

**Studies on Seed Germination and Epicotyl Grafting in  
Walnut (*Juglans regia* L.)**

**Suja Nabi Qureshi**  
(2010-310-D)



**Division of Fruit Science**  
**Faculty of Postgraduate Studies**  
**Sher-e-Kashmir University of Agricultural Sciences &  
Technology of Kashmir**  
**2014**

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**Thesis**

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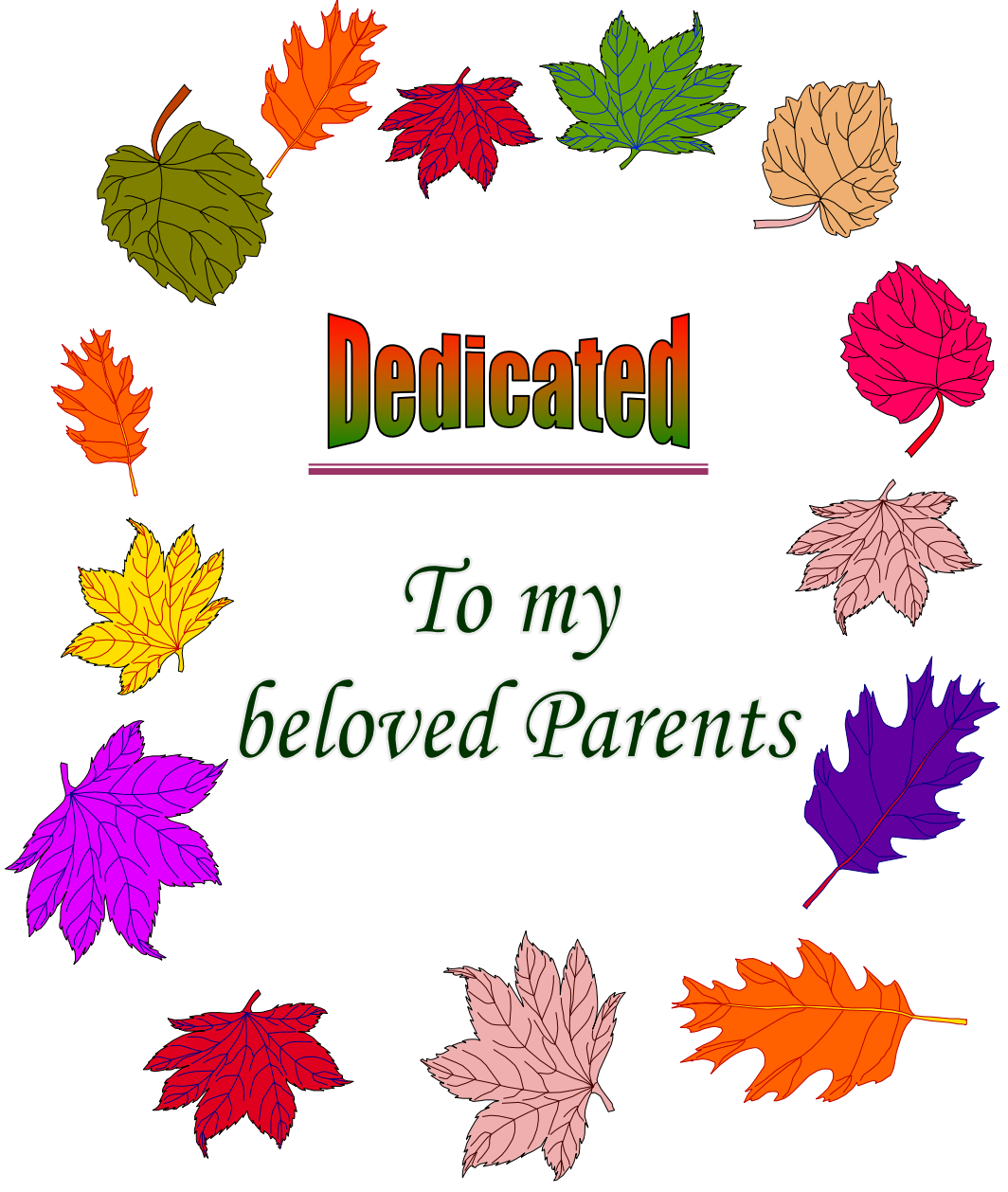
**The Faculty of Postgraduate Studies**

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**in partial fulfilment of requirement for the award of the degree of**

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**2014**



**Dedicated**

*To my  
beloved Parents*

**Sher-e-Kashmir**  
**University of Agricultural Sciences & Technology of Kashmir**  
**Division of Fruit Science, Shalimar Campus Srinagar**  
**190 025**

**Certificate – I**

This is to certify that the thesis entitled, “**Studies on Seed Germination and Epicotyl Grafting in Walnut (*Juglans regia* L.)**” submitted in partial fulfillment of the requirements for the award of the degree of **Doctor of Philosophy in Horticulture (Fruit Science)**, to the Faculty of Postgraduate Studies, Sher-e-Kashmir University of Agricultural Sciences & Technology of Kashmir is a record of bonafide research work carried out by **Mr. Suja Nabi Qureshi (Regd. No. 2010-310-D)** under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

It is further certified that information received during the course of investigation has duly been acknowledged.

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This is to certify that the thesis entitled, “**Studies on Seed Germination and Epicotyl Grafting in Walnut (*Juglans regia* L.)**” submitted by **Mr. Suja Nabi Qureshi (Regd. No. 2010-310-D)** to the **Faculty of Postgraduate Studies, Sher-e-Kashmir University of Agricultural Sciences & Technology of Kashmir** in partial fulfilment of the requirements for the award of the degree of **Doctor of Philosophy in Horticulture (Fruit Science)** was examined and approved by the Advisory Committee and External Examiner on .....

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### **ABSTRACT**

The present investigation entitled “Studies on seed Germination and Epicotyl Grafting in Walnut (*Juglans regia* L.)” was carried out in the Fruit Plant Nursery of Division of Fruit Science, SKUAST-Kashmir, Shalimar, Srinagar, J&K during the year 2011 and 2012. The walnut seeds were treated with four chemicals each at two levels, storage viz., cold and ambient treatments and seed treatments viz., un-soaked and soaked. Epicotyl grafting was performed on the basis of age of rootstocks (15, 30 and 45) days after germination and height of graftings viz., point of attachment cotyledons, 2cm, 4cm and 6cm above the point of attachment of cotyledons. The chemical treatments viz. GA<sub>3</sub>, sulphuric acid, ethrel and potassium nitrate were significantly effective in breaking the seed dormancy and promoting seedling growth of walnut. GA<sub>3</sub> at 100ppm was significantly effective in inducing seed germination compared to other chemical treatments and control. Highest seed germination percentages (58.24) during 2011 and (58.57) during 2012 were recorded with GA<sub>3</sub> 100ppm followed by GA<sub>3</sub> at 50ppm (54.23) during 2011 and (56.32) during 2012. The storage treatments under cold environment recorded maximum germination percentages 46.51 during 2011 and 46.56 during 2012 whereas the germination percentages were 42.21 during 2011 and 42.79 during 2012 under ambient storage (10-12°C). Un-soaked seed treatment under cold storage resulted in germination percentage of 45.36 during 2011 and 45.53 during 2012, whereas in pre-soaked treatment germination

percentages to the tune of 43.36 during 2011 and 43.83 during 2012 were recorded. The interactions between storage and chemical treatments were found significant both in seed germination and seedling growth. Highest graft take (67.38 and 70.55%) and graft sprout (40.93 and 41.32% ) were observed in epicotyl grafting performed on 15 day old walnut rootstocks at the point of cotyledon attachment followed by grafting at 2cm height both during 2011 and 2012. With regard to the quality of scion wood, terminal portion of scion wood was found superior as for as scion take and scion sprouting is concerned. Maximum scion take percentages (62.99 and 64.41) and scion sprouting percentages of (32.18 and 31.85) were recorded under terminal portion of scion wood during both 2011 and 2012, in comparison to basal portion of scion wood in which scion take of 59.01 and 63.74 per cent and scion sprouting of 29.21 and 29.63 per cent were recorded during both 2011 and 2102. Minimum graft success per cent was recorded in case of epicotyl grafting performed at height of 6cm from the point of attachment of cotyledons. The overall results indicate that germination of walnuts at late stage could be feasible with the use of GA<sub>3</sub> at 100ppm and 50ppm concentrations without pre-soaking treatment. Epicotyl grafting under open conditions seems to be viable option for mass propagation of walnuts. However, skill development of nurserymen with respect to grafting technique as well as proper management of propagated material seems to be of vital importance.

**Key Words :** Epicotyl grafting, Seed germination and Walnut (*Juglans regia* L.)

Signature of Student

Dated : \_\_\_\_\_

Signature of Major Advisor

Dated: \_\_\_\_\_

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***Suja Nabi Qureshi***

**Place :** Shalimar, Srinagar

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## Chapter – 1

### INTRODUCTION

The Persian walnut (*Juglans regia* L.) known as English walnut is the most valuable commercial species in its genus belonging to family Juglandaceae, containing about 60 species and 21 of which are placed in genus *Juglans*. The Persian walnut has origin in eastern Europe, Asia minor, extending from Turkey, Iran and western China to eastward to the Himalayan regions (Lesile and McGranahan, 1998). All juglans species are monoecious, with catkins being borne laterally on one year old wood and pistillate flowers borne terminally on current seasons wood. Although walnuts are genetically self fruitful they exhibit the phenomenon of dichogamy, being either protandrous or protogynous depending on cultivar, therefore are mostly cross pollinated by wind (Polito, 1998). Walnut is a rich source of fat (64%) in which 49 per cent is poly unsaturated (PUFA) and 6 per cent is in form of Linolenic acid, protein (20.8%), carbohydrate (10.5%) and is concentrated source of energy (687 calories/100 gm). Kernals contain good amount of vitamin B1 (Thiamine 450 mg/100gm) (Anonymous, 2010a).Walnuts are commercially grown in 48 countries on an area of 66,58,966,00 hectares with a total production of about 16,70,109 MT (Anonymous, 2010b). The chief walnut producing countries are China (22%), USA (20%), Iran (12%), Turkey (10%) and India (2%). In India, the state of Jammu and Kashmir occupies an important position, as for as growing of walnuts is concerned, producing about 85 per cent of total production of walnuts of the country, and has monopoly in production of export quality walnuts. At present the state of Jammu & Kashmir has an area of about 93641 hectares with a production of 1,65024 MT, giving an average productivity of 1.89 metric tonnes/ha. The state enjoys prestigious position in fruit map of India on account of export of walnuts to foreign market, which brings an income of about 120 crore for the state exchequer (Anonymous, 2010c). The traditional walnut plantations in J&K consist of mature seedling trees characterized by long juvenile period, poor productivity with long period required

for sprouting of nuts and inconsistent fruiting behaviour. The germination of walnuts does not take place uniformly due to the fact that seeds are mainly obtained from seedling origin trees and from mixed nut populations. Secondly, during last few years, it has been observed that due to worldwide phenomenon of climate change, snowfall/or rainfall takes place during October-December, which otherwise used to be dry periods. The early precipitation does not allow nurserymen to go for seed sowing germination during normal sowing months i.e., November-December. Various chemical as well as seed treatment procedures have been tried in different countries so as to ensure uniform germination of walnut seeds.

The Persian walnut is difficult to propagate vegetatively than other fruit species. As a result, establishment of seedling orchards of walnut is common in several walnut producing countries. Seedling orchards suffer from high variability of nut yield and quality. Due to higher market demand for quality nuts and increased productivity, satisfactory methods of vegetative production in Persian walnut are needed (Vaddati, 2000). Secondly after 15-20 days accumulation of the phenolic compound like 4-hydroxynaphthoquinine is more harmful for growth of walnut callus (Prataviera *et al.*, 1983). Since entire walnut population of J&K is seedling origin, there is non availability of planting material of known pedigree. For vegetative propagation of walnut different methods had been tried in this institution so as to obviate the difficulty of raising walnuts vegetatively which includes various methods of budding and grafting, both through conventional means as well as high tech procedures like hot callusing technique, use of zero energy polyhouses and tissue culture. But this difficulty still remains to be addressed because of inconsistent results. Due to these facts availability of quality planting material to the farmers is not adequate as per the demand. Lately in some countries epicotyl grafting has been tried in different hard to propagate fruit species which has given some promising results in different fruit crops including nut crops like walnut and chestnut. Furthermore, walnut seed germination needs

to be re-evaluated to shorten the time of sprouting, by way of finding alternatives to compensate chilling requirement of seeds for germination. The study entitled “Studies on seed Germination and Epicotyl Grafting in Walnut (*Juglans regia L.*)” was therefore undertaken at the Farm of Division of Fruit Science, SKUAST-Kashmir, Shalimar, Srinagar, J&K with the following objectives :

- 1) To determine the effect of various chemical treatments in breaking seed dormancy, promoting germination and growth of walnut seedling.
- 2) To assess the feasibility of off-season sowing and germination of walnut seeds.
- 3) To evaluate the feasibility of using Epicotyl method of grafting as a procedure for vegetative propagation of walnut.

## Chapter – 2

### REVIEW OF LITERATURE

#### 2.1 Dormancy

Dormancy is the temporary suspension of growth of any plant structure containing a meristem, based on mechanism of seed germination (Amen, 1968, 1963). The germination obstacles vary both among and within species. It is essential to understand the effect of stratification and sowing time on germination both for practical nursery applications and conservation. One of the hypothesis advanced to explain dormancy is that levels of endogenous gibberellins are too low for growth to occur and levels of these increase during chilling treatment (Sinka and Lewak, 1970; Mathur *et al.*, 1971). Furthermore, each tree species has specific chilling requirement that are related to the accumulated hours below a chilling temperature threshold or cumulative chill units which are hours that are weighted for temperature effectiveness for breaking dormancy (Richardson *et al.*, 1974). Dormancy is the condition of bud or seed characterised by lack of visible growth (Westwood, 1988).The embryo of many seeds don't germinate because oxygen doesn't diffuse through the seed coat. At cold temperatures, more oxygen is soluble in water, so the oxygen requirement of embryo are better satisfied. Cold moist stratification imitates overwintering in field seedbed (Young and Young, 1992). The seed production is an important stage in any higher plant life's cycle with respect to its survival as a species. For this survival, the seed, mainly in a dry state, is well equipped to sustain extended periods of unfavourable conditions. To optimize germination over time, the seed enters a dormant state. Dormancy also prevents pre-harvest germination. Numerous studies have been performed to better understand how germination is controlled by various environmental factors. However, still very little is known about the process by which the embryo emerges from the seed to complete germination and how embryo emergence is blocked in dormant seeds (Bewley, 1997; Baskin *et al.*, 2002; Baskin and Baskin, 2004). The environmental conditions, such as light and temperature, are the most

important factors for plant growth and development. Temperature has a considerable influence throughout the development of a plant too (Koorneef *et al.*, 2002), stimulating developmental processes such as growth, seed dormancy and release, germination and vernalization (Heggie and Halliday, 2005). Many fruit and nut crops require cold temperature in winter to break dormancy. Considering the climatic changes which show rise in average air temperature all chilling models have predicated substantial decrease in winter chill at all sites. Across all sites between 1950 and 2050 mean chilling was predicated to decrease by 33 per cent (chilling Hours), 26 per cent (Utah Model), 16 per cent (Dynamic Model) and 14 per cent (Positive Utah Model). Research efforts are needed to identify most reliable chilling model and an alternative means for fruit and nut growers for imminent effects of climate change (Eike *et al.*, 2009).

## **2.2 Seed germination**

Seeds of numerous temperate fruit crops such as peach, plum, apple, pear and apricot require a definite period of stratification (moist chilling) for uniform germination (Hartmann and Kester, 1968). Eight to ten weeks of stratification period is found effective for uniform germination in almond cultivars (Grigorian, 1972). The germination after 10 days was 29 per cent in apricot and peach respectively. Cent per cent germination in self-fertile species *Prunus davidiana* was observed where as, to the extent of only 20 per cent in J.H. Hale a cross pollinated pollinator (Nagoo, 1975). Seedlings rootstocks of *Pistacia vera* made more lateral roots and thicker stems than the other species and they can reach budding size in a shorter time on having successful germination percentage (Sakoury, 1976). Thirty to 50 days of stratification period was found effective for early germination and *P. arabica* seeds were stratified for different durations and significantly high germination percentage was obtained at 45 Or 60 days, while non stratified seeds did not germinate (Grasselly, 1977). Variation in germination characteristics among 13 open- pollinated families of chokecherry (*P. virginiana*) seeds were observed, and the lowest germination was found to be 80 per cent in

three families; 83 per cent in four families; 93 per cent in four families; 97 per cent in two families with highest 100 per cent in two other families following sixteen weeks stratification period and nine weeks re-stratification period at 10-16 °C in different cultivars of plum viz Titron, late yellow, Alubukhara, Sharabati and Alpha, first two did not germinate and remaining gave maximum germination (Lockley, 1980). Lack of success has been attributed to poor germination in black walnut (*Juglans nigra* L.) and growing conditions in the field (Robison *et al.*, 1997).

Germination, in general, starts with uptake of water by the seed and ends with the (start of) elongation of the embryonic axis. It includes numerous events such as respiration, cell elongation, protein hydration and sub-cellular structural changes. Successful seed germination depends on numerous factors both internal and external. Internal factors are linked to those present in the seed structure (pericarp, seed coat and embryo) whereas external factors include those pertaining to the seed's environment, environmental factors can be classified in two groups: (1) chemical and (2) physical factors. In many cases, even when all conditions for germination are optimal, seeds still fail to germinate. Seeds that are in this situation are called dormant. This dormancy has to be broken before seeds can germinate (Bewley, 1997).

To achieve the required number of germinated seeds, walnuts were sown into flats and covered with growing media, following the recommended procedure for *Juglans* spp. of covering nuts to a depth of up to 7.62 cm. Seeds were examined regularly for germination, and those that germinated were removed from the flats and stored under moist towels at 2°C until target quantities of germinated walnuts were available. Minimal development of the radicle of germinated seeds was important to maintain uniform seedling development (Kujawski and Davis, 2001).

Peach seeds have a mechanical dormancy found in the seed coat and a physiological dormancy. To overcome the physiological dormancy, stratification

is required. These dormancy mechanisms ensure that the conditions are optimal for the peach seed so that when it germinates it has a high chance of survival (Dicenta and Martinez-Gomez, 2001).

Germination of black walnut (*Juglans nigra* L.) prior to sowing improves the likelihood of maintaining constant density in the nursery bed. In containerized seedling production, the number of containers and the amount of soil media needed are directly dependent upon the number of containers sown. Therefore, an increase in the percentage of cells containing successfully germinated seedlings would reduce production costs (Michler *et al.*, 2004).

Exogenous application of growth regulators such as auxins, gibberellins and cytokinins, and chemicals such as potassium nitrate or thiourea, have been shown to improve seed germination in fruit crops like peach, plum, apple and grapes (Nadjafi *et al.*, 2006).

For almond there was significant difference between the highest germination percentages from the experiments in 2004 to 2005 and 2005 to 2006 years and it was considered that the cause of this difference was not only the variation in climate conditions, but also endosperm lack of the seeds. For proper germination the seed needed adequate humidity, temperature and light to germinate (Gezer and Yucedag, 2006).

The highest seeds germination percentages (81.07) were found when the seeds were soaked for 12 hours in 200 mg l<sup>-1</sup> GA<sub>3</sub>, and (77.83%) for 12 hour in 100 mg GA<sub>3</sub>. This increase in seeds germination percentage might be related to the initial enzyme induction and to the activation of reserve food – mobilizing systems by Gibberellins which have also been used to enhance early germination (Nabil and Al-Imam, 2007).

“Alook” (*Prunus scoparia* Spach) and “Arjan” (*Prunus webbii* Vierh) are two wild almond species which grow naturally in vast area of Iran. Seeds of both

species covered by stony endocarp have very poor seed germination, an important problem in propagation of these species (Heidari *et al.*, 2008).

There have been few studies using different methods and techniques to overcome the germination obstacles of almond species (Rahemi *et al.*, 2009). Almond nuts should be boiled for ten minutes before sowing to obtain maximum germination (Nasir *et al.*, 2010).

## **2.3 Methods of improving seed germination**

### **2.3.1 Scarification**

Softening of endocarp by scarification hasten or increase germination in mazzard cherry {*Prunus avium*} (Zielinski, 1958) and almond *Prunus dulcis* (Gaudio and Pedone, 1963). The germination of blackberry increased upto 70 per cent by treating seeds with concentrated sulphuric acid for 30 minutes (Wenzel and Smith, 1975). The Peach x Almond hybrid (sloh) stones, treated with H<sub>2</sub>SO<sub>4</sub> increased germination from 40 to 65 per cent (Singh *et al.*, 1983). Scarification by concentrated Sulphuric acid and Stratification at 5+1 °C for 30 days increased seed germination in Pistachio spp. up to 90.72 per cent (Sehgal and Sing, 1990).

Seed scarification involves breaking, scratching or softening the seed coat so that water can enter and germination can begin, immersion of nuts in H<sub>2</sub>SO<sub>4</sub> for 30 minutes plus 30 days stratification increased the germination percentage of *Prunus scorparia* seeds (74.5%) and treating the endocarp of *Prunus webbii* seeds with H<sub>2</sub>SO<sub>4</sub> for 60 minutes plus stratification for 45 days, increased the germination about 73 per cent (Hartmann *et al.*, 1997a).

In acid scarification, seeds of almond nuts were scarified with concentrated H<sub>2</sub>SO<sub>4</sub> (70 and 90%) in order to dissolve the exocarp, mesocarp and endocarp. Treatment with H<sub>2</sub>SO<sub>4</sub> was effective in breaking the seed dormancy and Seeds soaked in concentrated sulphuric acid for 3 minutes gave the highest germination of 50 per cent. The treatment with 90 per cent concentration of the

acid for 3 min gave 28.6 per cent germination, No germination was recorded from seeds in the control for the period of the experiment (Nasir *et al.*, 2010).

The highest percentage germination of the hard endocarp seeds group was obtained by immersion of seeds in H<sub>2</sub>SO<sub>4</sub> for 10 minutes. Scarification can be another possibility which can increase seed germination. Stony endocarps exist in all members of *Prunus* species and seeds often have seed coat dormancy. Endocarp may also offer some resistance to germination and removal of the endocarp may hasten or increase germination in stone species (Heidari *et al.*, 2008).

### **2.3.2 Stratification**

Different seed pre-treatments were used to enhance Pistacia seed germination. Scarification and cold stratification were found to improve the seed germination (Ayfer, 1961). *Prunus* seeds are often thought to have only endocarp dormancy and required a certain period of cold treatment for germination of stony endocarps, even though the endocarp may offer some resistance to germination (Heit, 1967).

Seeds of temperate fruit crops such as peach, plum, apple, pear and apricot require a definite period of stratification (cold, moist chilling) for uniform germination (Hartmann and Kester, 1968). Stratification is the process of pre-treating seeds to simulate natural conditions that a seed must endure before germination. In nature, moist chilling occurs in wet soils combined with winter coldness. Removal of the stony endocarp or pericarp of fruits such as *Prunus* spp. often reduces the number of days of chilling needed for germination to occur in stone fruit (Grisez *et al.*, 1974).

The percent seed germination of all tested cultivars of walnut increased with the overall trend of increase in stratification period from one week to eight weeks (from 36.8% in control compared with 86.3% after eight weeks). Protease

and lipase enzymes have been shown to increase during chilling stratification (Ruduicki, 1977).

