) indicated that in addition to wide gap, callus differentiation took place only in stock side and no rupturing of wound periderm in any point.

Plate XI. Anatomy of graft union 24 hours after grafting (X15X5X6.)

Plate XII. Callus formation of graft five days after grafting (X15X10X6.)

Plate XIII. Formation of cambial bridge between stock and scion after 15 days of grafting (X15X5X6.)

Plate XIV. Cambial layers from stock extended circumferentially and arched on the cambium of scion side (X15X10X6.)

Plate XV. Healed union 90 days after grafting (X15X5X6.)

Plate XVI. A closer views of healed union (X15X10X6.)

Stock Scion
← necrotic layers

XI

Stock Scion
← callus formation

necrotic layers

XII

scion
stock
← cambial bridge

XIII

stock scion
← cambial bridge

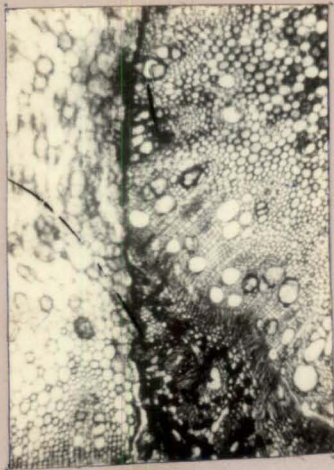
XIV

stock scion
← healed union

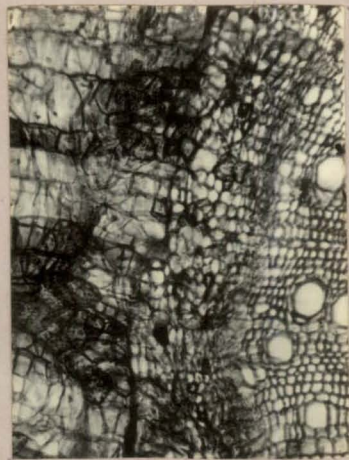
XV

stock scion
← healed union

XVI



XI



XII



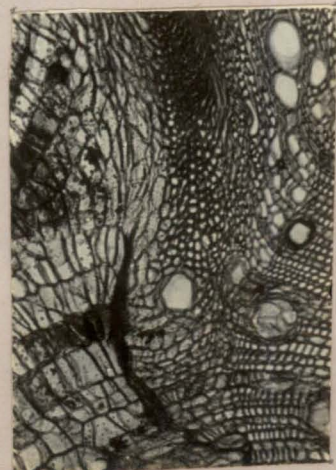
XIII



XIV



XV



XVI

Plate XVII. Anatomy of grafts failures showing no callus differentiation from both stock and scion (X15X5X6.)

Plate XVIII. Thick necrotic layers in unsuccessful grafts (X15X10X6.)

Plate XIX. Wide gap between stock and scion (X15X5X6)

Plate XX. Callus differentiation from stock side only even after 60 days of grafting (X15X10X6)