An optimum stratification period of 8 to 10 weeks in some almond cultivars, 30 to 50 days of stratification is required for 8 wild almond species. *P. Arabica* seeds were stratified for different durations and significantly high germination percentage was obtained at 45 or 60 days, while the non stratified ones did not germinate (Kester *et al.*, 1977; Grasselly, 1977).

Removal of seed coats in pome fruit such a *Malus* spp. and *Pyrus* spp. results in complete germination without chilling. However, seedlings obtained in this way are often stunted and their epicotyls will not grow until they are chilled (at 1-7 °C for 30-120 days) or treated with GA (Westwood, 1988).

Stratification of 4 weeks followed by pre-soaking treatments of seeds with gibberellic acid (GA) improved germination percentage and increased plant height in peach (Abo-Hassan, 1986).

Average seedling development (height and number of leaves) increased with GA<sub>3</sub> at 250 ppm + stratification for 3 weeks at 5°C treatment (Dahshan *et al.*, 1987). Peach (*P. dividiانا*) seeds have high ABA-levels and low GA levels. During stratification ABA-levels decreased and GA levels increased (Ji and Wang, 1987). *Prunus* seeds have embryo dormancy and require a period of after-ripening in the presence of moisture and oxygen to overcome it (Westwood, 1993).

Direct application of chemical on dry nut led to the best germination results (70%). Water pre-treatment produced a lesser entry of chemical solutions into seeds. In fact, gradient of humidity between chemical solutions like GA<sub>3</sub> and tissue of seeds ought to be bigger in no water treatment, therefore the effect of this extra chemicals would increase the growth parameters like height of seedling (Frutos, 1993).

The concentration of ABA like substances was high when Pistachio seeds were soaked for 12 hours in Gibberellic acid but decreased with the time of stratification (Ozguven *et al.*, 1995).

Germination of apricot seeds increased from 1.7 to 67.8 per cent when stratification period was increased from 0 to 63 days (Ercisli and Guleryuz, 1995). Stratified seeds of wild apricot had performed significantly higher mean germination (80.13%) in comparison to un-stratified seeds (74.54%). Potassium nitrate @ 0.4% treatment significantly improved the germination (91.24%) in comparison to all other treatments except potassium nitrate @ 0.2% treatment, which was statistically *at par* with this treatment. Interaction effect of stratification and chemical treatments also significantly influenced the germination of seeds. Seeds stratified first and then treated with potassium nitrate @ 0.4% exhibited highest germination (94.12%); however, least germination was observed in untreated and un-stratified seeds (Bhan and Sharma, 2007).

High germination percentage, seedling length and stem number in wild almond (*Prunus scoparia*) seeds were obtained after application of 500 ppm of gibberellic acid (Garcia-Gusano *et al.*, 2004). Percent of seed germination increased with the overall cold stratification in walnut seeds var. “Serr” and “Lara” (from 36.8 in the control compared with 86.3% after eight weeks). Also the mean time to germinate significantly decreased with the chilling treatment (Aslamarz *et al.*, 2007).

Pistachio seeds soaking for 12 hours significantly increased seedlings height and diameter as compared with seeds soaking for 24 hours. The highest seedling height (18.70 cm), seedling diameter (3.92mm) were found when the seeds soaked for 12 hours in 200 mg l<sup>-1</sup> GA<sub>3</sub> (Nabil and Al-Ameen, 2007).

Cold stratification was applied to the seeds of hazelnut. Seeds kept in water for 24 hours were planted into moist peat and incubated at 4°C for 100, 110 and 120 days which improved germination percentage upto 65-70 per cent

(Aygün *et al.*, 2008). Fully matured seeds/kernels of Pistachio were stratified for 60 days and then subjected to soaking in GA<sub>3</sub> and thiourea at different concentrations and combinations for 90 minutes before sowing in the nursery beds in polyhouse. The observations on seed germination percentage, number of leaves per seedling, seedling length, number of branches and roots were recorded. Among various treatments GA<sub>3</sub> @ 200 ppm + thiourea @ 10,000 ppm gave maximum seed germination percentage and seedling growth and root length as compared to other treatments and seeds which were only stratified (Negi *et al.*, 2010).

The mean chlorophyll (chlorophyll a & b) pigment levels ranged from a minimum of 0.34 mg/g for Fernor to a maximum of 2.37 mg/g for Pedro, cultivar of walnut. Chlorophyll A was determined predominant chlorophyll in total chlorophyll. The chlorophyll A pigment levels ranged from a minimum 0.28 mg/g for Fernor to a maximum of 1.95 mg/g for Pedro (Muradođlu and Gündođdu, 2011).

The average leaf area in walnut was found 53.02 cm<sup>2</sup>. This parameter ranged from a minimum of 26.77 cm<sup>2</sup> for Fernor cultivar to a maximum of 86.92 cm<sup>2</sup> for Howard cultivars. Generally the leaf area of foreign cultivars (Chandler, Fernor, Franquette, Hartley, Midland & Pedro) except Howard were found higher than domestic cultivars Kaman-1, Maraş-12, Maraş-18 & Şebin (Muradoglu and Gündođdu, 2011). The root length of the below-ground part of seedlings varied from 5.3 to 102.0 cm, with the amplitude of 96.7 cm. The greatest average root length was recorded in the seedlings from the locality '2', amounting to 45.6 cm, followed by the root length recorded at the locality '1' amounting to 43.4 cm, while the shortest root, of 40.4 cm on average was recorded in the seedlings from the locality '3'. There was no statistically significant difference between the localities, while the average root length for all seedlings was 43.3 cm (Tatjana *et al.*, 2011).

## 2.4 Use of chemicals

Gibberallic acid ( $GA_3$ ) in range of 50-200 mg/l resulted in significant breaking of dormancy in filbert seeds (Robert and Lagersteddt, 1969). Unshelled seeds of *P. terebinthus* were immersed in 50 ppm  $GA_3$  for seven days, the germination increased from 50-79 per cent (Casini and Conticni, 1979). In a study *Pistacia vera*, *P. khinjuk* and *P. atlantica* had more than 95 per cent germination after removing green nut hulls and soaking in 100 ppm  $GA_3$  solution for 7 days before sowing (Dahab *et al.*, 1975).

Non-chilled Nemaguard peach seeds treated with gibberellic acid at 50-500 mg/l gave 80 per cent germination (Mehana *et al.*, 1985). In another study it was found that peach seeds treated with Gibberellic acid and no cold storage had a final germination percentage of 80 but when they were treated with Gibberellic acid and stratified the germination percentage jumped to 100 (Martin *et al.*, 1985).

A number of plant growth substances are reported to be effective chiefly by way of overcoming the problem of impermeability of seed coats to water and dissolved gases particularly oxygen and carbon dioxide (Hartman and Kester, 1989). In another study, it was found that the highest germination percentage (73.3%) was obtained when seeds were soaked for 48 hours in 125 ppm  $GA_3$  solution (Ak *et al.*, 1995). More than 95 per cent seed germination was obtained in *Pistacia* species after seeds were soaked in for 12 hours in 100 ppm  $GA_3$  solution (Kuru and Aksu, 1995).

The highest germination rates (73.3%) were obtained with 125 ppm  $GA_3$  and 48 hr soaking in water (Ak *et al.*, 1995). The germination rate of *Pistacia* seeds ranged from 40 to 96 per cent after the seeds were exposed to different treatments. The highest germination rates were obtained with stratified seeds (Kafkas and Kaska, 1998).

Paradox walnut (*J. hindii* x *J. regia*) seeds treated with Gibberellic acid and potassium nitrate had increased percentage of germination and also resulted in stronger subsequent seedling growth (Hamed and Fathi, 1999). Mean time to complete germination of *P. atlantica*, *P. terebinthus* and a hybrid was minimum in seeds treated with 1000 ppm GA<sub>3</sub> and stratified at 4 °C for 15-45 days (Isfendiyaroglu and Ozeker, 2002). In another study, it was found that the highest germination percentage (73.3%) was obtained when seeds were soaked for 48 hours in 125 ppm (Hartmann *et al.*, 1997; Grisez *et al.*, 1974). Soaking seeds of almond in gibberellic acid (GA<sub>3</sub>) with concentrations of 750, 1000 or 1500 ppm increased germination percentage significantly when combined with stratification (Khalil and Al-Eisawi, 2000).

Germination in *Pistacia* species (*Pistaciavera* 'Mateur', *P. atlantica*, *P. terebinthus* and *P. lentiscus*) was positively affected by immersion in concentrated sulphuric acid. The increase in seeds germination percentage might be related to the initial enzyme induction and to the activation of reserve food-mobilizing systems by Gibberellins which have also been used to enhance germination and stimulate early seedling emergence and growth (Hopkins and Hüner, 2004).

The highest (84.78) percentages of seed germination in apricot was found with GA<sub>3</sub> at 250 ppm and stratification for 3 weeks at 5°C (Samaan *et al.*, 2000). Highest germination (100%) in (*Prunus mahleb* L.) was obtained from 75 ppm application, followed by 50 ppm (92.1%) and 100 ppm (84.7%) GA<sub>3</sub>. Germination speed was also determined in GA<sub>3</sub> treatments. The fastest germination occurred in 75 ppm application followed by 50 ppm and 25 ppm applications (Aygün *et al.*, 2008).

The best results in *Prunus mahaleb* L. seeds were obtained from stratification for 12 weeks at 2-4 °C + GA<sub>3</sub> at 1000 ppm (Gerçekcioglu and Cekic 1999). Fully matured seeds/kernels were stratified for 60 days and then subjected to soaking in GA<sub>3</sub> and thiourea at different concentrations and combinations for 90 minutes before sowing in the nursery beds in Polyhouse. The

observations on seed germination percentage, number of leaves per seedling, seedling length, number of branches and roots were recorded. Among various treatments GA<sub>3</sub> @ 200 ppm + thiourea @10,000 ppm gave maximum seed germination percentage (80.4), seedling growth and root length in pistachio nut as compared to other treatments and seeds which were only stratified (Negi *et al.*, 2008).

Pistachio seed soaking for 12 hrs + 100 ppm GA<sub>3</sub> significantly increased seedlings diameter (3.13 mm) as compared with seed soaking for 24 hours 2.77mm (Nabil and Al-Imam, 2007). Many attempts have been made to use GA<sub>3</sub> for improving seed germination and subsequent seedling growth of plants (Yousif *et al.*, 1984) on *Pistacia vera* L. seeds; (Al-Fawaier, 1994) on *Pistacia atlantica* seeds; (Beyhan, *et al.*, 1999) on hazelnut seeds; (Abdullah and Younis, 2002) on *P. atlantica* and *P. terebinthus*; and (Al-Imam and Al-Brifkany, 2006) on three cultivars of hazelnut seeds; (Abdullah and Younis, 2002) on *P. atlantica* and *P. terebinthus*. To improve germination of wild almond seeds were immersed in different concentrations of sulphuric acid for 10 and 60 minutes in combination of stratification for 15, 30 and 45days and immersion in sulphuric acid for 10 minutes in combination of stratification for 30 days and has improved germination percentage of *Prunus* spp. to 94 and 82.5 per cent, respectively (Heidari *et al.*, 2008). Gibberellin and GA is an important endogenous growth regulator having profound and diverse effects on plant growth and development. One of the roles of gibberellin is the induction of seed germination and promotion of radical elongation and mobilization of endosperm reserves during early embryo growth, as well as flower and fruit development (Hopkins, 1999; Peng and Harberd, 2002; Hartmann *et al.*, 2002).

Soaking seeds for 12 hours gave the highest seed germination percentage (88.50), seedling height (16.72 cm), stem diameter (3.25 mm), Which significantly differ from seeds soaked for 24 hours in Pistachio nut (Nabil and Al-Imam, 2007). The increase in seedling height with GA might be related to the fact that

GA promote stem and shoot elongation through the increase of both cell division and from internodes elongation in higher plant (Hopkins and Hüner, 2004; Harris *et al.*, 2004).

Chloroplasts in organs of plants developing in shade tend to have more photosynthetic pigments per unit volume than chloroplasts developing under high irradiance (Haehnel, 1984). However, the ratio of chlorophyll a and chlorophyll b may be altered by a change in light or irradiance subsequent to leaf development and expansion (Burkey and Wells, 1991).

## **2.5 Experiment No. 2 : Epicotyl grafting in walnut (*J. regia*)**

The multiplication of fruit trees including nuts through vegetative propagation is as old as human civilization. The new technologies in fruit science have forced the Horticulturists, all over the world, to make intensive use of new vegetative methods like tissue culture, hypocotyl and epicotyl grafting for propagation of horticultural plants. Walnut cultivars are principally propagated by traditional budding and grafting methods. One of the biggest obstacles with budding and grafting of walnut during the growing season is the detrimental effect of sap flow (bleeding) on bud and graft-take. Inconsistent results encountered with conventional grafting and budding of walnuts compelled researchers to conduct trials under controlled as well as open conditions involving epicotyl grafting. Lack of good success in vegetative propagation of walnuts could be attributed to the presence of Juglone in the rootstock and scion wood which is harmful for callus formation (Prataviera *et al.*, 1983).

Poor callus formation in walnut made it a difficult to propagate fruit species (Coggeshall and Beineke, 1997). Many factors, involving genetic and environmental, affect plant growth and development. Plants are able to modify their growth, development and physiology according to their environment. This ability of plants has a plays a key role in determining their tolerance to stress and

their maintaining efficient growth (Murchie and Horton, 1997; Walters *et al.*, 2003).

## **2.6 Epicotyl grafting**

The nursery seed grafting was first described by Moore (1964). In this method twigs with dormant buds of chestnut cultivars were used as scion material. Once winter dormancy had been met, seedlings quickly grown were utilized for grafting in containers at temperature around 25 °C (Moore, 1964).

The hypocotyl and epicotyl grafting as modifications of nurse seed grafting were used and described by Park (1968). The success with Nut grafting ranged from 0 to 90 per cent depending on size of seedling used for grafting. (McKay and Jaynes, 1969; Keys, 1978). Sixty to 100 per cent graft success was achieved using a modified chip bud method on epicotyls. Newly germinated chestnut seedling were used as root stocks, twigs with dormant buds were used as scion material. Once the seedlings were 2 cm long, grafting was carried and scions of thicker size were inserted into the stock seedling (Ackerman and Jayne, 1980). Cent percent graft success was achieved in the greenhouse and 83 per cent in the open field using cleft graft method on epicotyl graft (Sawano *et al.*, 1983). Seventy per cent success was observed when epicotyl grafting in mango was performed from July to September in glass house, whereas it was only 40 per cent in open in the month of July (Desai and Patil, 1984). Epicotyl grafting of mango in Jammu district revealed 50-75 per cent graft success (Gupta *et al.*, 1988).

The flush grafting in cashew with four different ages of scions (21, 28, 35 and 42 days) on five different ages of rootstocks (14, 21, 28, 35 and 42 days) was carried out at Bhubaneshwar. Highest success (45%) was noticed in combination of 28 days flush with 45 days stock followed by 21 days old scion and 35 day old stock (Anonymous, 1990).

Grafting of scion on the germinating seedling in epicotyl grafting has been successfully used as an efficient, economic and rapid method for propagation of mango (Lal, 1993; Singh and Srivastava, 1981; Srivastava, 1989).

Thirty per cent success with 14 genotypes on large nuts (20-25 g) was achieved in chestnut using nurse grafting (Balta and Basaran, 2001). Temperature had a pronounced effect on callus formation and optimum temperature for callusing varies between temperate zones (Hartmann *et al.*, 2002). Sixty three to 75 per cent graft success was achieved on epicotyls using cleft method of grafting, generally less on other methods (Duman and Serdar, 2005).

Swelling of union may be caused by mis-matched stock and scion sizes or from the genetic interaction of different stock-scion combinations (Serdar *et al.*, 2005). The survival rate of Epicotyl grafting in walnut ranged from 65 to 87 per cent according to grafting time, the average rate was 78.9 per cent (Suk-In, 2006). Seventy per cent success was achieved in hypocotyl grafting of walnut using one year old scion (Vahdatii and Zareie, 2008). Cleft grafting was conducted with one-year old scion on 10 to 15 day old seedlings of walnut. Scion thickness corresponded to the thickness of new shoot of the rootstock. Polyethylene band was used for tying to provide good aeration at the place of grafting. After 12 to 16 days the grafted plants started developing new shoots (Gandev, 2009). The highest rates for graft success and plant survival were obtained from shaded greenhouse conditions in the European chestnut (Ozturk and Serdar, 2009). When walnut seeds formed a well developed root and a juvenile epicotyl, split grafting was performed with 5-8 cm long one year old scions. Survival rate shown to range from 51.7 to 75.00 per cent (Gandew and Arnaudov, 2011).

## **2.7 Influence of factors on epicotyl grafting in walnut**

### **2.7.1 Time factor**

There are number of factors which influence the healing of graft union in which weather conditions, especially the temperature and humidity play a very

crucial role in the success of grafting (Hartman and Kester, 1979). Epicotyl grafting trials in cashew, conducted during 1980-82 at Vengurla (Maharashtra) revealed that this method can be commercially exploited under agro climatic conditions and February to May was the congenial period with 62.4 to 67.7 per cent success on 10 day old seedlings (Sawke, 1983). The success of epicotyl grafting in cashew using 15 days old seedling rootstocks was studied where the highest success was recorded between June and August (60-68%) which declined to 45-47 per cent in November (Nagabhushanam, 1983). Significantly more survival percentage (83.33 and 63.33) of graft after 4 months of grafting was noticed in mango when grafting was performed on 7-day old rootstock followed by 9-day old rootstocks. Only 2.38 percent graft on 15-day old rootstock survived after 4 months of grafting (Gagandeep and Malhi, 2003). The survival rate was shown in the range from 65 to 87.5 per cent according to grafting time. The rate in February was greater than that in March and April. The epicotyl grafting in walnut can be performed all the times or seasons of the year under controlled temperature and humidity, and if sound scion could be sampled (Suk-In, 2006). The climatic analysis made showed that the air temperature characteristics of June-August of experiment 2009 give grounds to make conclusion that in Bulgaria the successful hypocotyl grafting of walnuts is possible under unconditioned surroundings (Gandev, 2009).

### **2.7.2 Temperature and humidity**

Temperature has a pronounced effect on the production of callus tissue and optimum temperature range required for callusing varies between different temperature zone fruit species (Hartmann *et al.*, 2002). Optimum temperature for walnut to produce callus is about 27 °C (Sitton, 1931). Later studies confirmed that temperatures around 26 to 27 °C are optimum, but temperatures down to 22°C could still be sufficient for good callus formation of walnut (Rongting and Pinghai, 1993). Below 20 °C callus formation in walnut becomes unsatisfactory (Reil *et al.*, 1998; Hartmann *et al.*, 2002). Temperature did not only influence the

amount of callus tissue but also the rate of callus formation. At 22 °C, callus was initiated six days after grafting, while it took five days at 27 °C. When temperature was further further increased to 32 °C, callus initiated after only four days, but less callus tissue was produced at this temperature (Rongting and Pinghai, 1993). A temperature of 27 °C±2 has become the standard utilized by many propagators and researchers all over the world to graft walnut successfully (Lagerstedt, 1981; Lantos, 1990).

Optimum humidity of the immediate environment of grafted trees was regarded as vital, this was achieved by maintaining the correct moisture content of the medium in callus containers. Generally, a high relative humidity of 80±10% is preferred (Cerny, 1965; Germain *et al.*, 1999). An experiment was conducted at Madakkathara to find out the effect of mist chamber (90% humidity, max temperature 27 °C and min temperature 23 °C) on sprouting in flush grafting of cashew with suitable control. It was found that mist had definite effect on reducing the time taken for sprouting and also enhanced sprouting percentage (Anon, 1989b). A scion sprouting percentage of 71.7 per cent was observed under controlled conditions of temperature and humidity (Dhuria *et al.*, 1977). While studying the standardisation of suitable media and interval for walnut propagation under controlled conditions (Gandev and Arivindov, 2011) found that three weeks time in saw dust was most suitable for keeping the grafts in incubator at 25±1°C and 75±2 relative humidity and then planted in the field. With this technique, a graft success of 60.22 to 64 per cent was achieved.

Walnut graft union was wrapped with aluminum foil to standardise the heat and was maintained at a temperature 27 °C for a period of 24 days. A success of 73 per cent was achieved through heating of graft unions (Avanzato and Tampani, 1988).

Studies on effect of scion moisture, substrate humidity, environmental temperature, scion state and cultivar on callus formation in walnut was observed and the scion moisture content was the key factor influencing callus formation and

graft survival. Eighty five per cent success was obtained by using bark grafting coupled with surrounding the grafts with moist saw dust (Xi and Ding, 1993). One year old pot grown seedlings of walnut were subjected to different environments after top grafting and was found that plant growth efficiency was positively influenced by increasing the relative humidity around the leaf apparatus (Avanzato, 2001). Callus formation in walnut graft is closely related to biochemical and hormonal conditions of tissues depending on ecological factors like temperature and relative humidity. The biochemical structure, Juglone and phenolic contents of rootstock and scion are main causes of graft failure (Kuniyuki and Forde, 1995). The environmental conditions during and following grafting are factors influencing the healing of the graft union and plant growth (Hartmann *et al.*, 1990).

The changes in the length and diameter of the graft shoot, leaf number, and the mean and total leaf areas per plant of two chestnut genotypes (SE 3-12 and Marigoule), were influenced by temperature, shade and open field nursery conditions. It was also determined whether the length and diameter of the graft shoot of chestnuts depend on the leaf number, mean and total leaf areas per plant (Chapa and Verlhac, 1978).

Maximum sprouting percentage with younger (5-10 days) rootstocks was observed in mango. The reason behind better performance of the young rootstock producing more number of sprouted grafts seems to be high meristematic activity in just germinating seedlings and higher reserved food material in the cotyledons (Dhunaga *et al.*, 1989). Grafting of deciduous plant species can only be performed while both rootstock and scion material are in a dormant condition (Farmer, 1973).

Various authors evaluated the influence of different grafting times during the dormant period on the success of walnut grafting. In Oregon, USA grafting of various species, including walnut is preferred in mid December to mid January (mid dormancy) rather than February and March (late dormancy to early

vegetative period) (Lagerstedt, 1982). Epicotyl grafting in cashewnut, conducted at vengurla (Maharashtra) revealed that this method can be commercially exploited under coastal conditions. February to May was the congenial period with 62.4 to 67.7 per cent success on 10 day old seedling (Sawke, 1983).

An investigation was undertaken to assess the effect of season and age of the rootstock on success of grafting in Cashewnut varieties V-4 and V-5 and the most appropriate age of rootstock for cashew grafting was 45 days (Shingre *et al.*, 2003). Epicotyl grafting can be done successfully on 15 day old seedling during June to November by adopting cleft grafting technique in Cashew. Grafting success was highest (60-68%) between June and August and declined (45-47%) in November under South Kanara conditions of Karanataka (Nagabhushnam, 1983). Percentage of success in softwood grafting of cashew varied in different agroclimatic regions, seventy per cent success was obtained in the month of April (Ghosh, 1990).

Poor root system quality is the foremost influential factor on post planting graft failure in various fruit species and accentuated the selection of high quality rootstocks (Lagerstedt, 1882). Seedling of Persian walnut (*J. regia*) are most often used as rootstocks in grafting of walnut (Pieniasek, 1972; Avanzato and Tamponi, 1988). Better results were achieved with *J. regia* compared to *J. nigra* (Lantos, 1990). Northern Californian black walnut (*J. hindsii* Jeps. Rehder) seedlings were also utilized (Pieniasek, 1972). The healing rate of one year old seedling is better than two year old seedling (Vahdati and Zareie, 2008).

### **2.7.3 Height of grafting**

The seedling of (Persian walnut) 5 days old were grafted at height of 2 and 4 cm from the ground and the was cut off vertically, one year old scion stick was inserted on same centre of cut with cleft grafting method. Survival rate was shown to range from 65 to 87.5 per cent (Suk-In *et al.*, 2006).

Nurse grafting of chestnut was performed on 2cm long radicle after germination. The radicle including 1-2 mm of its base, was cut off and the scion was cut into wedge- shape and was inserted into the seed (Duman and Serdar, 2006). The non-lignified stem of chestnut was is cut-off at height of 4-7 cm and the bottom of scion was cut-off and was inserted into preformed cut on stem. Survival rate shown to range from 80 to 100 per cent (Sawano *et al.*, 1983). Cleft grafting at height of point of attachment of cotyledons was conducted with one-year old scions on 10 to 15-day-old new shoots of germinating walnut fruit. The survival rate was 77.5 per cent (Gandev, 2009)

#### **2.7.4 Scion material**

Grafting success depends also on the correct timing for collection of scion material. Scion-wood be must cut in late autumn before severe frost is experienced (Tsurkan, 1990). Scion material can be collected on same day of grafting or cold stored for several days (Erdogan, 2006). Scion cultivar had no effect on grafting success (Rezaee and Vahdati, 2008) but contrary to (Rongting and Pinghai, 1993) who reported that graftsuccess depends upon scion cultivar.

Scion quality mainly affects graft success rather than their genetic makeup and scions were used for bench grafting of walnuts containing two or three buds, 7 to 15 cm in length and 25mm in diameter (Vahdati and Zareie, 2008). Scion material was stored in cold storage at 4°C prior to grafting (Achim and Botu, 2001). Insufficient callus formation by scion was found to be main cause of graft failure in walnuts. Of various growth substances tested, auxins and kinetins were found to promote callus formation (Hartmann and Kester, 1973). Scions from upper, middle and lower parts of walnut shoots gave 94.59, 91.82 and 86.46 per cent respectively. The scion wood was collected during dormancy and kept in cold storage (Anadoliev, 1975). Effects of scion moisture, substrate humidity, environmental temperature on callus formation was studied. Results revealed that the uniting process included five stages, i). no callus formation ii). callus initiation iii). callus junction, iv. cambium differentiation and connection v). vascular

differentiation and connection. Scion moisture content was the main factor influencing callus formation and graft survival (Xi and Ding, 1993).

### **2.7.5 Graft cover**

The polythene covering of the grafts proved to be superior to other covering materials in achieving maximum scion sprouting and scion take per cent. In fact, polythene covering maintained the optimum moisture content for successful uniting of scion and stock. Eighty percent success was recorded under temperature of 27°C with polythene covering (Stritzke, 1959).

### **2.7.6 Grafting technique**

Cleft grafting gave 63-73 percent success under open conditions, whereas, whip and tongue grafts had less success (Mittempergher, 1963). Saddle, cleft and bark saddle grafting have been found suitable methods for walnut propagation. The optimum conditions for callus formation and union were found to be at a temperature of 25 to 27°C and relative humidity of 70-80 per cent. A sharp drop in temperature below 10-15C greatly reduced the “take” percent (Komanic, 1967). Good percentage of success and field survival was achieved with cleft grafting 67-72 per cent by direct field planting. Field survival was also highest about 56 percent (Dhuria *et al.*, 1977).

## Chapter – 3

### MATERIAL AND METHODS

The present investigation entitled “Studies on Seed Germination and Epicotyl Grafting in Walnut (*Juglans regia* L.)” was undertaken in the Fruit Plant Nursery, Division of Fruit Science, Shere-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Shalimar, Srinagar, J&K during 2011 and 2012. The site is situated at an elevation of 1630 meters above mean sea level having latitude 34°-80' N and longitude 74°-08' E.

Research work was confined to 2 separate experiments. The material and methods employed in programme are presented here under:

#### **3.1 Experiment No. 1 : Studies on seed germination and seedling growth in walnut**

##### **Treatments**

##### **a) Storage Environment**

- i) Cold storage treatment (3-5 °C) for 30 days.
- ii) Dry storage under ambient conditions (10-12°C)

##### **b) Seed treatment**

Walnut seeds were divided into two lots. Each lot was divided into two sub lots. Half of seeds were soaked in water for 12 hours and half of seeds were used without soaking for treatment with chemicals.

- i) GA<sub>3</sub> at 50 ppm and 100 ppm for 12 hours
- ii) Ethrel 1000 ppm and 1500 ppm for 12 hours.

- iii) Sulphuric acid 7.5 and 9.5 per cent for 10 minutes
- iv) Potassium nitrate 0.2 and 0.4 per cent for 10 minutes
- v) Control (water soaked and dry)

Plot size : 20 nuts

Replications : 3

### c) **Sowing of seeds**

The seeds were sown in polythene bags containing uniform rooting medium of sand, soil and organic manure in the ratio of 1: 1:1 respectively. Seeds were placed approximately 7.5 cm deep in punched polythene bags and kept in the open field.

#### **3.1.1 Selection of walnut seed**

Walnut seeds were collected from a single tree which were freshly harvested and used for experimentation during 2011 and 2012.

#### **3.1.2 Storage of walnut seed**

Selected walnut seeds were divided into two main lots, one lot of seeds was stored in cold temperature (3-4 °C) in a refrigerator for 30 days and second lot was kept under ambient conditions.

#### **3.1.3 Preparation of chemicals**

The chemical solutions were made according to formula  $N_1V_1=N_2V_2$

$N_1$  = Strength of stock solution

$V_1$  = Volume to be taken

$N_2$  = Required concentration

$V_2$  = Volume made

#### **3.1.3.1 Gibberellic acid**

1 gm of gibberellic acid was dissolved in 10 ml of ethanol. To this solution distilled water was added to make volume upto 1000ml. From this stock solution further dilutions of 50 ppm and 100 ppm were made accordingly.

#### **3.1.3.2 Ethrel**

4 ml of ethrel was taken in a volumetric flask and volume was made upto 1000 ml by adding distilled water which forms 4000 ppm stock solution. Further dilutions of 1000 ppm and 1500 ppm were made from this stock solution.

#### **3.1.3.3 Sulphuric acid**

10 ml was diluted in 90 ml of distilled water. From this solution further dilutions of 7.5 and 9.5 per cent were made accordingly.

#### **3.1.3.4 KNO<sub>3</sub>**

1 gm of KNO<sub>3</sub> was dissolved in 100 ml of water. From this stock solution 0.2 and 0.4 per cent was made with further dilutions accordingly.

#### **3.1.3.5 Seed treatment**

Walnut seeds were divided equally into two lots, each lot was further subdivided into two sub-lots. One of the lots was kept in refrigerator at 3-4 °C for 30 days for cold treatment and another lot was kept at ambient conditions. Cold stored lot was equally divided into two parts, one part was soaked in water for 12 hours and after soaking seeds were dipped in different chemicals. GA<sub>3</sub> and ethrel

treatments were given for 12 hours, sulphuric acid and potassium nitrate for 10 minutes. Another part of cold stored walnut seeds were directly dipped into chemicals without soaking. Walnut seeds kept at ambient conditions were also equally divided into two parts. One part was soaked in water for 12 hours and after soaking seeds were dipped into chemicals. Another equal part of nuts was dipped into chemicals without soaking.

### **3.1.4 Observations recorded**

#### **3.1.4.1 Germination of seed (%)**

The germination of seeds were recorded periodically. The seeds were considered germinated when plumules just began to be visible on the surface. Water was applied as a spray to emerging seedlings for ensuring optimum moisture level. The spray was used once a day with the help of sprayer.

#### **3.1.4.2 Height of seedling (cm)**

Height of seedling was recorded at the end of growing season. The height of seedling was measured from collar region to the apex. The height was measured with digital Vernier's Caliper's in centimetres.

#### **3.1.4.3 Stem diameter (mm)**

The stem diameter was measured with the help of Vernier's Caliper's in millimetres (mm)

#### **3.1.4.4 Root length (mm)**

Polythene bags of each sub-plot were cut-off and kept in water for some time in order to make roots free from rooting medium. Then seedling's were uprooted carefully without disturbing root system. The roots were washed under

tap water and excess water was removed. Roots were kept in stretched manner during measurement. The root length was measured with the help of Vernier's Caliper's in millimetres (mm).

#### **3.1.4.5 Number of secondary roots**

Secondary roots emerging from the tap roots were counted and expressed in numbers.

#### **3.1.4.6 Number of leaves**

Walnut compound leaves were counted at the end of growing season.

#### **3.1.4.7 Leaflet size (cm<sup>2</sup>)**

A random sample of 10 leaflets was taken from each treatments. The leaf area was measured with the help of systronics 211 leaf area meter, care was taken that leaflets were fully expanded without overlapping during measurement. The total leaf area was used to calculate average leaflet size in cm<sup>2</sup>.

$$\text{Leaf area (cm}^2\text{)} = \frac{\text{Total leaf area of samples}}{\text{Total number of leaflets}}$$

#### **3.1.4.8 Leaf chlorophyll (mg/g of fresh weight)**

The leaflets of walnut seedlings were picked from middle portion of leaves in mid July.

#### **Procedure**

Collected leaflet samples from middle portion of leaves and weighed 100 mg sample on digital balance. This sample was chaffed in Pistle bowl. Removed chaffed sample with spatula into 50 ml flasks then 20 ml of DMSO was added.

Flasks were wrapped and were kept at hot oven at 65<sup>0</sup>C for 5 days. Absorbance was measured at 645nm and 663nm using at Spectronics UV-viz 130 Spectrophotometer. A blank of pure DMSO was included in each run. The absorbance of this blank was subtracted from the absorbance readings of each sample before any calculations have been made.

For estimation of chlorophyll (mg/g fresh weight) following formula was used :

$$\text{Total chlorophyll (mg/g fresh weight)} = \frac{20.2 A_{645} + 8.02 A_{663}}{A \times 1000 \times W} \times V$$

Where,

A = The length of light path (usually 1 centimeter)

V = Volume of extract

W = Fresh weight of sample

A<sub>645</sub> = Absorbance at 645nm

A<sub>663</sub> = Absorbance at 663nm

#### **3.1.4.9 Days from germination to leaf fall**

The germination of walnuts was observed keenly and the dates were recorded accordingly till germination was almost over. Simultaneously at 80 per cent leaf shedding dates of leaf fall were recorded. The total days were counted and average mean for each treatment was recorded.

#### **3.1.4.10 Statistical Analysis**

Statistical Analysis of Experiment No. 1 (SPD) with 36 treatment combinations and three replications was done in Split Plot Design. The Storage Treatments constitute main plot while as Chemical Treatments constitute sub-plot.

### **3.2 Experiment No. 2 : Study on Epicotyl grafting of walnut under open temperature conditions**

The experiment was conducted in the fruit plant nursery of the Division of Fruit Science at Sher-e-Kashmir University of Agricultural Sciences and Technology, Shalimar campus, Srinagar, J&K. Seedlings of uniform growth and age was raised from walnuts sown in punched polythene bags and used as rootstocks. The graft wood was collected from healthy, strong and disease free tree during dormancy in last week of February to first week of March which were stored in refrigerator. Same were used as scion material for epicotyls grafting. The experiment was laid-out as detailed below :

#### **Treatment combinations**

##### **a) Height of grafting**

- i) At the point of attachment of cotyledons
- ii) 2 cm above point of attachment of cotyledons
- iii) 4 cm above the cotyledon
- iv) 6 cm above the cotyledon

##### **b) Time of grafting**

- i) 15 days after germination
- ii) 30 days after germination
- iii) 45 days after germination

**c) Scion wood used**

Graft wood of previous season s growth comprising of one to two buds was used as scion material. The graft wood was used consisted of

- 1) Terminal portion
- 2) Basal portion

Design of the experiment : Completely Randomised Design (CRD)  
Replications : 3  
Experimental plot size : 10 plants  
Treatment combinations :  $2 \times 4 \times 3 = 24$

**3.2.1 Selection of rootstock**

Seedling (Persian walnut) rootstocks was raised from nuts sown in early spring 2011 and 2012 and used for epicotyl grafting. The young 15, 30 and 45 day old seedlings were used as rootstock.

**3.2.2 Scion material**

Selection of proper scion material is very crucial for getting high percentage of graft success. One year old scion wood of about 5-10 cm length with thickness of 3-5 mm containing 2-3 buds were collected from healthy, disease free trees in the first week of March 2011 and 2012. Scion wood collected was wrapped in polythene and were kept in refrigerator until time of epicotyl grafting.

### **3.2.3 Time of grafting**

Grafting was performed in three sets after germination of walnut seed. First set of grafting was performed when seedlings attained age of 15days (5<sup>th</sup> May). Second set of grafting was performed when seedlings attained age of 30 days (20<sup>th</sup> May) and third set was performed when seedlings got 45 days old(First of June).

### **3.2.4 Height of grafting**

Four different heights of rootstocks were used. Simple scale was used to measure height of newly germinated rootstocks.

- i. At Point of attachment of cotyledons.
- ii. 2 cm above from point of attachment of cotyledons
- iii. 4 cm above from point of attachment of cotyledons
- iv. 6 cm above from point of attachment of cotyledons

### **3.2.5 Method of grafting**

Epicotyls were cut at heights of 0, 2, 4 and 6 cm. The non lignified epicotyl was split vertically in the form of cleft to a length of 3 cm downwards with surgical blade. A scion stick of about same thickness was used for grafting. At the lower cut end of scion, about 3 cm was mended to form wedge using sharp knife without injuring cambial tissues. The wedge shaped scions were inserted into the cleft of the stocks. Polyethylene band was used for tying at the place of grafting.

### **3.2.6 Observations recorded**

#### **3.2.6.1 Scion take percentage**

The grafts were counted after every 15 days. The 15 day average of scion take percentage was calculated for whole grafted plants in the experiment. The scion take percentage was calculated on the basis of total number of grafts prepared in each treatment combinations.

#### **3.2.6.2 Scion sprouting percentage**

The number of successful grafts were counted after the complete sprouting and commencement of new shoot growth on scion at 15, 30 and 45 days after grafting. The scion sprouting percentage was worked out on the basis of total number of grafts prepared in each treatment combination.

#### **3.2.6.3 Scion growth (cm)**

The length of new shoot growth, developed or initiated from the scion stick grafted was recorded at end of growing season with the help of Vernier's Caliper's in centimetres.

#### **3.2.6.4 Number of leaves**

The number of functional leaves produced on grafted plants was counted at end of growing season.

#### **3.2.6.5 Leaflet size (cm<sup>2</sup>)**

A random sample of 10 leaflets was taken from each of treatments. The total area of random samples was measured with the help of a Systronics211 leaf area meter. Care was taken that samples were fully expanded without overlapping during measurement. The total leaflet area was used to calculate average leaf size in cm<sup>2</sup>.

$$\text{Leaflet area (cm}^2\text{)} = \frac{\text{Total leaflet area of ten leaves}}{\text{No. of leaflets taken (10)}}$$

### **3.2.6.6 Plant height (cm)**

The height of seedling was recorded at the end of season. The height of seedling was measured from collar region to the apex in centimetres.

### **3.2.6.7 Stem diameter (mm)**

Stem diameter was measured with the help of Vernier's Caliper's in millimetres (mm).

### **3.2.6.8 Statistical analysis**

Statistical analysis of experiment No. 2 consisted of 24 treatment combinations was done in completely randomized block design (CRD). O.P STAT statistical package for agricultural research workers was used}.

## Chapter – 4

### EXPERIMENTAL FINDINGS

The results of the present investigation entitled “Studies on Seed Germination and Epicotyl Grafting in Persian walnut” are presented in this chapter. The observations were recorded on the basis of three replications and the results are interpreted at 0.05 per cent level of significance for the year 2011 and 2012.

#### 4.1 Studies on seed germination and seedling growth in walnut.

##### 4.1.1 Germination percentage

The perusal of data on germination of walnut seeds (Table-1 & 2) indicate that the chemical and storage treatments had a significant effect on the germination percentage during 2011 and 2012.

The data for the year 2011 (Table-1) indicates that among the chemical treatments given to walnut seeds, GA<sub>3</sub> at 100 ppm resulted in highest seed germination (58.24%) followed by GA<sub>3</sub> at 50 ppm (54.23%). Sulphuric acid (7.5%) recorded 49.66 per cent, sulphuric acid (9.5%) 47.99 per cent and Potassium nitrate (0.4%) recorded 44.24 per cent. Amongst the chemical treatments lowest germination (35.58%) was observed with the application of ethrel at 1000 ppm which was significantly higher when compared to control 29.99 per cent.

Amongst the storage treatments cold storage resulted in significantly higher germination (46.51%) in comparison to the ambient storage 42.21 per cent. Significantly higher germination (45.36%) was also recorded under un-soaked seed treatment than the soaked treatment (43.36%).

The perusal of data in Table-1 further indicates that the interactions between chemical x storage, seed treatment x storage and chemical x seed treatments were statistically significant. Maximum germination (63.49%) was

recorded with the treatment combination of GA<sub>3</sub> at 100 ppm and 30 days chilling. Significantly higher germination (47.73%) was recorded with treatment combination of un-soaked treatment and cold storage.

Interaction between chemical x seed treatment reveal that the seed treatment with GA<sub>3</sub> at 100 ppm under un-soaked conditions recorded significantly higher germination (59.33%). The interactions between chemical x storage and seed treatment were also statistically significant. Higher percentage of germination was recorded in GA<sub>3</sub>100 ppm under cold storage with un-soaked seed treatments (65.33%).

The data on influence of chemical and storage treatments for the year 2012 (Table-2) reveals that among the chemical treatments GA<sub>3</sub> at 100 ppm recorded maximum germination (58.57%) followed by GA<sub>3</sub> 50 ppm (56.32%). Sulphuric acid (7.5%) was also effective and recorded 48.57 per cent germination. Amongst chemical treatments the lowest germination (38.74%) was recorded with the application of ethrel at 1000 ppm which was however significantly higher when compared to control (29.38%). Cold storage treatment was found more effective and produced higher germination (46.56%) than the ambient storage (42.79%). Un-soaked seed treatment recorded (45.53%) comparatively higher germination than the soaked treatments (43.83%).

The perusal of data in Table-2 reveals that interactions between chemical x storage, seed treatment x storage and chemical x seed treatment were statistically significant. Interaction of chemical and seed storage indicates that GA<sub>3</sub> at 100 ppm under one month cold storage recorded highest germination (62.66%). Un-soaked seed treatment with GA<sub>3</sub> 100 ppm recorded significantly higher germination (59.66%) than GA<sub>3</sub> at 100 ppm and seeds soaked in water (57.49%). However, germination percentage obtained with GA<sub>3</sub> (50 ppm) under un-soaked seed treatment was at par with GA<sub>3</sub> at 100 ppm with soaking. The interaction between storage x chemical and seed treatment was significant during the second year as well. Highest germination (63.66%) was recorded in cold stored seeds

**Table-1 : Influence of chemical and storage treatments on percentage germination of walnut seed during the year 2011**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	61.66	53.53	57.59	51.66	50.33	50.99	54.23	56.66	51.83
	100 ppm	65.33	61.26	63.29	53.33	52.66	52.99	58.24	59.33	57.16
Etherel	1000 ppm	38.33	35.33	36.83	35.33	33.33	34.33	35.58	36.83	34.33
	1500 ppm	40.66	36.66	38.66	38.33	36.66	37.49	38.07	39.49	36.66
H <sub>2</sub> SO <sub>4</sub>	7.5 %	53.33	51.66	52.49	48.33	45.33	46.83	49.66	50.83	48.49
	9.5 %	51.66	50.33	50.99	46.66	43.33	44.99	47.99	49.16	46.83
KNO <sub>3</sub>	0.2 %	43.33	41.66	42.49	41.66	38.33	39.99	41.24	42.49	39.99
	0.4 %	46.66	45.33	45.99	43.33	41.66	42.49	44.24	44.99	43.49
Control	-	28.26	31.16	30.16	28.33	31.33	29.83	29.99	28.49	31.49
Mean		47.73	45.29	46.51	42.99	41.44	42.21		45.36	43.36

**C.D(p≤0.05)**

Storage (S)	:	2.05
Chemical (C)	:	2.11
Seed treatment (ST)	:	1.01
C x S	:	2.32
ST x S	:	2.05
ST x C	:	2.08
S x C x ST	:	2.37

**Table-2 : Influence of chemical and storage treatments on percentage germination of walnut seed during the year 2012**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	60.60	56.66	58.63	54.33	53.66	53.99	56.32	57.46	55.16
	100 ppm	63.66	61.66	62.66	55.66	53.33	54.49	58.57	59.66	57.49
Etherel	1000 ppm	40.33	37.66	38.99	37.33	35.33	36.33	38.74	38.83	36.49
	1500 ppm	41.66	40.66	41.16	39.66	37.66	38.66	38.82	40.66	39.16
H <sub>2</sub> SO <sub>4</sub>	7.5 %	52.66	50.33	51.49	47.66	43.66	45.66	48.57	50.16	46.99
	9.5 %	50.66	48.33	49.49	45.33	41.66	43.49	46.49	47.99	44.99
KNO <sub>3</sub>	0.2 %	43.33	40.66	41.99	40.33	38.33	39.33	40.66	41.83	39.49
	0.4 %	46.33	43.33	44.83	43.33	42.33	42.83	43.83	44.83	42.83
Control	-	28.33	31.33	29.83	28.13	31.53	29.83	29.38	28.23	31.13
Mean		47.51	45.64	46.56	43.55	42.03	42.79		45.53	43.83

**C.D(p≤0.05)**

Storage (S)	:	2.01
Chemical (C)	:	2.09
Seed treatment (ST)	:	0.99
C x S	:	2.28
ST x S	:	2.01
ST x C	:	2.04
S x C x ST	:	2.32

under un-soaked seed treatment with GA<sub>3</sub>100 ppm. The lowest germination (28.13%) was recorded in ambient stored seeds with un-soaked seed treatment without (control) any chemical treatment.

#### **4.1.2 Height of seedling (cm)**

The perusal of data for the year 2011 (Table-3) reveals that among the chemical treatments given, GA<sub>3</sub> (100 ppm) recorded maximum height (23.24 cm) followed by GA<sub>3</sub> at 50 ppm (20.24 cm). Sulphuric acid (7.5%) recorded 19.62 cm, sulphuric acid (9.5%) 18.48 cm and potassium nitrate (0.4%) recorded 18.01 cm. Amongst the chemical treatments minimum height (13.50 cm) was observed with the application of ethrel at 1000 ppm which was significantly higher when compared to control (12.78cm).

Among the storage treatments cold storage resulted in significantly higher stem length (21.28 cm) in comparison to the ambient storage (16.83cm). Significantly higher stem length (17.78 cm) was also recorded under un-soaked seed treatment than the soaked seed treatment (16.93 cm).

The data presented in table-3 further reveals that the interactions between chemical x storage, seed treatment x storage and chemical x seed treatment were statistically significant. Interaction of chemical x storage recorded maximum height (24.17cm) with GA<sub>3</sub> 100 ppm and 30 days cold storage followed by GA<sub>3</sub> 100 ppm (22.31 cm) under ambient storage. Among the chemical x storage interaction the minimum height was observed in ethrel 1000 ppm under ambient storage (13.25 cm) which was significantly higher when compared to control under ambient storage (12.56 cm).

Interaction between seed treatment x storage had a significant effect on height and maximum height (18.66 cm) was observed in un-soaked seed treatment under cold storage. The minimum height (16.76 cm) was recorded in the interaction of seed treatment x storage under ambient storage with soaked seed treatments.