stock scion
← no callus
 differentiation
 necrotic layer

XVII

stock scion
 ← thick necrotic
 layer

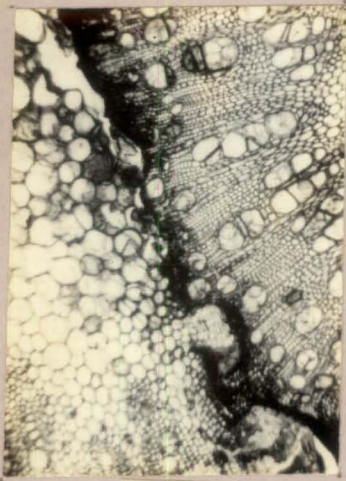
XVIII

stock scion
 ← wide gap

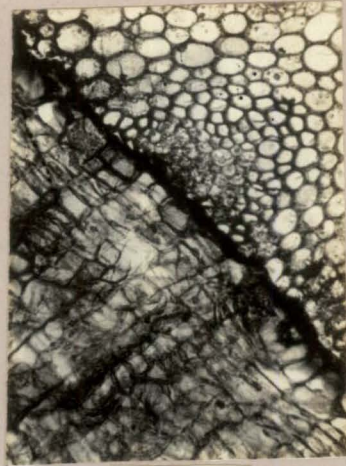
XIX

stock scion
 ← wide gap
 callus tissue
 formation

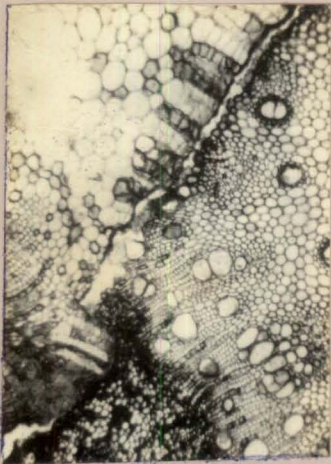
XX



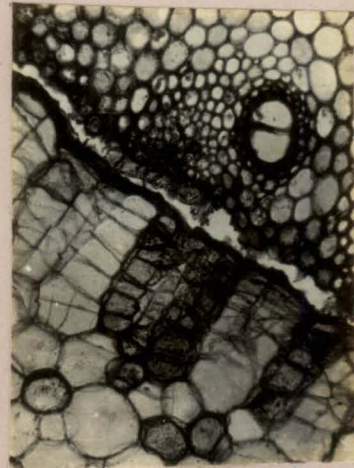
XVII



XVIII



XIX



XX

DISCUSSION

DISCUSSION

Vegetative propagation in mango has been hitherto carried out largely through approach grafting. Methods like veneer or epicotyl grafting which are comparatively of newer origin have virtually replaced the traditional method of inarching in several other states in India. Surprisingly neither veneer nor epicotyl grafting has become a commercial proposition among the nurserymen of Kerala, which has been mainly due to the absence of standardised technologies in these two methods suited to our conditions. The work initiated in the Department of Pomology, College of Horticulture during the years 1983-84 had given promising results and pointed out the possibility of adopting epicotyl grafting on a commercial basis.

In the earlier studies the season of grafting, precuring of scion, and age of stock and scion for stone grafting were standardised. It was, however, felt that there existed scope to improve the percentage of success by further refinement of the techniques, like standardisation of the length of scion and rootstock or by adopting protective measure like covering the grafts with polythene bags or application of fungicides against nursery diseases.

5.1. Standardisation of height of rootstock and length of scion

The effect of the height of rootstock on the success of grafting was demonstrated by Singh and Srivastava (1962) and Patel and Amin (1976) in mango budding and bench grafting respectively, Gunjate et al. (1980) in jackfruit and Nagabhusanam (1982) in cashew obtained maximum success when stone grafting was done at a height of 5 to 6 cm. Singh and Srivastava (1979) reported that tight fitting of the stock and scion in veneer grafting of mango gave better cambial activity resulting in easy and better union. Majumdar and Rathore (1970), Rajput and Haribabu (1971), Patel and Amin (1976) also reported that stock and scion should be of uniform thickness for the tight fitting of grafts.

The effect of scion length on the success of graft 'take' is also well established (Persai, 1963 and Kanwar and Bajwa, 1974 in side grafting of mango; Maity and Biswas, 1980 in stone grafting of mango; Ram and Bist, 1982 in veneer grafting of mango). The results of the present studies indicated that scion length influenced significantly the success of

graft 'take'. It was found that the scion length 8 cm was the best compared to scions of 5 cm or 6 cm length. Torres (1949) and Kanwar and Bajwa (1974) also reported the superiority of scion of 7.5 to 8 cm length for side grafting in mango. Dhungana (1984) had also used 8 to 10 cm long scions for stone grafting mango.

The superiority of 8 cm long scions could perhaps be attributed to higher reserve food materials in them compared to shorter scions. The low graft 'take' in the case of 5 cm and 6 cm long scions suggests that in shorter scions exhaustion of food material would take place before union is completed. Fahmy (1952) observed strong positive correlation between the amount of carbohydrates content in scion shoots and graft union in macademia and sapodilla.