Seed treatment and chemical interaction was found effective in promoting height and maximum height (23.83 cm) was observed in GA<sub>3</sub> 100 ppm with un-soaked seed treatment which was 22.16 cm in GA<sub>3</sub> 100 ppm in soaked seed treatment. Among the interaction of seed treatment x chemical the minimum height (12.85 cm) was recorded in control under un-soaked seed treatment.

The interaction between chemical x storage x seed treatment was also statistically significant and maximum stem length (25.35 cm) was observed in GA<sub>3</sub>100 ppm under cold storage in un-soaked seed treatment. Minimum height (12.09 cm) was observed in control at ambient storage under un-soaked seed treatment.

The data on influence of chemical and storage treatments for the year 2012 (Table-4) reveal that among the chemical treatments GA<sub>3</sub> at 100 ppm recorded maximum height (23.00 cm) followed by GA<sub>3</sub> 50 ppm (20.31cm). Sulphuric acid (7.5%) was also effective and recorded 19.55 cm stem height. Amongst chemical treatments the minimum height (13.40 cm) was recorded with the application of ethrel 1000 ppm which was, however, significantly higher when compared to control (12.90 cm). Cold storage treatment was found more effective and produced maximum height (17.93 cm) than the ambient storage (16.99 cm).Un-soaked seed treatment gave significantly higher height in walnut seedlings (17.79 cm) than the soaked seed treatments (16.96 cm).

The perusal of data in Table-4 further indicates that interactions between chemical x storage, seed treatment x storage and chemical x seed treatment were statistically significant. Interaction of chemical x seed storage indicates that GA<sub>3</sub> at 100 ppm under one month cold storage recorded maximum height (24.27 cm) whereas in GA<sub>3</sub> 100 ppm under ambient storage seedling height was 21.73 cm. Un-soaked treatment with 30 day cold treatment recorded significantly better height (18.68 cm) in comparison to the soaked treatment under ambient storage (16.75 cm).Un-soaked seed treatment with GA<sub>3</sub> 100 ppm was observed to give

**Table-3 : Influence of chemical and storage treatments on height (cm) of walnut seedling during the year 2011**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	22.85	19.72	21.28	19.21	19.20	19.21	20.24	21.03	19.46
	100 ppm	25.35	22.99	24.17	22.31	21.34	22.31	23.24	23.83	22.16
Etherel	1000 ppm	15.02	14.83	13.92	13.37	13.20	13.25	13.50	14.19	13.81
	1500 ppm	15.62	12.52	14.07	13.43	13.49	13.38	13.70	14.52	13.97
H <sub>2</sub> SO <sub>4</sub>	7.5 %	19.89	18.61	19.25	19.99	19.28	19.99	19.62	19.94	18.94
	9.5 %	19.40	18.06	18.73	18.23	19.02	18.23	18.48	18.81	18.54
KNO <sub>3</sub>	0.2 %	18.00	17.44	17.74	17.29	16.98	17.29	17.51	17.64	17.21
	0.4 %	18.86	17.96	18.40	17.62	17.54	17.62	18.01	18.24	17.75
Control	-	12.91	13.12	13.01	12.09	13.02	12.56	12.78	12.85	13.23
Mean		18.66	17.11	21.28	16.91	16.76	16.83		17.78	16.93

**C.D(p≤0.05)**

Storage (S)	:	0.13
Chemical (C)	:	0.17
Seed treatment (ST)	:	0.26
C x S	:	0.24
ST x S	:	0.31
ST x C	:	0.32
S x C x ST	:	1.13

**Table-4 : Influence of chemical and storage treatments on height (cm) of walnut seedling during the year 2012**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	23.09	19.63	21.36	19.21	19.17	19.27	20.31	21.15	19.40
	100 ppm	24.91	23.64	24.27	22.31	21.12	21.73	23.00	23.61	22.38
Etherel	1000 ppm	15.74	12.73	14.23	12.37	12.18	12.28	13.40	14.05	12.45
	1500 ppm	15.29	13.58	14.43	12.43	12.36	12.43	13.43	13.86	12.97
H <sub>2</sub> SO <sub>4</sub>	7.5 %	19.72	18.84	19.28	19.99	19.32	19.83	19.55	19.85	19.08
	9.5 %	19.13	18.32	18.72	18.23	19.08	18.75	18.73	18.68	18.70
KNO <sub>3</sub>	0.2 %	18.59	17.29	17.94	17.29	17.06	17.22	17.58	17.94	17.17
	0.4 %	18.92	17.73	18.32	17.62	17.45	17.56	17.94	18.27	17.59
Control	-	12.77	13.49	13.13	12.79	13.03	12.68	12.90	12.78	13.26
Mean		18.68	17.18	17.93	16.91	16.75	16.99		17.79	16.96

**C.D(p≤0.05)**

Storage (S)	:	0.10
Chemical (C)	:	0.17
Seed treatment (ST)	:	0.23
C x S	:	0.21
ST x S	:	0.28
ST x C	:	0.28
S x C x ST	:	1.10

significantly higher height (23.61 cm) than GA<sub>3</sub> at 100 ppm and seeds soaked in water (22.38 cm).

The interaction between storage, chemical and seed treatment was statistically significant during the second year as well. Maximum height (24.91 cm) was recorded in cold stored seeds under un-soaked treatment with GA<sub>3</sub>100 ppm. Among the interactions the minimum height (12.77 cm) was recorded in control under ambient stored seeds with un-soaked seed treatment.

#### **4.1.3 Stem diameter (mm)**

The data for the year 2011 (Table-5) indicates that among the chemical treatments given to walnut seeds, GA<sub>3</sub> at 100 ppm resulted in highest stem diameter(4.75 mm) followed by GA<sub>3</sub> at 50 ppm (4.19 mm) and sulphuric acid 7.5 per cent (4.06 mm). Amongst the chemical treatments lowest stem diameter (3.70 mm) was observed with the application of ethrel 1000 ppm which was statistically significant when compared to control (2.80 mm).

Among the storage treatments cold storage resulted in higher stem diameter (4.09 mm) in comparison to the ambient storage (3.53 mm). Un-soaked seed treatment recorded higher stem diameter (4.01 mm) which was 3.83 mm in case of soaked seed treatment.

The perusal of data in Table-5 further reveals that the interactions between chemical x storage, seed treatment x storage and chemical x seed treatments were effective in promoting stem diameter. Maximum stem diameter (5.10 mm) was recorded in GA<sub>3</sub>100 ppm under 30 days chilling. The next higher stem diameter (4.40 mm) was observed in GA<sub>3</sub> 100 ppm under 30 day ambient treatment. Among interaction of chemical x storage the stem diameter (3.43 mm) observed in ethrel 1000 ppm under ambient storage was significantly higher when compared to control (2.69 mm).

Significantly higher stem diameter (4.23 mm) was recorded with un-soaked seed treatment under cold storage. Among interactions of seed treatment x

storage the lowest stem diameter (3.73 mm) was observed in ambient storage under soaked seed treatment.

Interaction of chemical x seed treatment reveals that the seed treatment with GA<sub>3</sub> at 100 ppm under un-soaked seed treatment recorded significantly higher stem diameter (5.19 mm) followed by GA<sub>3</sub> 100 ppm under soaked seed treatment (4.31 mm). The stem diameter recorded in 1000 ppm ethrel (3.61 mm) was significantly higher when compared to control (2.70 mm) under un-soaked seed treatment.

The interactions between chemical, storage and seed treatment were also statistically significant. Higher stem diameter was recorded in GA<sub>3</sub>100 ppm under cold storage treatment with un-soaked seed (5.71 mm). Among the interaction of chemical x seed treatment x storage the lowest stem diameter (2.59 mm) was observed in control at ambient storage under un-soaked seed treatment.

The data on influence of chemical and storage treatments for the year 2012 (Table-6) indicates that among the chemical treatments GA<sub>3</sub>100 ppm recorded maximum stem diameter (4.92 mm) followed by GA<sub>3</sub> 50 ppm (4.04 mm). Sulphuric acid 7.5 per cent was also effective and recorded 3.88 mm stem diameter. Amongst chemical treatments the lowest stem diameter (3.23 mm) was recorded with the application of ethrel 1000 ppm which was however, significantly higher when compared to control (2.63 mm).

Cold storage treatment was significant in promoting stem diameter (3.93 mm) in comparison to the ambient storage (3.44 mm). Un-soaked seed treatment recorded stem diameter of 3.80 mm which was 3.57 mm in case of soaked seed treatment.

The perusal of data in Table-6 reveals that interactions between chemical x storage, seed treatment x storage and chemical x seed treatment were statistically significant. Interaction of chemical and seed storage indicates that GA<sub>3</sub> at 100 ppm under one month chilling recorded highest stem diameter (5.53 mm).

**Table-5 : Influence of chemical and storage treatments on stem diameter (mm) of walnut seedling during the year 2011**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	4.29	4.12	4.20	4.01	3.98	3.99	4.19	4.15	4.05
	100 ppm	5.71	4.49	5.10	4.68	4.13	4.40	4.75	5.19	4.31
Etherel	1000 ppm	4.01	3.93	3.97	3.57	3.39	3.43	3.70	3.79	3.61
	1500 ppm	4.20	3.99	4.09	3.62	3.40	3.56	3.80	3.91	3.74
H <sub>2</sub> SO <sub>4</sub>	7.5 %	4.52	4.05	4.28	3.92	3.79	3.85	4.06	4.22	3.92
	9.5 %	4.40	4.12	4.26	3.80	3.71	3.75	4.00	4.10	3.91
KNO <sub>3</sub>	0.2 %	3.79	3.50	3.64	3.72	3.69	3.70	3.67	3.75	3.59
	0.4 %	4.41	3.88	4.14	4.10	3.50	3.80	3.97	4.25	3.69
Control	-	2.82	3.03	2.91	2.59	2.80	2.69	2.80	2.70	2.91
Mean		4.23	3.93	4.09	3.79	3.73	3.53		4.01	3.83

**C.D(p≤0.05)**

Storage (S)	:	0.21
Chemical (C)	:	0.17
Seed treatment (ST)	:	0.13
C x S	:	0.19
ST x S	:	0.24
ST x C	:	0.27
S x C x ST	:	0.32

**Table-6 : Influence of chemical and storage treatments on stem diameter (mm) of walnut seedling during the year 2012**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	4.32	4.21	4.26	4.04	3.61	3.82	4.04	4.18	3.91
	100 ppm	5.63	5.43	5.53	4.98	3.64	4.31	4.92	5.30	4.53
Etherel	1000 ppm	3.42	3.39	3.40	3.10	3.03	3.06	3.23	3.26	3.21
	1500 ppm	4.01	3.98	3.99	3.71	3.36	3.53	3.76	3.86	3.67
H <sub>2</sub> SO <sub>4</sub>	7.5 %	4.16	3.97	4.06	3.86	3.55	3.70	3.88	4.01	3.76
	9.5 %	3.99	3.84	3.91	3.49	3.21	3.35	3.63	3.74	3.52
KNO <sub>3</sub>	0.2 %	3.65	3.42	3.53	3.21	3.04	3.12	3.32	3.43	3.23
	0.4 %	3.87	3.79	3.83	3.39	3.07	3.23	3.53	3.63	3.43
Control	-	2.82	2.83	2.86	2.66	2.14	2.52	2.69	2.63	2.48
Mean		4.00	3.87	3.93	3.60	3.27	3.44		3.80	3.57

**C.D(p≤0.05)**

Storage (S)	:	0.18
Chemical (C)	:	0.20
Seed treatment (ST)	:	0.15
C x S	:	0.16
ST x S	:	0.26
ST x C	:	0.14
S x C x ST	:	0.30

The next higher stem diameter (4.31 mm) was observed in GA<sub>3</sub> 100 ppm under ambient storage treatment. Among interaction of chemical x storage the lowest stem diameter (2.52 mm) was recorded in control under ambient storage.

Seed treatment x storage interaction recorded highest stem diameter (4.00 mm) in un-soaked seed treatment under cold storage. Lowest stem diameter was recorded in soaked seed treatment under ambient storage (3.27 mm).

Un-soaked seed treatment with GA<sub>3</sub>100 ppm recorded significantly higher stem diameter (5.30 mm) than GA<sub>3</sub> at 100 ppm and seeds soaked in water (4.53 mm).

The interaction between storage x chemical and seed treatment were significant during the second year as well. Highest stem diameter (5.63mm) was recorded in cold stored seeds under un-soaked seed treatment with GA<sub>3</sub>100 ppm. The lowest stem diameter (2.14 mm) was recorded in ambient stored seeds with soaking treatment without any chemical treatment.

#### **4.1.4 Root length (mm)**

The data on influence of chemical and storage treatments on root length of walnut seedlings for the year 2011 (table-7) reveal that among the chemical treatments GA<sub>3</sub> 100 ppm recorded maximum root length (191.57 mm) followed by GA<sub>3</sub> 50 ppm (181.24mm), sulphuric acid 7.5 per cent (167.66 mm), sulphuric acid 9.5 per cent (154.16 mm) and potassium nitrate 0.4 per cent (132.74 mm). Amongst chemical treatments the lowest root length (103.50 mm) was observed with the application of ethrel 1500 ppm which was significantly higher when compared to control (93.24 mm).

Among the storage treatments cold stored seeds resulted significantly higher root length (148.36 mm) which was 134.82 mm in case of ambient stored seeds. Significantly higher root length (144.92 mm) was also recorded under un-soaked seed treatment than the soaked seed treatments (138.27mm).

The perusal of data (Table-7) further indicates that the interactions between chemical x storage, seed treatment x storage and chemical x seed treatments were statistically significant. Maximum root length (198.49 mm) under interaction of chemical x storage was observed in GA<sub>3</sub> 100 ppm and 30 days chilling. The next higher root length (190.99 mm) was recorded with the application of GA<sub>3</sub> 50 ppm and 30 days cold treatment. Among the interaction of chemical x storage the lowest root length (91.48 mm) was recorded in control (untreated seed) under ambient storage.

In the interaction of seed treatment x storage maximum root length (150.03 mm) was recorded with the treatment combination of un-soaked treatment and cold storage followed by root length (146.69 mm) recorded in soaked seed treatment under cold storage. Among seed treatment x storage interaction the lowest root length (129.84 mm) was recorded in soaked seeds under ambient storage.

Seed treatment x chemical interaction recorded significantly higher root length (196.16 mm) with the application of GA<sub>3</sub> 100 ppm under un-soaked seed treatment. The next higher root length (188.00 mm) was observed with the application of GA<sub>3</sub> 50 ppm under un-soaked seed treatment. Among seed treatment x chemical interaction lowest root length (83.50 mm) was recorded with control under soaked seed treatment.

The interaction between chemical, storage and seed treatment were also statistically significant (Table-7). Higher root length was recorded in GA<sub>3</sub> 100 ppm under cold storage treatment with un-soaked seed treatment (201.66 mm). Among the interactions between chemical x seed treatment x storage the lowest root length (84.66) was recorded in control under ambient storage treatment with un-soaked seeds.

The data on the influence of chemical and storage treatments on root length of walnut seedlings for the year 2012 (Table-8) reveal that among

**Table-7 : Influence of chemical and storage treatments on root length (mm) of walnut seedling during the year 2011**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	193.33	188.66	190.99	182.66	160.33	171.49	181.24	188.00	174.50
	100 ppm	201.66	195.33	198.49	190.66	178.66	184.66	191.57	196.16	187.00
Etherel	1000 ppm	151.66	133.66	142.66	115.33	114.66	114.99	128.82	133.50	124.16
	1500 ppm	104.66	100.33	102.49	106.66	102.33	104.49	103.50	108.66	100.33
H <sub>2</sub> SO <sub>4</sub>	7.5 %	171.33	169.33	170.33	167.66	162.33	164.99	167.66	169.50	165.83
	9.5 %	162.66	160.33	161.49	162.33	131.33	146.83	154.16	162.50	145.83
KNO <sub>3</sub>	0.2 %	136.33	130.33	133.33	126.66	114.33	120.49	126.91	131.50	122.33
	0.4 %	143.33	137.66	140.49	131.66	118.33	124.99	132.74	137.50	128.00
Control	-	85.33	104.66	94.99	84.66	98.33	91.48	93.24	85.00	83.50
Mean		150.03	146.69	148.36	139.80	129.84	134.82		144.92	138.27

**C.D(p≤0.05)**

Storage (S)	:	3.32
Chemical (C)	:	5.42
Seed treatment (ST)	:	2.02
C x S	:	2.84
ST x S	:	3.52
ST x C	:	3.11
S x C x ST	:	3.67

**Table-8 : Influence of chemical and storage treatments on root length (mm) of walnut seedling during the year 2012**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	192.66	190.66	191.66	187.66	165.66	176.66	184.16	190.16	178.16
	100 ppm	203.33	198.66	200.99	195.33	182.33	188.83	194.91	199.33	190.50
Etherel	1000 ppm	155.66	140.33	147.99	130.66	125.33	127.99	137.99	143.16	132.83
	1500 ppm	119.33	111.33	115.31	114.33	112.21	113.27	114.29	111.83	104.00
H <sub>2</sub> SO <sub>4</sub>	7.5 %	186.66	180.33	183.49	173.66	168.66	171.16	177.32	180.16	174.50
	9.5 %	181.66	175.33	178.49	164.66	149.66	157.16	167.82	173.16	162.50
KNO <sub>3</sub>	0.2 %	149.33	141.66	145.49	130.33	122.33	126.33	135.91	139.83	132.00
	0.4 %	158.66	143.33	150.99	137.66	129.33	133.49	142.24	148.16	136.33
Control	-	86.33	105.33	95.83	85.33	98.33	91.83	93.83	85.83	98.83
Mean		159.29	154.10	156.69	145.51	137.58	141.55		152.40	145.84

**C.D(p≤0.05)**

Storage (S)	:	3.39
Chemical (C)	:	2.47
Seed treatment (ST)	:	2.09
C x S	:	2.89
ST x S	:	3.59
ST x C	:	3.42
S x C x ST	:	3.72

chemicals maximum root length was recorded in GA<sub>3</sub>100 ppm (194.91 mm) followed by GA<sub>3</sub> 50 ppm (184.16 mm). Sulphuric acid was also effective and recorded 177.32 mm root length. Among chemical treatments the lowest root length (114.29 mm) was recorded with the application of ethrel 1500 ppm which was however significantly higher when compared to control (93.83 mm).

Cold storage treatment was found effective in promoting root length and produced significantly higher root length (156.69 mm) than ambient storage (141.55 mm). Seed treatment also had a significant effect on root length and higher root length (152.40 mm) was recorded in un-soaked seed treatment which was only 145.84 mm in case of soaked seed treatment.

The interaction of chemical x seed storage (Table-8) indicates that GA<sub>3</sub> 100 ppm under one month cold storage recorded highest (200.99 mm) root length. The next higher root length (191.66 mm) was observed in GA<sub>3</sub> 50 ppm under one month cold treatment. Among the interaction of chemical x seed storage the lowest (91.83 mm) root length was recorded in control under ambient storage.

Seed treatment x storage had a significant effect on root length and highest root length (159.29 mm) was recorded with 30 days cold under un-soaked treatment which was 137.58 mm in case of soaked seed treatment under ambient storage.

Seed treatment x chemical interaction had also a significant effect on root length and highest (199.33 mm) root length was recorded in GA<sub>3</sub> 100 ppm under un-soaked seed treatment. The next higher root length (190.16 mm) was recorded under un-soaked seeds in GA<sub>3</sub> 50 ppm. Among the interaction of seed treatment x chemicals the lowest (85.83mm) root length was recorded with control under un-soaked seed treatment.

The interactions between chemical x storage and seed treatment were also statistically significant. Highest root length (203.33 mm) was recorded in GA<sub>3</sub> 100 ppm under cold storage treatment with un-soaked seeds. The next higher

(195.33 mm) root length was obtained in GA<sub>3</sub> 100 ppm under ambient storage with un-soaked seed treatment. Among the chemical x seed treatment x storage interactions the lowest (85.13 mm) root length was recorded in control under ambient storage with un-soaked seeds.

#### **4.1.5 Number of secondary roots**

The perusal of data in Table-9 reveal that all chemical and storage treatments were statistically significant, as far as their influence on number of secondary roots of walnut seedling is concerned. Among the chemical treatments GA<sub>3</sub> 100 ppm recorded maximum (27.49) number of secondary roots followed by GA<sub>3</sub> 50 ppm (25.99) and sulphuric acid 7.5 per cent (21.49). Amongst the chemical treatments the lowest (18.39) number of secondary roots was observed with the application of ethrel 1500 ppm which was significantly higher when compared to control (15.24).

Among the storage treatments cold stored seeds resulted in higher (21.18) number of secondary roots than the ambient stored seeds (20.16). Significantly higher (21.02) number of secondary roots was also recorded under un-soaked seed treatments than the soaked seeds (20.32).

The perusal of data (Table-9) further indicates that the interactions between chemical x storage, seed treatment x storage and chemical x seed treatment were statistically significant. Maximum secondary roots (28.33) under interaction of chemical x storage were observed in GA<sub>3</sub> 100 ppm under 30 days cold storage. The next higher (26.99) number of secondary roots was recorded with the application of GA<sub>3</sub> 50 ppm under 30 days cold treatment. The lowest (14.99) number of secondary roots was recorded in control under cold storage treatments.

Interaction of seed treatment x storage recorded maximum (21.66) number of secondary roots with un-soaked seed treatment under cold storage followed by 20.69 number of secondary roots in soaked seed treatment under cold storage. In

seed treatment x storage interaction the lowest (19.94) number of secondary roots were recorded in soaked seed treatments under ambient storage.

Seed treatment x chemical interaction recorded significantly higher number of secondary roots (28.00) with the application of GA<sub>3</sub> 100 ppm under un-soaked seed treatment. The next higher (27.00) was observed with the application of GA<sub>3</sub>100 ppm under soaked seed treatment. Among seed treatment x chemical interaction the lowest (15.50) number of secondary roots was recorded in control under soaked seed treatment.

The interaction between chemical, storage and seed treatment was also statistically significant (Table-9). Higher secondary roots (29.33) were recorded in GA<sub>3</sub> 100 ppm under cold storage treatment with un-soaked treatments. Among the interactions between chemical x seed treatment x storage the lowest number of secondary roots (14.33) was recorded in control under cold storage treatment with un-soaked seeds.

The data on the influence of chemical and storage treatments on number of secondary roots for the second year (Table-10) reveal that among the chemical treatments maximum number of secondary roots was recorded in GA<sub>3</sub> 100 ppm (27.74) followed by GA<sub>3</sub> 50 ppm (25.74). Sulphuric acid was also effective and recorded 22.24 number of secondary roots. Among chemical treatments the lowest number of secondary roots (18.42) was recorded with the application of ethrel 1500 ppm which was however, significantly higher when compared to control (14.91).

Cold storage treatment was found effective in promoting number of secondary roots and produced higher number of secondary roots (22.29) than ambient storage (19.45). Seed treatment also had a significant effect on number of secondary roots and higher number of secondary roots (21.23) was recorded under un-soaked seed treatment than the soaked seeds 20.51.

**Table-9 : Influence of chemical and storage treatments on number of secondary roots of walnut seedling during the year 2011**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	27.33	26.66	26.99	25.33	24.66	24.99	25.99	26.33	25.66
	100 ppm	29.33	27.33	28.33	26.66	26.66	26.66	27.49	28.00	27.00
Etherel	1000 ppm	19.43	18.33	18.83	18.86	18.33	18.59	18.66	19.20	18.34
	1500 ppm	19.33	18.23	18.63	18.18	18.22	18.15	18.39	19.17	18.28
H <sub>2</sub> SO <sub>4</sub>	7.5 %	22.66	21.66	22.16	21.33	20.33	20.83	21.49	22.00	21.00
	9.5 %	21.66	19.66	20.66	18.92	19.02	18.97	19.81	20.29	19.34
KNO <sub>3</sub>	0.2 %	20.66	18.33	19.49	19.33	18.33	18.83	19.16	19.83	18.50
	0.4 %	20.33	20.33	20.33	18.66	18.66	18.66	19.49	19.66	19.50
Control	-	14.35	15.60	14.99	15.66	15.33	15.49	15.24	15.60	15.50
Mean		21.66	20.69	21.18	20.38	19.94	20.16		21.02	20.32

**C.D(p≤0.05)**

Storage (S)	:	0.48
Chemical (C)	:	1.28
Seed treatment (ST)	:	0.63
C x S	:	1.55
ST x S	:	1.29
ST x C	:	0.48
S x C x ST	:	1.42

**Table-10 : Influence of chemical and storage treatments on number of secondary roots of walnut seedling during the year 2012**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	28.66	26.33	27.49	24.33	23.66	23.99	25.74	26.50	25.00
	100 ppm	29.43	28.66	28.99	27.66	25.33	26.49	27.74	28.50	27.00
Etherel	1000 ppm	21.33	19.66	20.99	17.88	16.90	17.77	18.88	19.50	18.26
	1500 ppm	20.66	19.33	19.49	16.33	16.69	16.16	18.42	18.51	18.16
H <sub>2</sub> SO <sub>4</sub>	7.5 %	24.33	23.66	23.99	20.66	20.33	20.49	22.24	22.50	22.00
	9.5 %	22.33	21.66	21.99	19.33	18.33	18.83	20.41	20.83	20.05
KNO <sub>3</sub>	0.2 %	20.66	21.13	20.99	18.66	17.33	17.99	19.49	19.66	19.33
	0.4 %	21.66	21.33	21.49	18.99	18.34	18.66	20.07	20.33	19.84
Control	-	14.66	15.16	15.26	14.76	14.16	14.66	14.91	14.06	15.16
Mean		22.62	21.95	22.29	19.83	19.07	19.45		21.23	20.51

**C.D(p≤0.05)**

Storage (S)	:	0.51
Chemical (C)	:	1.32
Seed treatment (ST)	:	0.54
C x S	:	1.51
ST x S	:	1.27
ST x C	:	0.29
S x C x ST	:	1.39

The interaction of chemical x seed storage indicates that GA<sub>3</sub> 100 ppm under one month cold storage recorded highest (28.99) number of secondary roots. The next higher (27.49) was observed in GA<sub>3</sub> 50 ppm under one month cold treatment. Among the interaction of chemical x seed storage the lowest (14.66) number of secondary roots was recorded in control under ambient storage.

Seed treatment x storage interaction had a significant effect on number of secondary roots and highest (22.62) was recorded with 30 days cold under un-soaked treatment and lowest (19.07) in case of soaked seeds under ambient storage.

Seed treatment x chemical interaction had a significant effect on number of secondary roots and highest (28.50) number of secondary roots was recorded in GA<sub>3</sub> 100 ppm under un-soaked seed treatment. The next higher number of secondary roots (27.00) was recorded under soaked seeds in GA<sub>3</sub> 100 ppm. Among the interaction of seed treatment x chemicals the lowest (14.06) number of secondary roots was recorded with control under un-soaked seed treatment.

The interactions between chemical x storage and seed treatment were also statistically significant. Highest (29.43) number of secondary roots were recorded in GA<sub>3</sub> 100 ppm under cold storage with un-soaked seed treatment. The next higher (28.66) number of secondary roots was obtained in GA<sub>3</sub> 50 ppm under cold storage with un-soaked seed treatment. Among the chemical x seed treatment x storage interactions the lowest (14.16) number of secondary roots was recorded in control under ambient storage with soaked seed treatment.

#### **4.1.6 Number of leaves**

The perusal of data (Table-11) reveal that chemical treatments had a significant effect on number of leaves during 2011. Maximum number of leaves (6.22) was obtained with the application of GA<sub>3</sub> 100 ppm followed by sulphuric acid 7.5 per cent (5.65). Amongst chemical treatments the lowest number of

leaves (3.56) was obtained in ethrel 1000 ppm which was higher when compared to control (3.47).

The storage treatments had a non significant effect on number of leaves during 2011. However maximum number of leaves (4.96) was observed under cold storage treatment which was 4.46 in case of ambient storage treatment. Seed treatment was also found non- significant, however, maximum number of leaves (4.85) was observed under un-soaked treatment in comparison to soaked seed treatment (4.57).

The interaction between chemicals x storage (Table-11) reveals that interaction had statistically significant effect on number of leaves. Highest (6.03) number of leaves was observed in GA<sub>3</sub>100 ppm under cold storage however, number of leaves obtained with the application of GA<sub>3</sub> 50 ppm under cold treatment was found at par with GA<sub>3</sub>100 ppm under cold treatments. The next higher (5.92) number of leaves were observed in sulphuric acid (7.5%) under 30 days cold treatments. Among the interaction of chemical x storage the lowest (3.01) number of leaves were recorded in ethrel 1000 ppm under ambient storage.

The interaction between storage, chemical and seed treatment was significant during the 2011. Highest number of leaves (6.66) was recorded in cold stored seeds under un-soaked seed treatment with GA<sub>3</sub>100 ppm. The next higher number of leaves was observed in GA<sub>3</sub>50 ppm with un-soaked seed treatment under cold storage. The lowest number of leaves (3.61) was recorded in ambient stored seeds with soaked seed treatment which was however, significantly higher when compared to control (3.02).

The data on influence of chemical treatments (Table-12) indicate statistically significant effect on number of leaves for the second year as well. Highest number of leaves (6.08) was recorded with the application of GA<sub>3</sub> 100 ppm followed by GA<sub>3</sub> 50 ppm (5.29). Sulphuric acid 7.5 per cent recorded number of leaves at par with GA<sub>3</sub> 50 ppm. Amongst chemical treatments lowest

**Table-11 : Influence of chemical and storage treatments on number of leaves of walnut seedling during the year 2011**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	6.43	5.68	6.05	4.96	4.11	4.53	5.29	5.69	4.89
	100 ppm	6.66	6.03	6.34	6.26	5.96	6.11	6.22	6.46	5.99
Etherel	1000 ppm	4.02	3.96	3.99	3.28	3.61	3.14	3.56	3.65	3.48
	1500 ppm	4.10	4.06	4.08	3.98	3.38	3.68	3.88	4.04	3.72
H <sub>2</sub> SO <sub>4</sub>	7.5 %	6.11	5.73	5.92	5.40	5.37	5.38	5.65	5.75	5.55
	9.5 %	5.90	5.13	5.51	5.21	5.03	5.12	5.31	5.55	5.08
KNO <sub>3</sub>	0.2 %	4.32	4.18	4.25	4.13	4.09	4.11	4.18	4.22	4.13
	0.4 %	4.61	4.50	4.55	4.33	4.20	4.26	4.40	4.47	4.35
Control	-	3.20	3.66	3.43	3.40	3.02	3.51	3.47	3.85	3.64
	Mean	5.11	4.80	4.96	4.59	4.34	4.46		4.85	4.57

**C.D(p≤0.05)**

Storage (S)	:	N.S
Chemical (C)	:	0.44
Seed treatment (ST)	:	N.S
C x S	:	0.80
ST x S	:	N.S
ST x C	:	0.79
S x C x ST	:	0.89

**Table-12 : Influence of chemical and storage treatments on number of leaves of walnut seedling during the year 2012**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	6.12	5.67	5.89	4.90	4.51	4.70	5.29	5.51	5.09
	100 ppm	6.53	6.39	6.46	5.32	6.11	5.71	6.08	5.92	6.25
Etherel	1000 ppm	4.15	4.08	4.11	4.00	3.90	3.95	4.03	4.07	3.99
	1500 ppm	5.28	4.80	5.04	4.73	3.99	4.36	4.70	5.00	4.39
H <sub>2</sub> SO <sub>4</sub>	7.5 %	5.60	5.41	5.50	5.20	4.80	5.00	5.25	5.40	5.10
	9.5 %	5.46	5.29	5.37	4.92	4.67	4.79	5.08	5.19	4.98
KNO <sub>3</sub>	0.2 %	4.30	4.80	4.55	4.60	4.51	4.55	4.55	4.45	4.65
	0.4 %	4.91	4.93	4.92	4.80	4.70	4.75	4.83	4.85	4.81
Control	-	3.97	3.88	3.92	3.31	3.32	3.31	3.61	3.64	3.60
Mean		5.14	5.02	5.08	4.64	4.50	4.57		4.89	4.76

**C.D(p≤0.05)**

Storage (S)	:	N.S
Chemical (C)	:	0.49
Seed treatment (ST)	:	N.S
C x S	:	0.85
ST x S	:	N.S
ST x C	:	0.12
S x C x ST	:	0.93

number of leaves (4.03) observed in ethrel 1000 ppm which was significantly higher when compared to control (3.61).

Storage treatments had a non significant effect on number of leaves during second year as well however, maximum number of leaves (5.08) was recorded under cold storage which was 4.57 leaves in case of ambient storage. Seed treatment also had a non significant effect on number of leaves however, un-soaked seed treatment recorded higher (4.89) number of leaves which was 4.76 in case of soaked treatment.

Interaction between chemical x storage had a statistically significant effect on number of leaves during second year as well. Highest number of leaves (6.46) was recorded in GA<sub>3</sub>100 ppm under cold storage treatment. The next higher number of leaves (5.89) was observed in GA<sub>3</sub> 50 ppm under cold storage. Among the chemical x storage interaction the lowest number of leaves(3.95) was recorded in ethrel 1000 ppm under ambient storage which was significantly higher in comparison to control (3.31).

Interaction between seed treatment x storage had a non significant effect on number of leaves. However, maximum number of leaves(5.14) was recorded under cold storage with un-soaked treatment whereas in case of ambient storage under soaked seed treatment number of leaves was 4.50.

Seed treatment x chemical interaction was statistically significant and higher number of leaves (6.25) was recorded in GA<sub>3</sub>100 ppm with soaked seed treatment. The next higher number of leaves (5.92) was recorded in GA<sub>3</sub>100 ppm with un-soaked seed treatment. Among the seed treatment x chemical interaction the lowest number of leaves (3.99) was recorded in ethrel 1000 ppm under soaked seed treatment which was higher than control (3.60).

The interactions between chemical x seed storage x seed treatment were also statistically significant. Higher number of leaves (6.53) was recorded in GA<sub>3</sub> 100 ppm under cold storage with un-soaked seed treatment. Among the

interactions the lowest number of leaves (3.90) was recorded in ethrel 1000 ppm under ambient storage with soaked seed treatment which was however, significantly higher when compared to control (3.32)

#### **4.1.7 Leaflet size (cm<sup>2</sup>)**

The data presented in Table-13 indicate that all the chemical and storage treatments had a significant effect on leaflet size. Among the chemical treatments GA<sub>3</sub> 100 ppm recorded maximum leaflet size (74.12 cm<sup>2</sup>) followed by GA<sub>3</sub> 50 ppm (72.27 cm<sup>2</sup>) and sulphuric acid 7.5 per cent (68.72 cm<sup>2</sup>). Amongst chemical treatments the lowest (56.98 cm<sup>2</sup>) leaflet size was observed with the application of ethrel 1500 ppm which was significantly higher when compared to control (45.33 cm<sup>2</sup>).

Among the storage treatments cold stored seeds resulted in significantly higher (65.62 cm<sup>2</sup>) leaflet size in comparison to ambient storage (61.30 cm<sup>2</sup>). Higher leaflet size (63.47 cm<sup>2</sup>) was also recorded under un-soaked seed treatments than the soaked seeds (62.57 cm<sup>2</sup>).

The perusal of data (Table-13) further indicates that the interactions between chemical x storage, seed treatment x storage and chemical x seed treatment were statistically significant. Maximum (75.86 cm<sup>2</sup>) leaflet size at interaction of chemical x storage was observed in GA<sub>3</sub> 100 ppm under 30 days chilling. The next higher (73.54 cm<sup>2</sup>) leaflet size was recorded with the application of GA<sub>3</sub> 50 ppm with 30 days cold treatment. The lowest (44.51 cm<sup>2</sup>) leaflet size was recorded in control under ambient storage.

Interaction of seed treatment x storage was found effective and maximum leaflet size (65.22 cm<sup>2</sup>) was recorded with the treatment combination of un-soaked seed under cold storage. However, leaflet size in both the seed treatments (un-soaked and soaked) were at par under cold and ambient storage. Among seed treatment x storage interaction the lowest leaflet size (60.87 cm<sup>2</sup>) was recorded in soaked seed treatment under ambient storage.

Under seed treatment x chemical interaction significantly higher leaflet size ( $75.02 \text{ cm}^2$ ) was observed with the application of GA<sub>3</sub> 100 ppm under un-soaked seed treatment. The next higher ( $73.24 \text{ cm}^2$ ) was observed with the application of GA<sub>3</sub>100 ppm under soaked seed treatment. Among seed treatment x chemical interaction the lowest leaflet size ( $43.34 \text{ cm}^2$ ) was recorded with control under un-soaked seed treatment.

The interactions between chemical, storage and seed treatment were also statistically significant (Table-13). Significantly higher leaflet size was recorded in GA<sub>3</sub> 100 ppm under cold storage treatment with un-soaked seed ( $77.36 \text{ cm}^2$ ) in comparison to all other treatment combinations. Among the interaction between chemical x seed treatment x storage the lowest leaflet size ( $42.58 \text{ cm}^2$ ) was recorded in control with ambient storage under un-soaked seed treatment.

The data on the influence of chemical and storage treatments on leaflet size of walnut seedlings for the second year (Table-14) reveal that among chemical treatments the maximum leaflet size was recorded in GA<sub>3</sub>100 ppm ( $72.70 \text{ cm}^2$ ) followed by GA<sub>3</sub> 50 ppm( $71.04 \text{ cm}^2$ ). Sulphuric acid was also found effective and recorded  $67.84 \text{ cm}^2$  leaflet size. Among chemical treatments the lowest leaflet size ( $55.15 \text{ cm}^2$ ) was recorded with the application of ethrel 1500 ppm which was however significantly higher when compared to control ( $53.13 \text{ cm}^2$ ).

Cold storage treatment was found non-effective in promoting leaflet size and maximum leaflet size ( $62.77 \text{ cm}^2$ ) was recorded in cold storage treatment which was  $61.18 \text{ cm}^2$  in case of ambient storage. Seed treatment had a significant effect on leaflet size and higher ( $62.98 \text{ cm}^2$ ) was recorded under un-soaked seed treatment which was  $60.97 \text{ cm}^2$  in case of soaked seed treatment.

The interaction of chemical x seed storage (Table-14) indicates that GA<sub>3</sub> 100 ppm under 30 day cold storage recorded highest ( $73.97 \text{ cm}^2$ ) leaflet size. The next higher leaflet size ( $71.56 \text{ cm}^2$ ) was observed in GA<sub>3</sub> 50 ppm under one month

**Table-13 : Influence of chemical and storage treatments on leaflet size (cm<sup>2</sup>) of walnut seedling during the year 2011**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	74.02	73.06	73.54	71.58	70.44	71.01	72.27	72.80	71.75
	100 ppm	77.36	74.37	75.86	72.67	72.10	72.38	74.12	75.02	73.24
Etherel	1000 ppm	62.91	62.24	62.57	59.76	54.51	57.13	59.85	58.74	55.23
	1500 ppm	60.28	58.61	59.44	57.19	51.85	54.52	56.98	61.34	58.38
H <sub>2</sub> SO <sub>4</sub>	7.5 %	70.59	69.71	70.15	67.96	66.63	67.29	68.72	69.28	68.17
	9.5 %	68.63	67.04	67.83	61.81	63.61	62.71	65.27	65.22	65.33
KNO <sub>3</sub>	0.2 %	65.25	62.51	63.88	60.30	60.04	60.17	62.02	62.08	61.28
	0.4 %	63.86	62.63	63.24	61.71	62.27	61.99	62.61	63.48	62.45
Control	-	44.10	48.21	46.15	42.58	46.44	44.51	45.33	43.34	47.33
Mean		65.22	64.26	65.62	61.72	60.87	61.30		63.47	62.57

**C.D(p≤0.05)**

Storage (S)	:	2.11
Chemical (C)	:	1.43
Seed treatment (ST)	:	1.01
C x S	:	2.15
ST x S	:	2.19
ST x C	:	0.13
S x C x ST	:	2.32

**Table-14 : Influence of chemical and storage treatments on leaflet size (cm<sup>2</sup>) of walnut seedling during the year 2012**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	72.47	70.66	71.56	71.25	69.81	70.53	71.04	71.86	70.24
	100 ppm	75.40	72.55	73.97	72.07	70.79	71.43	72.7	73.74	71.67
Etherel	1000 ppm	60.36	59.61	59.98	59.766	54.74	57.25	58.61	57.95	52.36
	1500 ppm	58.70	53.26	55.98	57.19	51.45	54.32	55.15	60.06	57.18
H <sub>2</sub> SO <sub>4</sub>	7.5 %	70.59	66.41	68.50	67.80	66.57	67.18	67.84	69.20	66.49
	9.5 %	67.55	64.92	66.23	61.81	58.59	60.20	63.21	64.68	61.76
KNO <sub>3</sub>	0.2 %	63.91	59.016	61.46	60.20	64.95	62.57	62.01	61.45	61.98
	0.4 %	62.70	58.31	60.50	61.60	60.63	61.11	60.80	62.76	59.47
Control	-	45.47	47.99	46.73	44.92	47.15	46.03	53.13	45.20	47.57
Mean		64.12	61.41	62.77	61.84	60.52	61.18		62.98	60.97

**C.D(p≤0.05)**

Storage (S)	:	N.S
Chemical (C)	:	1.49
Seed treatment (ST)	:	1.05
C x S	:	2.16
ST x S	:	2.14
ST x C	:	0.42
S x C x ST	:	2.28

cold treatment. Among the interaction of chemical x seed storage the lowest (46.03cm<sup>2</sup>) leaflet size was recorded in control at ambient storage.

Seed treatment x storage interaction was found effective and recorded highest leaflet size (64.12 cm<sup>2</sup>) with 30 days cold under un-soaked seed treatment. Soaked seed treatment at ambient storage was at par with un-soaked seed treatment under ambient storage as well as soaked seed treatment under cold storage. Among the interaction the lowest leaflet size (60.52 cm<sup>2</sup>) was observed under ambient storage with soaked seed treatment.

Seed treatment x chemical interaction had a significant effect on leaflet size and highest (73.74 cm<sup>2</sup>) leaflet size was recorded in GA<sub>3</sub> 100 ppm under un-soaked seed treatment. The next higher leaflet size (71.86 cm<sup>2</sup>) was recorded in GA<sub>3</sub> 50 ppm with un-soaked seed treatment. Among the interaction of seed treatment x chemicals the lowest leaflet size (52.36 cm<sup>2</sup>) was recorded with the application of ethrel 1000 ppm under soaked seed treatment which was significantly higher when compared to control(45.57 cm<sup>2</sup>).

The interactions between chemical, storage and seed treatment were also statistically significant. Highest leaflet size (75.40 cm<sup>2</sup>) was recorded in GA<sub>3</sub> 100 ppm under cold storage with un-soaked seed treatment. The next higher (72.55 cm<sup>2</sup>) leaflet size was obtained in GA<sub>3</sub> 100 ppm under cold storage with soaked seed treatment. Among the chemical x seed treatment x storage interactions the lowest leaflet size (44.92 cm<sup>2</sup>) was recorded in control under ambient storage with un-soaked seed.

#### **4.1.8 Leaf chlorophyll (mg/g of fresh weight)**

Chlorophyll content of walnut seedlings (Table-15) was found non-significant in all chemical and storage treatments. The chlorophyll content under different chemical treatments ranged from 0.80 to 0.86 mg/g of fresh weight, however maximum chlorophyll content(0.86 mg/g) was recorded in GA<sub>3</sub> 100 ppm during 2011.

**Table-15 : Influence of chemical and storage treatments on leaf chlorophyll (mg/g) of walnut seedling during the year 2011**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	0.85	0.85	0.85	0.85	0.84	0.84	0.84	0.85	0.85
	100 ppm	0.86	0.86	0.86	0.86	0.86	0.86	0.86	0.86	0.86
Etherel	1000 ppm	0.81	0.79	0.80	0.82	0.81	0.81	0.80	0.82	0.80
	1500 ppm	0.82	0.81	0.81	0.81	0.80	0.80	0.80	0.82	0.81
H <sub>2</sub> SO <sub>4</sub>	7.5 %	0.84	0.84	0.84	0.85	0.84	0.84	0.84	0.85	0.84
	9.5 %	0.84	0.80	0.82	0.84	0.84	0.84	0.83	0.84	0.82
KNO <sub>3</sub>	0.2 %	0.83	0.83	0.83	0.82	0.81	0.81	0.82	0.83	0.82
	0.4 %	0.85	0.86	0.85	0.84	0.85	0.84	0.84	0.85	0.86
Control	-	0.80	0.80	0.80	0.80	0.81	0.80	0.80	0.80	0.81
Mean		0.83	0.82	0.83	0.83	0.82	0.83	0.83	0.83	0.82

**C.D(p≤0.05)**

Storage (S)	:	NS
Chemical (C)	:	NS
Seed treatment (ST)	:	NS
C x S	:	NS
ST x S	:	NS
ST x C	:	NS
S x C x ST	:	NS

**Table-16 : Influence of chemical and storage treatments on leaf chlorophyll (mg/g) of walnut seedling during the year 2012**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	0.87	0.85	0.84	0.84	0.84	0.84	0.84	0.86	0.85
	100 ppm	0.86	0.80	0.83	0.86	0.86	0.86	0.86	0.86	0.83
Etherel	1000 ppm	0.80	0.82	0.88	0.88	0.88	0.88	0.82	0.84	0.85
	1500 ppm	0.81	0.81	0.85	0.83	0.83	0.83	0.83	0.82	0.82
H <sub>2</sub> SO <sub>4</sub>	7.5 %	0.83	0.84	0.83	0.82	0.82	0.82	0.82	0.83	0.83
	9.5 %	0.80	0.88	0.84	0.80	0.80	0.8	0.82	0.80	0.84
KNO <sub>3</sub>	0.2 %	0.77	0.87	0.82	0.77	0.77	0.77	0.79	0.77	0.82
	0.4 %	0.82	0.85	0.83	0.82	0.82	0.82	0.82	0.82	0.84
Control	-	0.81	0.81	0.81	0.81	0.81	0.81	0.81	0.81	0.81
Mean		0.87	0.82	0.83	0.82	0.82	0.82		0.85	0.82

**C.D(p≤0.05)**

Storage (S)	:	NS
Chemical (C)	:	NS
Seed treatment (ST)	:	NS
C x S	:	NS
ST x S	:	NS
ST x C	:	NS
S x C x ST	:	NS

The data on influence of chemical and storage treatments for the second year (Table-16) was also found non-significant.

#### **4.1.9 Number of days from germination to leaf fall**

The perusal of data on leaf fall of walnut seedlings for the year 2011 (Table-17) indicates that among the chemical treatments maximum number of days from germination to leaf fall (225.94) was recorded in GA<sub>3</sub> 100 ppm followed by GA<sub>3</sub> at 50 ppm (223.85) and sulphuric acid 7.5 per cent (217.14). Amongst the chemical treatments lowest number of days from germination to leaf fall (208.57) was observed with the application of ethrel at 1000 ppm which was significantly higher when compared to control (193.49)

Among the storage treatments cold storage resulted in significantly higher number of days from germination to leaf fall (212.97) in comparison to the ambient storage (210.45). Number of days from germination to leaf fall (213.52) recorded under un-soaked treatment was significantly higher than observed under soaked seed treatment (212.15).