The length of the scion or rootstocks appears to be more concerned with graft union rather than the subsequent growth of the grafts. The growth of the grafts as exhibited by the girth of stock or scion, number of leaves produced was not seen influenced by any of these factors.

5.2. Effect of covering materials

It was observed that covering the graft with transparent polythene film gave significant effect on

the success of epicotyl grafting. The percentage of success was 95.83 when the grafts were covered with transparent polythene film compared to control where the success was only 88.54 per cent.

Reyes (1978) and Singh and Srivastava (1982) also obtained higher success when mango grafts were covered with white polythene film. They stated that covering the grafts with white polythene film created high humidity inside and reflected radiant energy resulting in a low temperature inside the cover.

Dhungana (1984) observed a significant positive correlation between scion sprouting and relative humidity. He obtained maximum percentage of success (69.33) per cent during August when relative humidity was maximum (87 per cent). Hartmann and Kester (1978) suggested that the presence of a thin film of water in the callusing surface was more congenial for abundant callus formation. The influence of the quality of polythene material on graft 'take' is also noteworthy. The grafts covered with black polythene film recorded minimum percentage of success (20.20 per cent). Teotia and Maurya (1970) obtained minimum percentage of success for chip budding in mango when black polythene film was used for

covering. The detrimental effect of black polythene as covering material in budding and grafting was also reported by several workers (Bringezu and Penning, 1969 in apple; Jauhari and Singh, 1970 and Singh and Srivastava, 1979 in mango budding). They suggested that black polythene film absorbed more radiant energy resulting in a higher temperature near the graft union which was injurious to cell division and their further multiplication. Hartmann and Kester (1978) observed that dark coloured bark of plants would raise the temperature of living cells below the bark to a lethal point, thereby reducing the percentage of success of grafting.

Although the percentage of success was influenced significantly by polythene covering further growth of scion was not influenced by covering materials.

5.3. Effect of growth regulators

The present studies revealed that the application of growth regulators like IAA and GA each at concentrations of 100 and 250 ppm did not in any way influence the graft 'take'. The results are thus in conformity with the findings of Hensen and Hartmann (1951) who observed that auxin applied to the walnut graft union did not produce any significant effect on the percentage of success. It cannot

however, be ruled out that the growth regulator will not influence the graft 'take'. The chemicals at the concentrations tried might not perhaps be at optimum level to influence the healing of the graft union. The recent theory that the growth regulators function in a system rather in isolation also suggests further detailed study in this particular aspect.

Thompson (1945) had observed that treatment of tissue with growth regulators which had already activated such as meristems usually caused either inhibition of growth or distortion of normal growth pattern. The precuring of the scion would have perhaps increased the meristematic activity of the scion's as has been reported by several workers in various fruits (Munch, 1930 in apple; Teotia and Maurya, 1970; Rajput and Haribabu, 1971; Kasyap et al., 1972; Persai, 1974; Singh and Srivastava, 1979; Maity and Biswas, 1980 in mango).

5.4. Effect of fungicides

A high incidence of fungal infection of sprouted grafts resulting a low percentage of final survival was reported by Dhungana (1964) in both

epicotyl and veneer grafting under Vellanikkara conditions. He observed maximum infection of stone grafts during August. It was observed that fungal infection occurred during the second week of August and continued upto October. The dieback disease of grafts was mainly caused by colletotrichum gloeosporioides. The trials with different fungicides viz., Bordeaux mixture at 1%, Thiram at 0.2% and Caftafol at 0.2% showed that Bordeaux mixture at 1% was the most effective in controlling the disease, when sprayed one month after grafting and subsequently applied at intervals of 15 days for three months. Kanwar and Jawanda (1983) obtained good control of fungal disease with Bordeaux mixture paste in young grafts. The effectiveness of Bordeaux mixture in controlling dieback disease has been reported earlier (Anon.1981). The results of the study further indicate the necessity of protecting the grafts especially in the earlier stages against fungal infections.