The perusal of data in Table-17 further indicate that the interaction between chemical x storage, seed treatment x storage and chemical x seed treatments were statistically significant. Interaction of chemical x storage recorded maximum number of days (226.39) with GA<sub>3</sub> 100 ppm under 30 days cold followed by GA<sub>3</sub> 100 ppm (225.49) under ambient storage. Among the chemical x storage interaction the lowest number of days from germination to leaf fall was observed in control (193.41).

Interaction between seed treatment x storage had a significant effect on number of days from germination to leaf fall and maximum days (213.56) were observed in un-soaked seed treatment under cold storage followed by 212.27 days in case of soaked seed treatment under cold storage. However, un-soaked and soaked seed treatment under cold and ambient storage were at par. The minimum

number of days from germination to leaf fall (212.02) was recorded under ambient storage with soaked seed treatment.

Seed treatment x chemical interaction was found effective in promoting number of days from germination to leaf fall and maximum number of days from germination to leaf fall (226.83) was observed in GA<sub>3</sub> 100 ppm with un-soaked seed treatment followed by 225.06 days in GA<sub>3</sub> 100 ppm in soaked seed treatment. Among the interaction of seed treatment x chemical the lowest number of days from germination to leaf fall (192.66) was recorded in control under un-soaked seed treatment which was at par with untreated control under soaked seed treatment.

The interaction between chemical, storage and seed treatment was statistically significant and maximum number of days from germination to leaf fall (227.33) was observed in GA<sub>3</sub>100 ppm under cold storage with un-soaked seed treatment. Lowest number of days from germination to leaf fall (192.63) was observed with control at ambient storage under un-soaked seed treatment.

The data on influence of chemical and storage treatments for the second year (Table-18) reveal that among the chemical treatments GA<sub>3</sub> at 100 ppm recorded highest number of days from germination to leaf fall (224.46) followed by GA<sub>3</sub> 50 ppm (223.10). Sulphuric acid (7.5%) was also effective and recorded 216.14 number of days from germination to leaf fall. Amongst chemical treatments the minimum number of days from germination to leaf fall (208.23) was recorded with the application of ethrel 1000 ppm which was however significantly higher when compared to control (193.49). Cold storage treatment was effective and recorded maximum number of days from germination to leaf fall (212.14) than the ambient storage (211.56). Un-soaked seed treatment recorded higher number of days (212.39) from germination to leaf fall than soaked seeds (211.25).

**Table-17 : Influence of chemical and storage treatments on days from germination to leaf fall of walnut seedling during the year 2011**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	225.30	223.49	224.39	224.30	222.33	223.31	223.85	224.80	222.91
	100 ppm	227.33	225.45	226.39	226.33	224.66	225.49	225.94	226.83	225.06
Etherel	1000 ppm	208.53	209.66	208.99	209.61	208.33	208.16	208.57	209.97	208.10
	1500 ppm	210.45	209.30	209.87	213.38	211.43	212.40	211.13	211.92	210.37
H <sub>2</sub> SO <sub>4</sub>	7.5 %	219.60	215.66	217.63	217.66	215.66	216.66	217.14	218.63	215.66
	9.5 %	212.33	210.33	213.50	215.65	214.49	215.07	214.28	213.99	212.41
KNO <sub>3</sub>	0.2 %	210.38	209.61	209.99	210.66	206.33	208.49	209.24	210.52	207.97
	0.4 %	215.66	212.66	212.50	212.66	210.66	211.66	212.08	214.16	211.66
Control	-	192.66	194.33	193.43	192.63	193.13	193.41	193.49	192.66	194.33
	Mean	213.56	212.27	212.97	213.47	212.02	210.45		213.52	212.15

**C.D(p≤0.05)**

Storage (S)	:	0.32
Chemical (C)	:	1.45
Seed treatment (ST)	:	0.45
C x S	:	1.49
ST x S	:	0.53
ST x C	:	0.49
S x C x ST	:	1.54

**Table-18: Influence of chemical and storage treatments on days from germination to leaf fall of walnut seedling during the year 2012**

Treatment		Storage						Factor Means		
Chemical	Concentration	Cold			Ambient			Chemical	Seed treatment	
		Seed treatment		Mean	Seed treatment		Mean		Unsoaked	Soaked
		Unsoaked	Soaked		Unsoaked	Soaked				
GA <sub>3</sub>	50 ppm	226.30	223.49	224.89	222.30	220.33	221.31	223.10	224.30	221.91
	100 ppm	227.43	223.45	225.44	224.33	222.66	223.49	224.46	225.88	223.06
Etherel	1000 ppm	208.33	207.66	207.99	208.61	209.33	208.47	208.23	208.47	208.50
	1500 ppm	209.45	209.30	209.37	209.38	210.43	208.80	209.08	209.42	208.37
H <sub>2</sub> SO <sub>4</sub>	7.5 %	217.60	216.66	217.13	216.66	213.66	215.16	216.14	217.13	215.16
	9.5 %	212.33	210.33	211.33	213.65	213.49	213.57	212.45	212.99	211.91
KNO <sub>3</sub>	0.2 %	209.38	207.61	208.49	209.66	209.33	209.49	208.99	209.52	208.47
	0.4 %	211.66	210.66	211.16	210.66	209.66	210.16	210.66	211.16	210.16
Control	-	192.66	193.33	193.19	192.66	192.33	193.29	193.49	192.66	193.33
	Mean	212.79	211.39	212.14	211.99	211.10	211.56		212.39	211.25

**C.D(p≤0.05)**

Storage (S)	:	0.32
Chemical (C)	:	1.45
Seed treatment (ST)	:	0.45
C x S	:	1.49
ST x S	:	0.53
ST x C	:	0.53
S x C x ST	:	1.54

The perusal of data in Table-18 further reveals that interactions between chemical x storage, seed treatment x storage and chemical x seed treatment were statistically significant. Interaction of chemical x seed storage indicates that GA<sub>3</sub> at 100 ppm under one month chilling recorded maximum number of days from germination to leaf fall (225.44) followed by 222.26 days in GA<sub>3</sub> 100 ppm under ambient storage. Un-soaked seed with 30 day cold treatment recorded maximum number of days from germination to leaf fall (212.79) in comparison to the soaked treatment under ambient storage (211.10). Un-soaked seed treatment with GA<sub>3</sub> 100 ppm recorded significantly higher number of days from germination to leaf fall (225.88) than GA<sub>3</sub> at 100 ppm and seeds soaked in water (223.06).

The interaction between storage, chemical and seed treatment were statistically significant during the second year as well. Maximum number of days from germination to leaf fall (227.43) was recorded in cold stored seeds under un-soaked seed treatment with GA<sub>3</sub>100 ppm. Among the interactions the lowest number of days from germination to leaf fall (192.33) was recorded in control under ambient stored seeds with soaked seed treatment.

## **4.2 Epicotyl grafting in walnut**

### **4.2.1 Scion take percentage after 15 days**

The perusal of data in Table-19 reveals that the height of grafting, time of grafting and portion of scion wood used had a significant effect on scion take percentage recorded after 15 days of grafting during 2011. Maximum scion take percent (67.38) was recorded when grafting was performed at the point of attachment of cotyledons followed by grafting at 2cm above the point of attachment of cotyledons (65.55). At 4cm height scion take was to the tune of 58.88 per cent. The lowest scion take percent (54.16) was recorded in grafting performed at a height of 6 cm.

Time of grafting had a significant effect on scion take percentage (table-19). Highest scion take (67.49%) was recorded when grafting was performed on



**Plate-1 :** Epicotyl grafting in walnut (*Juglans regia* L.)

15 day old stocks followed by (63.37%) grafting performed on 30 day old stocks. Minimum scion take (45.36%) was observed when grafting was performed on 45 day old stocks. Higher percentage of scion take (62.99) was recorded in terminal portion of scion wood as compared to basal portion which was to the tune of 59.01 per cent.

The perusal of data in Table-19 reveals that interactions between height of grafting x scion wood, time x height of grafting and scion wood x time were statistically significant. Maximum scion take percentage (69.10) was observed under interaction of scion wood x height in grafting performed at the point of attachment of cotyledons using terminal portion of scion wood which was at par with scion take percent (68.44) obtained in grafting performed at the height of 2cm using terminal portion of scion wood. The least scion take percent (51.77) was recorded at 6cm height of grafting under basal portion of scion wood.

Height x time interaction produced maximum scion take percentage (71.39) when grafting performed at the height of 2cm on 15 day old stocks. The scion take percent obtained in epicotyl grafting in performed at the height of 2cm on 15 day old stocks however was at par with that obtained when grafting performed at the point of attachment of cotyledons on 15 day old stocks. The least scion take per cent (46.06%) was obtained when grafting was performed on 45 day old stocks.

Scion take per cent of 67.16 was obtained under interaction of scion wood x time (W x T) when grafting was performed on 15 day old stocks using terminal portion of scion wood which was at par with that recorded when basal portion of scion wood was used. The interaction between height of grafting, scion wood used and time of grafting was significant during 2011. Highest scion take percent (75.33) was observed at interaction (H x T x W) when grafting was performed on 15 day old stocks at the point of attachment of cotyledons using terminal portion of scion wood.

During second year also (Table-20) the height of grafting, scion wood used and time of grafting had a significant effect on scion take percentage. Maximum scion take percentage (70.38) was recorded in grafting performed just at the point of attachment of cotyledons. Grafting at 2cm height resulted in 67.64 percent scion take and at 4cm height scion take percentage was to the tune of 59.20. Amongst the grafting heights lowest scion take percentage (49.30) was recorded in grafting performed at a height of 6 cm.

The time of grafting was effective in promoting scion take percentage recorded after 15 days of grafting. Highest scion take percentage (64.79) was observed under grafting conducted on 15 day old stocks. However, scion take percent (63.76) obtained on 30 day old stocks was at par with scion take percent obtained on 15 day old stocks. The scion take percentage got significantly lowered when grafting was performed on 45 day old stocks and was to the tune of 55.73 per cent during the year 2012.

The portion of scion material also exerted a significant influence on the scion take percentage observed after 15 days of grafting. Significantly higher percentage of scion take (64.52) was recorded when terminal portion of scion wood was used compared to scion take percentage (59.42) when basal portion of scion wood was used.

The perusal of data in Table-20 reveals that interactions between height x scion wood, time x height and scion wood x time were statistically significant. Higher scion take percentage (72.77) was observed under interaction (W x H) when grafting was performed at point of attachment of cotyledons using terminal portion of scion wood in comparison to scion take percent (67.99) using basal portion of scion wood and grafted at point of attachment of cotyledons. At 2cm height of grafting scion take percentage was 69.21 using terminal portion of scion wood which was 66.07% in case of basal portion of scion wood. Among treatment combinations of scion wood x height the lowest scion take (47.87%) was recorded when grafting was performed at 6cm height using basal portion of scion wood.

**Table-19 : Scion take percentage after 15 days in walnut epicotyl grafting during the year 2011**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Height	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	75.33	70.33	61.66	69.10	66.66	61.66	56.66	61.66	67.38	70.99	67.40	59.16	
2	71.66	70.03	63.33	68.44	71.66	66.66	61.66	66.66	65.55	71.39	68.49	62.49	
4	68.33	66.66	56.66	63.88	66.66	53.33	41.66	53.88	58.88	69.49	59.99	49.16	
6	53.33	56.66	41.66	53.55	63.33	58.33	51.66	51.77	54.16	58.33	57.09	46.06	
Mean	67.16	65.99	55.82	62.99	67.07	59.99	52.91	59.01		67.49	63.37	45.36	

**C.D(p≤0.05)**

Height (H)	:	2.23
Time (T)	:	2.12
Wood (W)	:	2.35
W x H	:	2.39
H x T	:	2.02
W x T	:	2.57
H x T x W	:	3.11

**Table-20 : Scion take percentage after 15 days in walnut epicotyl grafting during the year 2012**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Height	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	78.33	76.66	63.33	72.77	73.33	61.33	69.33	67.99	70.38	75.83	72.19	66.33	
2	73.03	71.66	62.66	69.21	71.66	69.32	57.06	66.07	67.64	72.34	70.49	59.86	
4	68.31	66.33	53.03	62.56	63.66	55.23	48.66	55.85	59.20	65.98	60.78	50.84	
6	53.33	56.66	51.21	50.73	49.33	52.98	41.16	47.87	49.30	51.33	54.82	46.18	
Mean	68.25	67.82	57.42	64.52	64.49	59.71	54.05	59.42		64.79	63.76	55.73	

**C.D(p≤0.05)**

Height (H)	:	3.02
Time (T)	:	2.73
Wood (W)	:	2.97
W x H	:	2.34
H x T	:	1.02
W x T	:	2.41
H x T x W	:	3.13

Height x time interaction was statistically significant and recorded maximum scion take (75.83%) in grafting performed at the point of attachment of cotyledons. Among the height x time interactions the lowest scion take percent (46.18) was observed when grafting was performed on 45 days old stocks at height of 6 cm.

Scion wood and time (W x T) interaction was also found effective and revealed higher scion take percent (68.25) under grafting performed on 15 day old stocks using terminal portion wood. However, the scion take percent (67.82) obtained on 30 day old stocks was at par with that obtained on 15 day old stocks using terminal portion of scion wood. Minimum scion take percent (54.05) was obtained in grafting performed on 45 day old stock using basal portion of scion wood.

Treatment combinations of height x time of grafting x scion wood (H x T x W) recorded highest scion take percent (78.33) when grafting was performed at the point of attachment of cotyledons on 15 day old stocks using terminal portion of scion wood followed by 76.66% in grafts on 30 day old stocks at point of attachment of cotyledons using terminal portion of scion wood. Least scion take percent (41.16) was recorded in grafting performed at height of 6cm on 45 day old root stocks using basal portion of scion wood.

#### **4.2.2 Scion take percentage after 30 days**

The perusal of data in Table-21 indicate that the height of grafting, time of grafting and scion wood used had a significant effect on scion take percentage recorded after 30 days of grafting during 2011. Maximum scion take percent (63.32) was recorded when grafting was performed at the point of attachment of cotyledons followed by grafting at 2cm above the point of attachment of cotyledons (57.90). At 4cm height scion take was to the tune of 50.25 per cent. The lowest scion take percent (44.40) was recorded when grafting performed at a height of 6 cm.

Time of grafting had a significant effect on scion take percentage (Table-21). Highest scion take (59.38%) was recorded in grafting performed on 15 day old stocks followed by (54.63%) in grafting performed on 30 day old stocks. Minimum scion take (47.88%) observed in grafting performed on 45 day old stocks. Higher percentage of scion take (54.82) was recorded in terminal portion of scion wood as compared to basal portion which was to the tune of 53.11 per cent.

The perusal of data in Table-21 reveal that interactions between height of grafting x scion wood, time x height of grafting and scion wood x time were statistically significant. Maximum scion take percentage (65.27) was observed under interaction of scion wood x height in grafting performed at the point of attachment of cotyledons using terminal portion of scion wood followed by 61.21% when grafting was performed at the point of attachment of cotyledons using basal portion of scion wood. The least scion take percent (44.06) was recorded at 6cm height of grafting under basal portion of scion wood.

Height x time interaction produced maximum scion take percentage (68.46) in grafting performed at the point of attachment of cotyledons on 15 days old stocks followed by the scion take percent (63.82) obtained in epicotyl grafting performed at the height of 2cm on 15 day old stocks. The least scion take percent (39.66) was recorded after 30 days in grafting performed on 45 day old stocks.

Scion take per cent (59.78) obtained under interaction of wood x time (W x T) in grafting performed on 15 day old stocks using terminal portion of scion wood was at par with that recorded when basal portion of scion wood was used. The least scion take per cent (47.60) was recorded in grafting performed on 45 day old stocks.

The interactions between height of grafting, scion wood used and time of grafting were significant during 2011. Highest scion take percent (69.23) was observed at interaction (H x T x W) in grafting performed on 15 day old stocks at

the point of attachment of cotyledons using terminal portion of scion wood. The least scion take per cent (38.66) were recorded in grafting performed on 45 day old stocks.

During second year also (Table-22) the height of grafting, scion wood used and time of grafting had a significant effect on scion take percentage. Maximum scion take percentage (64.84) was recorded when grafting was performed just at the point of attachment of cotyledons. Grafting at 2cm height resulted in 58.60 scion take per cent. Amongst the grafting heights lowest scion take percentage (47.25) was recorded in grafting performed at a height of 6cm.

Highest scion take percentage (61.75) was observed under grafting conducted on 15 day old stocks followed by 55.99 per cent in grafting performed on 30 day old stocks. The scion take percentage got significantly lowered in grafting performed on 45 day old stocks and was to the tune of 50.51 per cent during the year 2012.

The portion of scion material also exerted a significant influence on the scion take percentage observed after 30 days of grafting. Significantly higher percentage of scion take (56.69) was recorded when terminal portion of scion wood was used compared to scion take percentage (54.61) when basal portion of scion wood was used.

The perusal of data in Table-22 reveals that interactions between height x wood, time x height and wood x time were statistically significant. Higher scion take percentage (66.66) was observed under interaction (W x H) under grafting performed at point of attachment of cotyledons using terminal portion of scion wood in comparison to scion take percent (63.02) using basal portion of scion wood and grafted at point of attachment of cotyledons. At 2cm height of grafting scion take percentage was 59.53 using terminal portion of scion wood which was 57.68% in case of basal portion of scion wood. Among treatment combinations of

**Table-21 : Scion take percentage after 30 days in walnut epicotyl grafting during the year 2011**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Height	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	69.23	65.33	61.26	65.27	67.70	60.21	56.20	61.37	63.32	68.46	62.77	58.73	
2	65.32	65.00	45.10	58.47	62.33	59.30	50.36	57.33	57.90	63.82	62.15	47.73	
4	56.11	50.66	45.66	50.81	55.60	48.28	45.20	49.69	50.25	55.85	49.47	45.43	
6	48.46	45.12	40.66	44.74	50.33	43.20	38.66	44.06	44.40	49.39	44.16	39.66	
Mean	59.78	56.52	48.17	54.82	58.99	52.74	47.60	53.11		59.38	54.63	47.88	

**C.D(p≤0.05)**

Height (H)	:	1.50
Time (T)	:	1.82
Wood (W)	:	N.S
W x H	:	2.34
H x T	:	2.48
W x T	:	2.30
H x T x W	:	2.68

**Table-22 : Scion take percentage after 30 days in walnut epicotyl grafting during the year 2012**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Height	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	71.91	68.21	59.88	66.66	69.13	63.60	56.33	63.02	64.84	70.52	65.90	58.10	
2	66.50	63.11	49.00	59.53	60.60	61.33	51.11	57.68	58.60	63.55	62.22	50.05	
4	59.10	52.20	45.20	52.16	58.29	48.48	48.29	51.68	51.92	58.69	50.34	51.92	
6	56.20	45.70	43.33	48.41	52.33	45.30	40.66	46.09	47.25	54.26	45.50	41.99	
Mean	63.42	57.30	49.35	56.69	60.08	54.67	49.09	54.61		61.75	55.99	50.51	

**C.D(p≤0.05)**

Height (H)	:	1.54
Time (T)	:	1.87
Wood (W)	:	1.73
W x H	:	2.36
H x T	:	2.51
W x T	:	2.32
H x T x W	:	2.72

scion wood x height the lowest scion take (46.09%) was recorded under grafting performed at 6cm height using basal portion of scion wood.

Height x time interaction was statistically significant and recorded maximum scion take (70.52%) in grafting performed at the point of attachment of cotyledons. Among the height x time interactions the lowest scion take percent (41.99) was observed when grafting was performed on 45 days old stocks at height of 6 cm.

Wood and time (W x T) interaction was also found effective and revealed higher scion take percent (63.42) in grafting performed on 15 day old stocks using terminal portion wood followed by 57.30 per cent on 30 day old stocks. Minimum scion take percent (49.09) was obtained in grafting performed on 45 day old stocks using basal portion of scion wood.

Treatment combinations of height x time of grafting and wood (H x T x W) recorded highest scion take percent (71.91) in grafting performed at the point of attachment of cotyledons on 15 day old stocks using terminal portion of scion wood followed by 68.21 per cent under grafting on 30 day old stocks at point of attachment of cotyledons using terminal portion of scion wood. Least scion take percent (40.66) was recorded in grafting performed at height of 6cm on 45 day old root stocks using basal portion of scion wood.

#### **4.2.3 Scion take percentage after 45 days**

The perusal of data in Table-23 reveals that the height of grafting, time of grafting and scion wood used had a significant effect on scion take percentage recorded after 45days of grafting during 2011. Maximum scion take percent (43.16) was recorded under grafting performed at the point of attachment of cotyledons followed by grafting at 2cm above the point of attachment of cotyledons (39.32). At 4cm height scion take was to the tune of 33.82 per cent. The lowest scion take percent (28.16) was recorded in grafting performed at a height of 6cm.

Time of grafting had a significant effect on scion take percentage (Table-23). Highest scion take (42.53%) was recorded when grafting was performed on 15 day old stocks followed by (38.78%) grafting performed on 30 day old stocks. Minimum scion take (27.03%) was observed in grafting performed on 45 day old stocks. Higher percentage of scion take (45.77) was recorded in terminal portion of scion wood as compared to basal portion (34.43%).

The perusal of data in Table-23 reveals that interactions between height of grafting x scion wood, time x height of grafting and scion wood x time were statistically significant. Maximum scion take percentage (45.77) was observed under interaction of scion wood x height in grafting performed at the point of attachment of cotyledons using terminal portion of scion wood followed by 40.55% in grafting performed at the point of attachment of cotyledons using basal portion of scion wood. The least scion take percent (26.55) was recorded at 6cm height of grafting under basal portion of scion wood.

Height x time interaction produced maximum scion take percentage (49.99) under grafting performed at the point of attachment of cotyledons on 15 days old stocks. The next higher scion take percent (45.49) was obtained in epicotyl grafting performed at the point of attachment of cotyledons on 15 day old stocks. The least scion take percent (19.16) was recorded in grafting performed on 45 day old stocks.

The scion take per cent (43.91) obtained under interaction of wood x time (W x T) in grafting performed on 15 day old stocks using terminal portion of scion wood was at par with that recorded when basal portion of scion wood was used. The interaction between height of grafting x scion wood used and time of grafting was significant during 2011. Highest scion take percent (53.33) was observed at interaction (H x T x W) in grafting performed on 15 day old stocks at the point of attachment of cotyledons using terminal portion of scion wood.

During second year also (Table-24) the height of grafting, scion wood used and time of grafting had a significant effect on scion take percentage. Maximum scion take percentage (45.32) was recorded in grafting performed just at the point of attachment of cotyledons. Grafting at 2cm height resulted in 39.54 scion take percent. Amongst the grafting heights lowest scion take percentage (28.32) was recorded in grafting performed at a height of 6cm.

Highest scion take percentage (43.78) was observed under grafting conducted on 15 day old stocks followed by 39.33 per cent in grafting performed on 30 day old stocks. The scion take percentage got significantly lowered when grafting was performed on 45 day old stocks and was to the tune of 27.45 per cent during the year 2012. The portion of scion material also exerted a significant influence on the scion take percentage observed after 45 days of grafting. Significantly higher percentage of scion take (38.71) was recorded when terminal portion of scion wood was used compared to scion take percentage (34.99) when basal portion of scion wood was used.