5.5. Anatomical studies

The mechanism of the healing of graft union depends to a large extent on the activities of the

cambium and its derivative tissues (Esau, 1979; Fahn, 1982). The possible reasons for graft failures in a compatible species is often traced to anatomical deformities. It was found that the callus proliferation started within 5 days of grafting and continued upto 90 days. Most of the callus differentiated from stock side which contained maximum live cells than scions as was reported by Soule (1971) who observed that live cells were most important for graft union.

In cases of graft failures it was observed that there was no cell differentiation in the cut portions of stocks even after seven days of grafting. Moreover, in these cases necrotic layers formed in between stock and scion and they were found to be thicker than successful grafts. Soule (1971) stated that the thick necrotic layers were due to deep crushing of cells during wrapping which contributed to graft failures.

Another type of graft failure observed in the present study was where the scions remained green upto 60 days and did not sprout later. Anatomical studies

in such cases showed a wide gap between stock and scion. Irregular surfaces of the cut portions of stock and scion were also evident. This would mean that mechanical factors can as well impair the final success in any particular type of grafting. The necessity for skill in grafting operation especially in stone grafting cannot therefore be over emphasised. The observation of Copes (1969) that poorly matched stock and scion resulted in very slow cambial union and delayed bud sprouting points out this fact.

The genetic and biochemical factors interfering in the healing of grafts have been stressed by Robert (1949) and Robitaille and Carlson (1970). In mango although the stock and scion are not biochemically or genetically far apart, the possibility of some of these factors interfering the healing process cannot be ruled out.

Results of the present study as well as those obtained earlier clearly show that epicotyl grafting could be successfully employed in the vegetative propagation of mango in Kerala. However, epicotyl grafting is rather delicate compared to inarching, and has to be done with skill and under technical

supervision. The season of operation, precuring, height of rootstock and length of scion all play significant role in the ultimate analysis. The need for protective measure like covering the graft with suitable material and application of chemicals against diseases are necessary in order to obtain higher percentage of success. Stone grafting, in spite of all its inherent difficulties is worth adopting on a commercial scale in mango especially because the cost of production can be substantially reduced. Moreover, this method will be highly beneficial in propagating selected trees located in far off places.

SUMMARY

SUMMARY

In the earlier studies on vegetative propagation of mango at the College of Horticulture, Vellanikkara the season of stone grafting and precuring of scion shoots were standardised. However, it was felt that the percentage of graft 'take' could be further enhanced by standardising the height of rootstock, length of scion, covering the grafts with polythene bags, application of plant growth regulators and by protecting the grafts against fungal infection.

Hence, a series of experiments on epicotyl grafting in mango as reported in this thesis were carried out at the Department of Pomology and Floriculture, during the period from June 1983 to March 1985. The results are summarised below.

- 1) Studies on the standardisation of height of rootstock and length of scion were conducted during July-August 1983. The highest percentage of success (87.5 per cent) was obtained when grafting was done with scion of 8 cm length on rootstock at a height of 6 to 8 cm. The success decreased as the scion length decreased.

Growth studies of the grafts indicated that the height of stock or the length of scion did not influence their subsequent growth.

2) Covering the grafts with transparent polythene bags immediately after grafting was beneficial and the percentage of success and survival was enhanced to 95.83 per cent and 94.58 per cent respectively. Minimum percentage of sprouting and survival was obtained when grafts were covered with black polythene bags.

3) The studies on the application of different plant growth regulators like IAA and GA each at 100, 250 ppm on graft indicated that none of the treatments had significant effect on graft 'take' as well as on subsequent growth of the grafts.

4) The dieback disease of grafts was found to be caused by colletotrichum gloeosporioides and the infection was more intense during August to October. Treatments of grafts with different fungicides like Bordeaux mixture at 1%, Thiram at 0.2% and Caftafol at 0.2% after one month of grafting were effective in controlling the dieback disease. Of these chemicals, Bordeaux mixture at 1% was most effective in controlling the dieback disease which gave 84 per cent of survival.

5) Anatomical studies clearly indicated four distinct stages during the course of graft union. The possible reasons for graft failures appeared to be due to the formation of thick necrotic layers and wide gap between stock and scion.

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APPENDIX I

Chi-square values for comparisons between pair of height of rootstock length of scion with regard to the number of sprouting of grafts

scion/stock	5/2-4	6/2-4	8/2-4	5/4-6	6/4-6	8/4-6	5/6-8	6/6-8	8/6-8	5/8-10	6/8-10	8/8-10
5/2-4	-	4.58*	4.43*	32.76**	4.49*	189.08**	59.88**	79.35**	26.41**	4.07*	11.53**	62.52**
6/2-4	-	-	30.19**	19.74**	40.58**	98.45**	38.40**	54.84**	133.70**	0.20	7.85**	40.58**
8/2-4	-	-	-	0.41	1.65	44.66**	1.21	5.65*	114.87**	30.19**	12.00**	11.65**
5/4-6	-	-	-	-	3.39	50.00**	2.75	7.57**	125.52**	17.36**	10.90**	12.82**
6/4-6	-	-	-	-	-	38.23**	0.03	2.13	9.81**	31.34**	19.07**	1.33
8/4-6	-	-	-	-	-	-	40.43**	26.28**	14.25**	136.68**	112.16**	38.23**
5/6-8	-	-	-	-	-	-	-	2.70	101.40**	30.28**	17.54**	0.03
6/6-8	-	-	-	-	-	-	-	-	79.73**	44.17**	29.52**	2.13
8/6-8	-	-	-	-	-	-	-	-	-	204.21**	175.92**	83.36**
5/8-10	-	-	-	-	-	-	-	-	-	-	2.98	31.34**
6/8-10	-	-	-	-	-	-	-	-	-	-	-	19.07**
8/8-10	-	-	-	-	-	-	-	-	-	-	-	-

** Significant at 1% level

* Significant at 5% level

APPENDIX II

Chi-square values for comparison between pair of height of rootstock and length of scion with regard to the number of survival of grafts

scion/stock	5/2-4	6/2-4	8/2-4	5/4-6	6/4-6	8/4-6	5/6-8	6/6-8	8/6-8	5/8-10	6/8-10	8/8-10
5/2-4	-	8.62**	63.63**	34.49**	77.35**	212.78**	76.48**	98.18**	262.80**	2.60	11.64**	58.40**
6/2-4	-	-	34.84**	23.13**	45.95**	167.09**	43.63**	73.73**	214.28**	0.70	1.04	30.73**
8/2-4	-	-	-	0.60	1.73	64.82**	1.28	6.76*	99.43**	39.67**	29.40**	0.00
5/4-6	-	-	-	-	6.15*	83.25**	5.27*	14.10**	121.12**	27.13**	11.92**	1.36
6/4-6	-	-	-	-	-	51.30**	0.00	2.75	83.00**	51.37**	30.18**	2.24
8/4-6	-	-	-	-	-	-	43.63**	84.43**	6.40*	192.66**	173.64**	118.76**
5/6-8	-	-	-	-	-	-	-	3.75	6.23*	48.94**	28.25**	2.08
6/6-8	-	-	-	-	-	-	-	-	62.72**	69.61**	45.17**	8.88**
8/6-8	-	-	-	-	-	-	-	-	-	221.29**	202.64**	106.40**
5/8-10	-	-	-	-	-	-	-	-	-	-	6.00*	46.54**
6/8-10	-	-	-	-	-	-	-	-	-	-	-	6.91*
8/8-10	-	-	-	-	-	-	-	-	-	-	-	-

** Significant at 1% level

* Significant at 5% level

APPENDIX III

Chi-square values for comparisons between pair of rootstock heights with regard to the number of sprouting and survival of grafts.

Comparisons	Chi-square value	
	Sprouting	Survival
2-4 vs 4-6	49.59**	132.89**
2-4 vs 6-8	191.14**	201.51**
2-4 vs 8-10	0.54	2.26
4-6 vs 6-8	10.95**	9.34**
4-6 vs 8-10	8.20**	124.48**
6-8 vs 8-10	140.10**	195.51**

** Significant at 1% level.

APPENDIX IV

Chi-square values for comparisons between pair of scion length with regard to the number of sprouting and survival of grafts.