The perusal of data in Table-24 reveals that interaction between height x wood, time x height and wood x time were statistically significant. Higher scion take percentage (48.10) was observed under interaction (W x H) in grafting performed at point of attachment of cotyledons using terminal portion of scion wood in comparison to scion take percent (42.55) using basal portion of scion wood and grafted at point of attachment of cotyledons. At 2cm height of grafting scion take percentage was 40.88 using terminal portion of scion wood which was 38.2 per cent in case of basal portion of scion wood. Amongst the treatment combinations of scion wood x height the lowest scion take (26.76%) was recorded in grafting performed at 6cm height using basal portion of scion wood.

Height x time interaction was statistically significant and recorded maximum scion take (54.49%) in grafting performed at the point of attachment of cotyledons. Among the height x time interactions the lowest scion take per cent

**Table-23 : Scion take percentage after 45 days in walnut epicotyl grafting during the year 2011**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Height	Time		
	15 DAG	30 DAG	45 DAG	15 DAG		30 DAG	45 DAG	15 DAG	30 DAG		45 DAG		
	15 DAG	30 DAG	45 DAG	Mean	15 DAG	30 DAG	45 DAG	Mean	Height	15 DAG	30 DAG	45 DAG	
0	53.33	47.66	36.33	45.77	46.66	43.33	31.66	40.55	43.16	49.99	45.49	33.99	
2	45.33	43.66	32.33	40.44	44.66	42.33	27.66	38.21	39.32	44.99	42.99	29.99	
4	40.66	38.33	26.66	35.21	38.66	35.33	23.33	32.44	33.82	39.66	36.83	24.99	
6	36.33	31.33	21.66	29.77	34.66	28.33	16.66	26.55	28.16	35.49	29.83	19.16	
Mean	43.91	40.24	29.24	45.77	41.16	37.33	24.82	34.43		42.53	38.78	27.03	

**C.D(p≤0.05)**

Height (H)	:	2.11
Time (T)	:	2.07
Wood (W)	:	3.11
W x H	:	3.19
H x T	:	2.43
W x T	:	3.23
H x T x W	:	3.39

**Table-24 : Scion take percentage after 45 days in walnut epicotyl grafting during the year 2012**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Height	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	59.66	48.33	36.33	48.10	49.33	45.00	33.33	42.55	45.32	54.49	46.66	34.83	
2	46.33	43.33	33.00	40.88	44.66	42.66	27.33	38.21	39.54	45.49	42.99	30.16	
4	41.66	40.00	26.33	35.99	38.66	35.00	23.66	32.44	34.21	40.16	37.50	24.99	
6	36.66	31.66	21.33	29.88	33.30	28.66	18.33	26.76	28.32	34.98	30.16	19.83	
Mean	46.07	40.83	29.24	38.71	41.48	37.83	25.66	34.99		43.78	39.33	27.45	

**C.D(p≤0.05)**

Height (H)	:	2.13
Time (T)	:	2.09
Wood (W)	:	3.07
W x H	:	3.12
H x T	:	3.11
W x T	:	3.19
H x T x W	:	3.28

(19.83) was observed when grafting was performed on 45 day old stocks at height of 6cm.

Wood and time (W x T) interaction was also found effective and revealed higher scion take percent (46.06) in grafting performed on 15 day old stocks using terminal portion wood. The next higher scion take percent (40.83) was recorded with 30 day old stocks after 45 days of grafting. Minimum scion take percent (25.66) was obtained in grafting performed on 45 day old stocks using terminal portion of scion wood.

Treatment combinations of height x time of grafting x wood (H x T x W) recorded highest scion take percent (59.66) in grafting performed at the point of attachment of cotyledons on 15 day old stocks using terminal portion of scion wood followed by 49.33 per cent when grafted on 15 day old stocks at point of attachment of cotyledons using basal portion of scion wood. Least scion take percent (18.33) was recorded in grafting performed at height of 6cm on 45 day old root stocks using basal portion of scion wood.

#### **4.2.4 Scion sprouting percentage**

The data in Table-25 indicates that the height of grafting x time of grafting x scion wood used had a significant effect on scion sprouting percentage during the year 2011. Maximum scion sprouting percentage (40.93) was recorded in grafting performed at the point of attachment of cotyledons followed by grafting at 2cm above the point of attachment of cotyledons (35.88%). At 4cm height scion sprouting percentage was to the tune of 27.99. The lowest scion sprouting percentage (20.99) was recorded in grafting performed at a height of 6cm.

The time of grafting was effective in promoting scion sprouting percentage. Highest scion sprouting percentage (36.12) was observed under grafting conducted on 15 day old stocks followed by grafting on 30 days old stock (33.74%). The scion sprouting percentage got significantly lowered when grafting was performed on 45 day old stocks and was to the tune of 22.24 per cent during



**Epicotyl grafting at point of attachment of cotyledons**



**Epicotyl grafting at 2 cm**



**Epicotyl grafting at 4 cm**

**Plate-2 : Sprouted grafts**

the year 2011. Significantly higher scion sprouting per cent (32.18) was recorded in terminal portion of scion wood as compared to basal portion which was to the tune of 29.21 per cent.

The perusal of data in Table-25 further indicates that the interactions between height x scion wood, time x height and scion wood x time were statistically significant. Maximum scion sprouting percentage (43.88) was recorded under interaction scion wood x height in grafting performed at the point of attachment of cotyledons using terminal portion of scion wood followed by sprouting percentage (37.99) using basal portion of scion wood and grafted at the point of attachment of cotyledons. At 2cm height of grafting scion sprouting was 37.44 per cent using terminal portion of scion wood whereas it was 35.88 per cent in case of basal portion of scion wood grafted at height of 2cm. Among the treatment combination of wood x height the lowest scion sprouting (17.44%) was recorded in grafting performed at a height of 6cm using basal portion of scion wood.

Height x time interaction was statistically significant. Maximum scion sprouting (47.49%) was obtained on 15 day old stocks grafted at the point of attachment of cotyledons followed by scion sprouting per cent (43.49) in grafting performed on 30 day old stocks grafted just at the point of attachment of cotyledons. Among the height x time interaction the lowest scion sprouting per cent (13.66) was observed in grafting performed on 45 day old stocks at the height of 6cm.

Scion wood x time interaction was found effective which records significantly higher scion sprouting per cent (37.91) in grafting performed on 15 day old stock using terminal portion of scion wood. The next higher scion sprouting per cent (35.16) was obtained when grafting was performed on 30 day old stocks. Minimum scion sprouting per cent (20.99) was observed under interaction of scion wood x time in grafting performed on 45 day old stocks using basal portion of scion wood.

The interaction between height x time x scion wood (H x T x W) indicate that significantly higher scion sprouting (51.66 %) was obtained in grafting performed at the point of attachment of cotyledons on 15 day old stocks using terminal portion of scion wood followed by scion sprouting per cent of 46.66 when grafting was performed on 30 day old stocks at the point of attachment of cotyledons using terminal portion of scion wood. The minimum scion sprouting percentage (13.67) was recorded in grafting performed at a height of 6cm using 45 day old stocks and basal portion of scion wood.

During second year also (Table-26) height of grafting x scion wood used and time of scion grafting had a significant effect on scion sprouting percentage. Maximum scion sprouting percentage (41.32) was recorded in epicotyl grafting performed just at the point of attachment of cotyledons. Grafting at 2cm height resulted in 33.99 per cent scion sprouting and at 4cm height scion sprouting per cent was to the tune of 29.43. Amongst grafting heights the lowest scion sprouting per cent (18.21) was recorded in grafting performed at a height of 6cm.

Time of grafting had a significant effect on scion sprouting percentage (Table-26). Highest scion sprouting (37.99%) was recorded in grafting performed on 15 day old stocks followed by scion sprouting per cent (34.41) in grafting performed on 30 day old stocks. The minimum scion sprouting (19.82%) was observed in grafting performed on 45 day old stocks. Significantly higher percentage of scion sprouting (31.85) was recorded in terminal portion of scion wood as compared to basal portion in which it was to the tune of 29.63 per cent.

The perusal of data in Table- 26 further indicates that interactions between height x scion wood, time x height of grafting and scion wood x time of grafting were statistically significant. Maximum scion sprouting (43.77%) was recorded in the interaction of scion wood x height (W x H) in grafting performed at the point of attachment of cotyledons using terminal portion of scion wood which was 38.88 per cent in case of basal portion of scion wood and grafting at the point of attachment of cotyledons. Among treatment combination of scion wood x height

**Table-25 : Scion sprouting percentage in walnut epicotyl grafting during the year 2011**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal			Mean	Height	Time		
	15 DAG	30 DAG	45 DAG			15 DAG	30 DAG	45 DAG			15 DAG	30 DAG	45 DAG
0	51.66	46.66	33.33	43.88	43.33	40.33	30.33	37.99	40.93	47.49	43.49	31.83	
2	41.66	40.33	30.33	37.44	40.33	39.33	23.33	34.33	35.88	40.99	39.83	26.83	
4	36.66	33.33	16.66	28.88	33.33	31.33	16.66	27.10	27.99	34.99	32.33	16.66	
6	21.66	20.33	13.66	18.55	20.33	18.33	13.67	17.44	17.99	20.99	19.33	13.66	
Mean	37.91	35.16	23.49	32.18	34.33	32.33	20.99	29.21		36.12	33.74	22.24	

**C.D(p≤0.05)**

Height (H)	:	1.05
Time (T)	:	1.11
Wood (W)	:	1.23
W x H	:	1.27
H x T	:	3.09
W x T	:	1.32
H x T x W	:	2.54

**Table-26 : Scion sprouting percentage in walnut epicotyl grafting during the year 2012**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal			Mean	Height	Time		
	15 DAG	30 DAG	45 DAG			15 DAG	30 DAG	45 DAG			15 DAG	30 DAG	45 DAG
0	53.33	46.66	31.33	43.77	46.66	43.33	26.66	38.88	41.32	49.99	44.99	28.99	
2	43.33	40.33	20.33	34.66	40.33	39.33	20.33	33.33	33.99	41.83	39.83	20.33	
4	41.66	33.33	16.66	30.55	36.66	31.66	16.66	28.32	29.43	39.16	32.49	16.66	
6	21.66	20.33	13.33	18.44	20.33	20.33	13.23	17.99	18.21	20.99	20.33	13.43	
Mean	39.99	35.16	20.41	31.85	35.99	33.66	19.24	29.63		37.99	34.41	19.82	

**C.D(p≤0.05)**

Height (H)	:	1.08
Time (T)	:	1.12
Wood (W)	:	1.25
W x H	:	1.33
H x T	:	2.44
W x T	:	1.35
H x T x W	:	2.63

the lowest scion sprouting (17.99%) was recorded in grafting performed at a height of 6 cm using basal portion of scion wood.

Height x time interaction had a significant effect on scion sprouting percentage. Maximum scion sprouting (49.99%) was observed on 15 day old stocks grafted at the point of attachment of cotyledons followed by 30 day old stocks grafted at the height of point of attachment of cotyledons (44.99%). Amongst height x time interactions the lowest scion sprouting per cent (13.43) was observed under grafting performed on 45 day old stocks at a height of 6cm.

Scion wood x time interaction was found effective which revealed significantly higher scion sprouting per cent (39.99) in grafting conducted on 15 day old stocks using terminal portion of scion wood. The next higher scion sprouting per cent (35.16) was recorded in grafting performed on 30 day old stocks using terminal portion of scion wood. Among height x scion wood interactions minimum scion sprouting (19.24) was recorded in epicotyl grafting performed on 45 day old stocks using basal portion of scion wood.

The interaction between height of grafting x scion wood used x time of grafting (H x T x W) were statistically significant during 2012 as well. Highest scion sprouting per cent (53.33) was observed in grafting performed on 15 day old stocks at the point of attachment of cotyledons using terminal portion of scion wood. The lowest scion sprouting (13.33%) was recorded in epicotyl grafting performed at 6 cm height on 45 day old stocks using basal portion of scion wood.

#### **4.2.5 Scion growth (cm)**

The data (Table -27) indicates that height of grafting, scion wood used and time of grafting had a significant influence on scion growth in walnut epicotyl grafting during the year 2011. Maximum scion growth (15.75 cm) was recorded in grafting performed just at the point of attachment of cotyledons. Scion growth obtained at 2cm and 4cm graftings were at par with each other. Among grafting

heights the minimum scion growth (15.15 cm) was obtained in grafting performed at the height of 6cm.

Time of grafting had a significant effect on scion growth. Maximum scion growth (16.22 cm) was recorded when grafting was performed on 15 day old stocks followed by (15.74 cm) grafting performed on 30 day old stocks. The lowest scion growth (14.99 cm) was recorded in grafting performed on 45 day old stocks. The portion of scion wood used also affected scion growth and significantly higher scion growth (15.87 cm) was recorded in terminal portion than the scion growth 15.42 cm in basal portion of scion wood.

The perusal of data in Table-27 further indicates that the interactions between height of grafting, time of grafting and scion wood used had a significant effect on scion growth during 2011. Maximum scion growth (16.84 cm) was recorded under interaction of wood x height in grafting performed at the point of attachment of cotyledons using terminal portion of scion wood followed by 15.69 cm scion growth in grafts performed at the height of 2cm using basal portion of scion wood. Among treatment combinations of scion wood x height minimum scion growth (14.99 cm) was recorded in grafting performed at the height of 6cm using basal portion of scion wood.

Height x time interactions were statistically significant and higher scion growth (17.37 cm) was observed in grafts performed at the point of attachment of cotyledons on 15 day old stocks. The next higher scion growth (16.27 cm) was recorded in grafts performed at the point of attachment of cotyledons on 30 day old stocks. Among height x time interaction the lowest scion growth (14.37 cm) was obtained when grafting was performed at the height of 6cm on 45 day old stocks.

Scion wood x time interaction was also effective which revealed significantly higher scion growth (16.31 cm) in grafts performed on 15 day old stocks using terminal portion of scion wood. Scion growth (16.23 cm) recorded on

30 day old stocks using terminal portion was at par with 15 day old stocks with terminal portion of scion wood. Minimum scion growth (14.88 cm) was observed in grafting performed on 45 day old stocks using basal portion of scion wood.

The interaction between height of grafting x time of grafting x scion wood used reveals that highest scion growth (18.59 cm) was obtained when grafts performed at the point of attachment of cotyledons on 15 day old stocks using terminal portion of scion wood followed by scion growth (16.81 cm) in grafting performed at the point of attachment of cotyledons on 30 day old stocks using terminal portion of scion wood. Lowest scion growth (14.12 cm) was recorded in grafting performed at the height of 6cm on 45 day old stocks using basal portion of scion wood.

During second year (Table-28) also the height of grafting, time of grafting and portion of scion wood used had a significant effect on scion growth in walnut epicotyl grafting. The maximum scion growth (15.78 cm) was recorded in grafting performed on 15 day old stocks at a point of attachment of cotyledons. The minimum scion growth (15.23 cm) was observed under grafting performed at the height of 6cm.

Time of grafting reveals that highest scion growth (16.20 cm) was recorded in grafting was performed on 15 day old stock. The next higher scion growth (15.63 cm) was observed in grafting performed on 30 day old stocks. Lowest scion growth (14.82 cm) was recorded under grafting performed on 45 day old stocks. The portion of scion wood also exerted influence on scion growth and comparatively higher scion growth 15.71 cm was recorded in terminal portion of scion wood than basal portion of scion wood (15.41 cm).

The perusal of data (Table-28) further indicates that interactions between height of grafting, time of grafting and scion wood used were significant. Maximum scion growth (15.99 cm) was recorded under grafting performed at the point of attachment of cotyledons using terminal portion of scion wood. Scion

growth recorded at 2cm and 4cm grafting heights were at par with each other. Similarly scion growth recorded on grafting conducted at point of attachment of cotyledons and 2 cm heights were at par using basal portion of scion wood. Minimum scion growth (15.23 cm) was recorded in grafting performed at 6cm height using basal portion of scion wood.

The interactions between height x time of grafting were also statistically significant. Higher scion growth (16.62 cm) was recorded in grafting performed at the point of attachment of cotyledons on 15 day old stocks. The next higher growth (16.38 cm) was recorded under grafting performed at 2cm height on 15 day old stocks. Among the time x height interaction the lowest scion growth (14.33 cm) was observed in grafting performed on 45 day old stock at a height of 6cm.

Scion wood x time interactions was found effective which revealed significantly higher scion growth (16.35 cm) in grafting performed on 15 day old stocks using terminal portion of scion wood followed by grafting performed on 15 day old stocks using basal portion of scion wood which was to the tune of (16.06cm). Among scion wood x time interaction the lowest scion growth (14.65 cm) was observed when grafting was performed on 45 day old stocks using basal portion of scion wood.

The interactions between height of grafting x scion wood used x time of grafting were statistically significant during 2012. Highest scion growth (17.06 cm) was observed at the interaction of height x time x scion wood under grafting performed on 15 day old stock at the point of attachment of cotyledons using terminal portion of scion wood. Among interactions of height x time x scion wood the lowest scion growth (14.08 cm) was recorded in grafting performed at 6cm height on 45 day old stocks using basal portion of scion wood.

**Table-27: Scion growth (cm) in walnut epicotyl grafting during the year 2011**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal			Mean	Height	Time		
	15 DAG	30 DAG	45 DAG			15 DAG	30 DAG	45 DAG			15 DAG	30 DAG	45 DAG
0	18.59	16.81	15.12	16.84	16.15	15.73	15.15	15.67	15.75	17.37	16.27	15.13	
2	15.25	16.17	15.25	15.55	16.44	15.60	15.04	15.69	15.62	15.84	15.88	15.14	
4	15.99	16.04	15.40	15.81	16.17	14.63	15.24	15.34	15.57	16.08	15.33	15.32	
6	15.44	15.9	14.62	15.32	15.78	15.09	14.12	14.99	15.15	15.61	15.49	14.37	
Mean	16.31	16.23	15.09	15.87	16.13	15.26	14.88	15.42		16.22	15.74	14.99	

**C.D(p≤0.05)**

Height (H)	:	0.09
Time (T)	:	0.15
Wood (W)	:	0.20
W x H	:	0.23
H x T	:	1.12
W x T	:	0.27
H x T x W	:	0.33

**Table-28 : Scion growth (cm) in walnut epicotyl grafting during the year 2012**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Height	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	17.06	15.70	15.21	15.99	16.18	15.85	14.71	15.58	15.78	16.62	15.77	14.96	
2	16.37	15.87	15.15	15.79	16.40	15.72	14.56	15.56	15.67	16.38	15.79	14.85	
4	16.11	15.98	15.08	15.72	16.24	14.67	15.25	15.38	15.55	16.17	15.32	15.16	
6	15.87	15.56	14.59	15.34	15.44	15.88	14.08	15.13	15.23	15.65	15.72	14.33	
Mean	16.35	15.77	15.00	15.71	16.06	15.53	14.65	15.41		16.20	15.63	14.82	

**C.D(p≤0.05)**

Height (H)	:	0.07
Time (T)	:	0.13
Wood (W)	:	0.16
W x H	:	0.19
H x T	:	1.07
W x T	:	0.25
H x T x W	:	0.30

#### 4.2.6 Number of leaves

The data in Table-29 indicate that the height of grafting, time of grafting and scion wood used had a significant effect on number of leaves in walnut epicotyl grafting during 2011. Maximum number of leaves (4.88) was recorded in grafting performed at the point of attachment of cotyledons followed by grafting at 2cm above the point of attachment of cotyledons (4.66). At 4cm height number of leaves was to the tune of 4.65. Lowest number of leaves (4.10) was recorded in grafting performed at a height of 6cm.

The time of grafting had a significant effect on the number of leaves and highest number of leaves (5.47) was recorded under grafting performed on 15 day old stocks followed by grafting on 30 day old stocks (4.45). The number of leaves got significantly lowered when grafting was performed on 45 day old stocks and was to the tune of 3.78 during the year 2011. Higher number of leaves (4.69) was recorded under terminal portion of scion wood as compared to basal portion which was to the tune of 4.46 number of leaves.

The perusal of data in Table-29 indicates that interactions between height x scion wood, time x height and scion wood x time were statistically significant. Maximum number of leaves (5.33) were recorded under interaction of wood x height in grafting performed at the point of attachment of cotyledons using terminal portion of scion wood whereas it was 4.44 number of leaves in case of grafting performed at point of attachment of cotyledons using basal portion of scion wood. The minimum number of leaves (3.88) was recorded at interaction of scion wood x height under grafting performed at 6cm height using basal portion of scion wood.

Height x time interaction produced maximum number of leaves (5.99) in grafting performed at the point of attachment of cotyledons on 15 day old stocks. The number of leaves recorded at 2cm and 4cm on 15 day old stocks were at par

in grafts. Lowest number of leaves (3.31) at the interaction of height x time were observed in grafting performed at 6cm height on 45 day old stocks.

Wood x time interactions were found effective and higher number of leaves (5.75) were recorded in grafting performed on 15 day old stocks using terminal portion of scion wood. The next higher number of leaves (5.24) were observed when grafting was performed on 15 day old stocks using basal portion of scion wood. Among the interaction wood x height the lowest number of leaves (3.66) were recorded in grafting performed on 45 day old stocks using basal portion of scion wood.

The interactions between height of grafting x scion wood used x time of grafting were significant during 2011. Highest number of leaves (6.23) were observed at interaction of height x time x scion wood in grafting performed on 15 day old stocks at the point of attachment of cotyledons using terminal portion of scion wood. The next higher number of leaves (6.00) were recorded under interactions of height x time x scion wood under grafting performed at 4cm height on 15 day old stocks using terminal portion of scion wood. The lowest number of leaves (3.23) were recorded at interaction of height x time x wood in grafting performed at 6cm height on 45 day old stocks using basal portion of scion wood.

During second year also the height of grafting, time of grafting and scion wood used had a significant effect on number of leaves (Table-30). Maximum number of leaves (5.27) was recorded in grafting performed at the point of attachment of cotyledons. Grafting at 2cm height resulted 4.70 number of leaves and at 4cm height number of leaves was to the tune of 4.60. Among the grafting heights the lowest number of leaves (4.20) was recorded in grafting performed at a height of 6 cm.

The time of grafting had a significant effect on number of leaves (Table-30). Highest number of leaves (5.20) was recorded in grafting performed on 15 day old stocks followed by 4.37 number of leaves on grafts grafted on 30 day old

stocks. Minimum number of leaves (4.03) was recorded in grafting performed on 45 day old stocks. The number of leaves (4.69) recorded under terminal portion of scion wood used was at par with the number of leaves (4.63) recorded in basal portion of scion wood.

The perusal of data in Table-30 indicates that interactions between height x scion wood, time x height and scion wood x time of grafting were significant. Maximum number of leaves (5.33) was observed under interaction of wood x height in grafting performed at the point of attachment of cotyledons using terminal portion of scion wood. The next higher number of leaves (5.21) was observed under grafting performed on 15 day old stocks using basal portion of scion wood. Among the interaction of height x wood the lowest number of leaves (3.88) were observed under grafting performed on 45 day old stocks using terminal portion of scion wood.

The interaction height x time produced maximum number of leaves (5.89) in grafting performed at the point of attachment of cotyledons on 15 day old stocks. Grafting at 2 and 4cm heights on 15 day old stocks were at par with each other. Among the height x time interaction the lowest number of leaves (3.63) were recorded in grafting performed at the 6cm height on 45 day old stocks.

Scion wood x time interaction had also significant influence on number of leaves and higher number of leaves (5.66) were observed on 15 day old stocks using terminal portion of scion wood. The next higher number of leaves (5.24) were recorded under grafting performed on 15 day old stocks using basal portion of scion wood. Minimum number of leaves (3.80) were recorded in grafting performed on 45 day old stocks using basal portion of scion wood.