Comparisons	Chi-square value	
	Sprouting	Survival
5 vs 6	14.31**	14.24**
5 vs 8	211.55**	228.21**
6 vs 8	143.61**	137.96**

** Significant at 1% level.

APPENDIX V

Analyses of variance for the effect of height of rootstock and length of scion on extension growth of scion, number of leaves, girth of stock and scion at 30 days

source	d.f.	Mean squares at 30 days			
		scion	extension - no. of leaves	girth of stock	girth of scion
Scion length	2	1.17	2.16	0.30	0.48
Height of rootstock	3	1.76	2.24	0.19	0.86
Interaction	6	1.60	1.75	0.46	0.23
Error	24	1.40	1.35	0.13	0.16
Total	35	-	-	-	-

APPENDIX VI

Chi-square values for comparisons between pair of covering materials with regard to the number of sprouting and survival of grafts

Comparisons	Chi-square value	
	Sprouting	Survival
White covering vs black covering	114.63**	111.18**
White covering vs control	31.51**	55.62*
Black covering vs control	914.50**	479.75*

** Significant at 1% level.

APPENDIX VII

Analyses of variance for the effect of covering materials on scion extension number of leaves, girth of stock and scion at 30 days

source	d.f.	Mean squares at 30 days			
		scion extension	no. of leaves	girth of stock	girth of scion
Covering	2	1.42	2.10	0.44	0.45
error	12	0.48	1.14	0.44	0.92
Total	14	-	-	-	-

APPENDIX VIII

Chi-square values for comparisons between pair of growth regulators with regard to the number of sprouting and survival of grafts

Comparisons	Chi-square value	
	Sprouting	Survival
T ₁ vs T ₂	0.707	0.707
T ₁ vs T ₃	123.64**	14.60**
T ₁ vs T ₄	9.07**	10.27**
T ₁ vs T ₅	1.76	0.132
T ₂ vs T ₃	9.68**	12.00**
T ₂ vs T ₄	6.78**	8.09**
T ₂ vs T ₅	1.06	0.707
T ₃ vs T ₄	1.06	1.06
T ₃ vs T ₅	10.52**	14.20**
T ₄ vs T ₅	7.54**	9.96**

** Significant at 1% level

APPENDIX IX

Analyses of variance for the effect of growth regulators on scion extension, numbers of leaves, girth of stock and scion at 30 days

source	d.f.	Mean squares at 30 days			
		scion extension	no. of leaves	girth of stock	girth of scion
Treatments	4	0.87	1.10	0.42	0.24
error	15	1.19	1.16	0.16	0.13
Total	19	-	-	-	-

APPENDIX X

Chi-square values for comparisons between pair of fungicides with regard to the number of sprouting and survival of grafts

Comparisons	Chi-square value	
	Sprouting	Survival
Bordeaux mixture vs Thiram	2.06	50.50**
Bordeaux mixture vs Caftafol	1.02	48.55**
Bordeaux mixture vs water spray	1.49	78.63**
Thiram vs Caftafol	0.74	0.18
Thiram vs water spray	0.37	4.59*
Caftafol vs water spray	0.37	5.23*

** Significant at 1% level

* Significant at 5% level

APPENDIX XI

Analyses of variance for the effect of fungicides on scion extension, number of leaves, girth of stock and scion at 30 days

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source	d.f.	Mean squares at 30 days			
		scion extension	no. of leaves	girth of stock	girth of scion
Treatments	3	0.11	1.11	0.39	0.39
error	12	0.64	1.33	0.16	0.13
Total	15	-	-	-	-

APPENDIX XII

Weather data for the period from January 1983 to December 1983

Month	Temperature (0°C)		mean relative humidity (per cent)	Total rainfall (mm)	Total rainy days
	Maximum	minimum			
January	33.25	21.64	51.31	nil	nil
February	34.46	22.7	64.00	nil	nil
March	36.15	23.76	65.00	nil	nil
April	36.2	25.8	66.00	nil	nil
May	35.1	25.5	69.00	37.4	3
June	31.9	24.5	79.00	387.2	19
July	29.7	23.7	87.00	580.6	21
August	29.1	23.8	87.00	754.7	26
September	29.5	23.4	84.00	494.6	24
October	31.2	23.1	77.00	149.8	6
November	31.8	22.3	71.00	60.2	3
December	31.2	23.9	63.00	24.4	3

APPENDIX XIII

Weather data for the period from January 1984 to December 1984

Month	Temperature (0°C)		mean relative humidity (per cent)	Total rainfall (mm)	Total rainy days
	maximum	- minimum			
January	32.4	23.3	58	nil	nil
February	39.3	24.2	56	27.0	3
March	35.2	24.3	51	18.9	2
April	34.5	24.9	86	109.2	9
May	34.5	25.8	71	40.6	6
June	29.0	22.7	87	853.1	28
July	28.6	22.9	87	730.4	24
August	29.3	22.2	83.5	260.2	21
September	30.4	23.2	80.0	158.6	7
October	29.9	22.1	67.5	323.7	12
November	32.00	23.13	66.7	7.8	1
December	31.9	20.8	58.0	16.4	1

STANDARDISATION OF EPICOTYL GRAFTING IN MANGO

By

JOMBO RATAN

ABSTRACT OF A THESIS

submitted in partial fulfilment of
the requirement for the degree

MASTER OF SCIENCE IN HORTICULTURE

Faculty of Agriculture

Kerala Agricultural University

Department of Horticulture

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COLLEGE OF HORTICULTURE

Vellanikkara - Trichur

1985

ABSTRACT

The season of epicotyl grafting, age of root stock and scion and precuring of scions were standardised by Dhungana (1984) in the earlier studies. The present series of experiments were undertaken with the objective to enhance the grafts 'take' by different methods.

Studies reported in this thesis were carried out during the period from June 1983 to March 1985 and consisted of experiments on standardisation of the height of root stock, length of scion, effect of covering material, effect of different plant growth regulators and different fungicides on dieback disease. Anatomical studies were also undertaken to find out the possible reasons of graft failures.

The results of the experiments on the influence of the height of rootstocks (2 to 4 cm, 4 to 6 cm, 6 to 8 cm and 8 to 10 cm) and the length of scion (5 cm, 6 cm and 8 cm) indicated that the height of rootstock and length of scion had significant effect on the success of epicotyl grafting in mango. It was observed that when mature 3 to 4 months old, ten days precured scion of 8 cm were grafted on five to ten days old rootstock at 6 to 8 cm height gave maximum percentage of sprouting and survival during July-August.

In the experiments to find out the influence of covering the grafts with polythene bags both transparent and black polythene bags were used. The grafts were covered with bags (15 x 20 cm) leaving sufficient space at the top. It was observed that covering the grafts with transparent polythene bags and retaining them for one month gave maximum percentage of success as well as final survival compared to control. Black polythene material was detrimental for graft 'take'.

In another experiments the grafts were sprayed with IAA and GA each at 100, 250 ppm immediately after grafting in order to find out whether these growth regulators would help to increase the graft 'take'. Although no significant effect could noticed, it appeared that IAA at 100 ppm had some beneficial effect.

The dieback disease of grafts was found to be caused by colletotrichum gloeosporioides and the disease was more sever during August to October. The trials with different fungicides viz., Bordeaux mixture at 1%, Thiram at 0.2% and Caftafol at 0.2%, revealed that Bordeaux mixture was the most effective in controlling the disease when applied one month after grafting and subsequently sprayed at an intervals of 15 days till 90 days.

Anatomical studies of the successful and failed grafts were also studied in order to understand the possible reasons for graft failures. It was observed that in successful grafts callus proliferation commenced from 5th day onwards and the completion of cambial bridge of the stock and scion was attained 15 days after grafting. The completion of cambial union was indicated by sprouting of grafts. There were four distinct stages in the healing of the grafts.

In unsuccessful grafts there was no indication of callus proliferation. Thicker necrotic layers were also formed in the region of graft union.

In grafts which remained alive upto 60 days, but did not sprout there were wide gap between stock and scion which perhaps inhibited sprouting of the scions.