The interactions between height x time x scion wood reveal that the highest number of leaves (6.33) were obtained under grafting performed at the point of attachment of cotyledons on 15 day old stocks using terminal portion of scion wood. Number of leaves were at par in grafting performed at 4cm height on

**Table-29 : Number of leaves in walnut epicotyl grafting during the year 2011**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Height	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	6.23	5.33	4.13	5.33	5.66	4.33	3.93	4.44	4.88	5.99	4.83	3.83	
2	5.66	4.33	3.66	4.55	5.33	4.66	4.63	4.77	4.66	5.49	4.49	3.92	
4	6.00	4.23	4.13	4.91	4.60	4.60	3.66	4.32	4.65	5.40	4.46	3.90	
6	4.66	3.66	3.33	4.33	5.33	4.13	3.23	3.88	4.10	4.99	3.99	3.31	
Mean	5.75	4.41	3.91	4.69	5.24	4.49	3.66	4.46		5.47	4.45	3.78	

**C.D(p≤0.05)**

Height (H)	:	0.10
Time (T)	:	0.12
Wood (W)	:	0.11
W x H	:	0.13
H x T	:	0.18
W x T	:	0.16
H x T x W	:	0.21

**Table-30 : Number of leaves in walnut epicotyl grafting during the year 2012**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Height	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	6.33	5.33	4.33	5.33	5.38	5.38	4.26	5.21	5.27	5.09	4.35	4.90	
2	5.66	4.31	3.60	4.55	5.30	4.26	4.31	4.77	4.70	5.42	4.29	3.09	
4	6.02	4.32	4.18	4.19	4.66	4.06	3.33	4.21	4.60	5.40	4.19	3.83	
6	4.66	3.66	3.36	4.33	5.33	4.31	3.30	3.88	4.20	4.99	3.99	3.63	
Mean	5.66	4.41	3.97	4.69	5.24	4.44	3.80	4.63		5.20	4.37	4.03	

**C.D(p≤0.05)**

Height (H)	:	0.09
Time (T)	:	0.11
Wood (W)	:	N.S
W x H	:	0.15
H x T	:	0.15
W x T	:	0.16
H x T x W	:	0.24

15 day old stocks. The minimum number of leaves (3.30) were recorded at interaction of height x time x scion wood under grafting performed at 6cm height on 45 day old stocks using basal portion of scion wood.

#### **4.2.7 Leaflet size (cm<sup>2</sup>)**

The perusal of data in Table-31 reveals that height of grafting, scion wood used and time of grafting had a significant effect on leaflet size in walnut epicotyl grafting during the year 2011. Maximum leaflet size (38.52 cm<sup>2</sup>) was recorded under grafting performed just at the point of attachment of cotyledons. The leaflet size recorded at 2cm height (38.30 cm<sup>2</sup>) and 4 cm (36.26 cm<sup>2</sup>) were at par with each other. Among the grafting heights minimum scion leaflet size (36.29 cm<sup>2</sup>) was obtained in grafting performed at the height of 6cm.

Significantly higher leaflet size (38.76 cm<sup>2</sup>) was recorded under grafting performed on 15 day old stocks followed by 37.91cm<sup>2</sup> in grafting performed on 30 day old stocks. The lowest leaflet size (35.81cm<sup>2</sup>) was recorded in grafting performed on 45 day old stocks. The portion of scion wood used also affected leaflet size and higher leaflet size (37.87 cm<sup>2</sup>) was recorded under terminal portion of scion wood than the basal portion (37.11 cm<sup>2</sup>).

The perusal of data in Table-31 further reveals that interactions between the height of grafting, time of grafting and scion wood used had a significant effect on leaflet size during 2011. Maximum leaflet size (39.08 cm<sup>2</sup>) was recorded under interaction of wood x height in grafting performed at the point of attachment of cotyledons using terminal portion of scion wood followed by 38.46 cm<sup>2</sup> leaflet size in grafting performed at the height of 2cm using terminal portion of scion wood. Among wood x height interaction the lowest leaflet size (36.19 cm<sup>2</sup>) was recorded under grafting performed at the height of 6cm using basal portion of scion wood.

Height x time interaction was statistically significant and higher leaflet size (39.86 cm<sup>2</sup>) was observed in grafting performed at the point of attachment of

cotyledons on 15 day old stocks. The next higher leaflet size (39.37 cm<sup>2</sup>) was recorded in grafting performed at the 2cm height on 15 day old stocks. Among height x time interactions the lowest leaflet size (35.39 cm<sup>2</sup>) was obtained under grafting performed at the 6cm height on 45 day old stocks.

Wood x time interaction was also effective which revealed significantly higher leaflet size (39.82 cm<sup>2</sup>) in grafting performed on 15 day old stocks using terminal portion of scion wood. The next higher leaflet size (38.80 cm<sup>2</sup>) was recorded on 30 day old stocks using terminal portion of scion wood. The minimum leaflet size (34.61 cm<sup>2</sup>) was observed in grafting performed on 45 day old stocks using basal portion of scion wood.

The interactions between height of grafting x time of grafting x scion wood used was significant and highest leaflet size (40.33 cm<sup>2</sup>) was observed when grafting was performed at the point of attachment of cotyledons on 15 day old stocks using terminal portion of scion wood followed by leaflet size (39.98 cm<sup>2</sup>) when grafting was performed at 2cm height on 30 day old stocks using terminal portion of scion wood. Among height x time x scion wood interactions the lowest leaflet size (34.09 cm<sup>2</sup>) was recorded under grafting performed at the height of 6cm on 45 day old stocks using basal portion of scion wood.

During second year (Table-32) also the height of grafting, time of grafting and portion of scion wood used had a significant effect on leaflet size in walnut epicotyl grafting. The maximum leaflet size (38.78 cm<sup>2</sup>) was recorded in grafting performed at the point of attachment of cotyledons. At 2cm grafting height leaflet size (38.61 cm<sup>2</sup>) obtained was significantly better than grafting at 4 cm height (37.88 cm<sup>2</sup>). Minimum leaflet size (37.13 cm<sup>2</sup>) was observed under grafting performed at the height of 6cm.

Time of grafting had a significant effect on leaflet size in walnut epicotyl grafting. Highest leaflet size (40.23 cm<sup>2</sup>) was recorded in grafting performed on 15 day old stocks. The next higher leaflet size (38.43 cm<sup>2</sup>) was observed under

grafting performed on 30 day old stocks. Lowest leaflet size (34.90 cm<sup>2</sup>) was recorded in grafting performed on 45 day old stocks. The portion of scion wood also exerted influence on leaflet size and significantly higher leaflet size (38.24 cm<sup>2</sup>) was recorded under terminal portion of scion wood as compared basal portion of scion wood (37.47 cm<sup>2</sup>).

The data in Table-32 further reveals that interactions between height of grafting, time of grafting and scion wood used were statistically significant. Maximum leaflet size (38.93 cm<sup>2</sup>) was recorded under grafting performed at the point of attachment of cotyledons using terminal portion of scion wood. At 2cm grafting height using terminal portion of scion wood leaflet size of 38.72 cm<sup>2</sup> observed was at par with grafting performed at the point of attachment of cotyledons using basal portion of scion wood (38.64cm<sup>2</sup>). Minimum leaflet size (37.00 cm<sup>2</sup>) was recorded under interaction between scion wood x height under grafting performed at 6cm height using basal portion of scion wood.

The interaction between height x time of grafting were also statistically significant. Highest leaflet size (41.13 cm<sup>2</sup>) was recorded under grafting performed at the point of attachment of cotyledons on 15 day old stocks. The next higher (40.44 cm<sup>2</sup>) was recorded under grafting performed at 2cm height on 15 day old stocks. Among the time x height interactions the lowest leaflet size (33.95 cm<sup>2</sup>) was observed in grafting performed on 45 day old stocks at a height of 6cm.

Scion wood x time interaction was found effective which revealed significant results (Table-32). Highest leaflet size (40.56 cm<sup>2</sup>) was observed under grafting performed on 15 day old stocks using terminal portion of scion wood followed by grafting performed on 30 day old stocks using terminal portion of scion wood which was to the tune of 39.48 cm<sup>2</sup>. Among scion wood x time interaction the lowest leaflet size (34.14 cm<sup>2</sup>) was observed in grafting performed on 45 day old stocks using basal portion of scion wood.

**Table-31: Leaflet size (cm<sup>2</sup>) in walnut epicotyl grafting during the year 2011**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Height	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	40.33	39.09	37.82	39.08	39.40	38.54	35.99	37.97	38.52	39.86	38.81	36.90	
2	39.98	39.41	36.00	38.46	38.77	38.51	37.14	38.14	38.30	39.37	38.96	36.57	
4	39.88	39.27	33.53	37.56	40.01	33.22	35.25	36.16	36.26	39.14	36.24	34.39	
6	39.09	37.44	38.69	36.40	37.68	37.80	34.09	36.19	36.29	35.88	37.62	35.39	
Mean	39.82	38.80	35.01	37.87	37.71	37.01	34.61	37.11		38.76	37.91	35.81	

**C.D(p≤0.05)**

Height (H)	:	0.13
Time (T)	:	0.11
Wood (W)	:	0.16
W x H	:	0.19
H x T	:	0.25
W x T	:	0.20
H x T x W	:	0.23

**Table-32: Leaflet size (cm<sup>2</sup>) in walnut epicotyl grafting during the year 2012**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Height	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	41.59	39.58	35.63	38.93	40.67	39.63	35.63	38.64	38.78	41.13	39.60	35.63	
2	40.85	40.37	34.96	38.72	40.03	38.66	36.84	38.51	38.61	40.44	39.51	35.90	
4	40.09	39.85	34.13	38.02	40.71	32.40	34.13	35.74	37.88	40.40	36.12	34.13	
6	39.74	38.14	33.95	37.27	38.23	38.83	33.92	37.00	37.13	38.98	38.48	33.95	
Mean	40.56	39.48	35.66	38.24	39.91	37.38	34.14	37.47		40.23	38.43	34.90	

**C.D(p≤0.05)**

Height (H)	:	0.10
Time (T)	:	0.13
Wood (W)	:	0.14
W x H	:	0.20
H x T	:	0.24
W x T	:	0.24
H x T x W	:	0.27

The interactions between height of grafting x scion wood used x time of grafting were statistically significant during 2012 also. Highest leaflet size (41.59 cm<sup>2</sup>) was observed at the interaction of height x time of grafting x scion wood under grafting performed at the point of attachment of cotyledons on 15 day old stocks using terminal portion of scion wood. Among interactions of height x time x scion wood lowest leaflet size (33.92 cm<sup>2</sup>) was recorded in grafting performed at 6cm height on 45 day old stocks using basal portion of scion wood.

#### **4.2.8 Plant height (cm)**

The perusal of data in Table-33 reveals that height of grafting, scion wood used and time of grafting had a significant influence on plant height during 2011. Maximum plant height (20.16 cm) was observed under grafting performed at the height of 6cm. The next highest plant height (18.88 cm) was recorded in epicotyl grafting performed at 4cm height. Among grafting heights lowest plant height (17.84 cm) was recorded in grafting performed at the point of attachment of cotyledons.

Time of grafting had a significant effect on plant height (Table-33). Highest plant height (19.24 cm) was recorded in grafting performed on 15 day old stocks. The plant height obtained in grafting performed on 30 day old stocks was 19.13 cm. Lowest plant height (17.70 cm) was recorded in grafting performed on 45 day old stocks. Significantly higher plant height (19.14 cm) was recorded under terminal portion of scion wood which was 18.41 cm in case of basal portion of scion wood used.

The data in Table-33 further reveals that the interactions between height of grafting, time of grafting and scion wood used were statistically significant. Maximum plant height (20.43 cm) was observed at interaction of scion wood x height when grafting was performed at the height of 6cm using terminal portion of scion wood. The next higher plant height (19.85 cm) was recorded in grafting performed at the 4cm height using terminal portion of scion wood. Among

interaction of scion wood x height lowest plant height (17.44 cm) was observed under grafting performed at the point of attachment of cotyledons using basal portion of scion wood.

Height x time interaction produced maximum plant height (20.22 cm) was recorded under grafting performed at the 6cm height on 15 day old stocks. The next higher plant height (20.02 cm) was recorded in grafting performed at the 6cm height on 30 day old stocks. Among the interaction between height and time lowest plant height (17.20 cm) was observed in grafting performed at the point of attachment of cotyledons on 45 day old stocks.

Plant height of 19.59 cm was observed under interaction of wood x time in grafting performed on 15 day old stocks using terminal portion of scion wood. The next higher plant height (19.19cm) recorded in grafting conducted on 15 day old stocks using basal portion of scion wood. Among wood x time interaction lowest plant height (17.06 cm) was obtained in grafting performed on 45 day old stocks using basal portion of scion wood.

The interactions between height of grafting x scion wood used x time of grafting were significant during 2011. Highest plant height (20.90 cm) was recorded in grafting performed at 6cm height on 15 day old stocks using terminal portion of scion wood. The next higher plant height (20.85 cm) was recorded in grafting performed at 6cm height on 30 day old stocks using terminal portion of scion wood. The former and later interactions were at par with each other. Among interactions between height of grafting x time of grafting x scion wood used the lowest plant height (16.16 cm) was recorded in grafting performed at the point of attachment of cotyledons on 45 day old stocks using basal portion of scion wood.

During second year also the height of grafting, scion wood and time of grafting had a significant effect on plant height (Table-34). Maximum plant height (19.77 cm) was observed in grafting performed at the 6cm height followed by grafting at 4cm (18.47 cm). Among the grafting heights the lowest plant height

(18.00 cm) was observed in grafting performed at the point of attachment of cotyledons.

Time of grafting significantly affected plant height as revealed in Table-34. Highest plant height (19.14 cm) was recorded in grafting performed on 15 day old stocks. The next higher plant height (18.98 cm) was observed in grafting performed on 30 day old stocks. Among time of grafting lowest plant height (17.72 cm) was observed in grafting performed on 45 day old stocks. The portion of scion wood also influenced plant height significantly and higher plant height (18.89 cm) was observed in grafting using terminal portion scion wood which was 18.34 cm in case of basal portion of scion wood.

The perusal of data in Table-34 reveals that the interactions between height of grafting x scion wood were statistically significant. Maximum plant height (20.07 cm) was recorded under interaction of scion wood x height under grafting performed at 6cm height followed by 4cm (18.69 cm) using terminal portion of scion wood. Among the interaction of scion wood x height lowest plant height (17.51 cm) was obtained in grafting performed at the point of attachment of cotyledons using basal portion of scion wood.

Height x time interaction also influenced plant height significantly and higher plant height (19.92 cm) was recorded in grafting performed at 6 cm height on 15 day old stocks. The next higher plant height (19.68 cm) was recorded on 30 day old stocks in grafting performed at 6cm height. Among interaction of height x time lowest plant height (17.05 cm) was obtained in grafting performed at the point of attachment of cotyledons on 45 day old stocks.

Maximum plant height (19.60 cm) was recorded under interaction of wood x time in grafting performed on 15 day old stocks using terminal portion of scion wood. The next higher plant height (19.22 cm) was recorded in epicotyl grafting performed on 30 day old stocks using terminal portion of scion wood. The lowest

**Table-33 : Plant height (cm) in walnut epicotyl grafting during the year 2011**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Height	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	18.42	18.39	17.95	18.05	18.04	17.83	16.16	17.44	17.84	18.23	18.11	17.20	
2	19.23	18.25	17.28	18.25	18.26	18.16	16.41	17.51	17.88	18.74	18.20	16.69	
4	19.82	19.34	17.40	18.85	19.75	19.62	17.36	18.91	18.88	19.78	19.48	17.38	
6	20.90	20.85	20.74	20.43	20.74	20.60	18.34	19.79	20.16	20.02	20.72	19.54	
Mean	19.29	19.20	18.34	19.14	19.19	19.05	17.06	18.41		19.24	19.13	17.70	

**C.D(p≤0.05)**

Height (H)	:	0.10
Time (T)	:	0.08
Wood (W)	:	0.12
W x H	:	0.11
H x T	:	0.09
W x T	:	0.13
H x T x W	:	0.16

**Table-34 : Plant height (cm) in walnut epicotyl grafting during the year 2012**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal			Mean	Height	Time		
	15 DAG	30 DAG	45 DAG			15 DAG	30 DAG	45 DAG			15 DAG	30 DAG	45 DAG
0	18.73	18.69	18.05	18.49	18.21	18.08	16.25	17.51	18.00	18.47	18.38	17.05	
2	19.06	18.8	17.10	18.32	18.16	17.73	18.16	18.01	18.16	18.61	18.26	17.63	
4	19.71	19.04	17.34	18.69	19.45	19.11	16.72	18.26	18.47	19.58	19.07	17.13	
6	20.70	20.35	20.17	20.07	20.14	20.07	18.01	19.47	19.77	19.92	19.68	19.09	
Mean	19.60	19.22	18.16	18.89	18.99	18.74	17.28	18.34		19.14	18.98	17.72	

**C.D(p≤0.05)**

Height (H)	:	0.09
Time (T)	:	0.06
Wood (W)	:	0.10
W x H	:	0.12
H x T	:	0.13
W x T	:	0.15
H x T x W	:	0.17

plant height (17.28 cm) was recorded in grafting conducted on 45 day old stocks using basal portion of scion wood.

The interactions between height x time x scion wood reveal that highest plant height (20.70 cm) was recorded under grafting performed at 6cm height on 15 day old stocks using terminal portion of scion wood. The next higher plant height (20.35 cm) was obtained in grafting performed at 6cm height on 30 day old stocks using terminal portion of scion wood. Among interactions of height x time x scion wood the lowest plant height (16.25 cm) was observed under grafting performed at point of attachment of cotyledons on 45 day old stocks using basal portion of scion wood

#### **4.2.9 Stem diameter (mm)**

The perusal of data in Table-35 reveals that height of grafting, time of grafting and scion wood had a significant effect on stem diameter during 2011. Maximum stem diameter (3.73 mm) was recorded in grafting performed at the height of 4cm followed by stem diameter (3.59 mm) in grafting performed at 2cm height. The lowest stem diameter (3.43 mm) was recorded in grafting performed at the height of 6cm.

Time of grafting had a significant effect on stem diameter and higher stem diameter (3.67 mm) was recorded in grafting performed on 15 day old stocks. The next higher stem diameter (3.59 mm) was observed in grafting performed on 30 day old stocks. The lowest stem diameter (3.41 mm) was recorded in grafting performed on 45 day old stocks. Significantly higher stem diameter (3.67 mm) was recorded under terminal portion of scion wood which was 3.45 mm under basal portion of scion wood.

The interactions between height of grafting x time of grafting x scion wood used were significant. Maximum stem diameter (3.95 mm) was recorded in grafting performed at the height of 4cm using terminal portion of scion wood. The next higher stem diameter (3.59 mm) was observed in grafting performed at 2cm

height using terminal portion of scion wood. Among interaction of wood x height the lowest stem diameter (3.37 mm) was recorded in grafting performed at 6cm height using basal portion of scion wood.

Height x time interaction recorded maximum stem diameter (3.87 mm) in grafting was performed at 4cm height on 15 day old stocks followed by stem diameter (3.77 mm) in grafting performed on 30 day old stocks. Among the interaction of height x time lowest stem diameter (3.32 mm) was recorded under interaction of height x time in grafting performed at the height of 6cm on 45 day old stocks.

The data in Table-35 indicates that wood x time interaction was also effective in influencing stem diameter. Maximum stem diameter (3.81 mm) was recorded in grafting performed on 15 day old stocks using terminal portion of scion wood followed by stem diameter (3.71mm) in grafting performed on 30 day old stocks using terminal portion of scion wood. Among interaction between wood x time lowest stem diameter (3.36 mm) was recorded in grafting performed on 45 day old stocks using basal portion of scion wood.

The interactions between height of grafting x scion wood x time of grafting were significant during 2011. Highest stem diameter (4.13 mm) was recorded under interaction of height x time x scion wood under grafting performed at 4cm height on 15 day old stocks using terminal portion of scion wood. The next higher stem diameter (4.03 mm) was recorded in grafting was performed at 4cm height on 30 day old stocks using terminal portion of scion wood. Among interactions of height x scion wood x time lowest stem diameter (3.21 mm) was observed in grafting performed at the 6cm height on 45 day old stocks using basal portion of scion wood.

The data on stem diameter for the year 2012 is given in Table-36. The perusal of data reveals that height of grafting, time of grafting and scion wood used had a significant effect on stem diameter during second year. Highest stem

diameter (3.63 mm) was recorded when grafting was performed at the height of 4cm followed by stem diameter (3.56 mm) recorded in grafting performed at the height of 2cm. Lowest stem diameter (3.44 mm) was observed in grafting performed at the height of 6cm.

Time of grafting also had a significant effect on stem diameter (Table-36). Highest stem diameter (3.68 mm) was recorded under grafting performed on 15 day old stocks. The next higher stem diameter (3.56 mm) was recorded in grafting performed on 30 day old stocks. Lowest stem diameter (3.40 mm) was in grafting performed on 45 day old stocks. Significantly higher stem diameter (3.65 mm) was recorded in terminal portion of scion wood in comparison to basal portion of scion wood used (3.56mm).

The perusal of data in Table-36 further reveals that interactions between height of grafting, scion wood and time of grafting had a significant effect on stem diameter. Highest stem diameter (3.90 mm) was observed at interaction of wood x height in grafting performed at the height of 4cm. Among the interaction of wood x height lowest stem diameter (3.36 mm) was recorded in grafting performed at the height of 6cm.

The interaction between height x time recorded maximum stem diameter (3.91 mm) under grafting performed at the height of 4cm on 15 day old stocks. The next higher stem diameter (3.64 mm) was recorded in grafting performed at 2cm height on 15 day old stocks. Among interaction between height x time lowest stem diameter (3.30 mm) was observed in grafting performed at the height of 6cm on 45 day old stocks.

Wood x time interaction also had a significant effect on stem diameter (Table-36). Highest stem diameter (3.85 mm) was recorded under interaction between wood x height in grafting was performed on 15 day old stocks using terminal portion of scion wood. The next higher stem diameter (3.64 mm) was recorded in grafting performed on 30 day old stocks using terminal portion of

scion wood. Lowest stem diameter (3.37 mm) was observed at interaction of wood x height in grafting performed on 45 day old stocks using basal portion of scion wood.

The interactions between height of grafting x time of grafting x scion wood were statistically significant during second year as well. Highest stem diameter (4.21 mm) was observed under interactions of height x time x scion wood in grafting performed at the height of 4cm on 15 day old stocks using terminal portion of scion wood. The lowest stem diameter (3.30 mm) under interactions height x time x scion wood was observed in grafting performed on 45 day old stocks at 6cm height using basal portion of scion wood.

**Table-35 : Stem diameter (mm) in walnut epicotyl grafting during the year 2011**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Stem diameter	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	3.90	3.69	3.36	3.55	3.61	3.56	3.30	3.49	3.57	3.75	3.62	3.33	
2	3.75	3.57	3.47	3.59	3.58	3.50	3.31	3.42	3.59	3.66	3.53	3.41	
4	4.13	4.03	3.69	3.95	3.62	3.52	3.43	3.52	3.73	3.87	3.77	3.56	
6	3.59	3.40	3.49	3.49	3.28	3.49	3.21	3.37	3.43	3.43	3.44	3.32	
Mean	3.81	3.71	3.52	3.67	3.52	3.51	3.36	3.45		3.67	3.59	3.41	

**C.D(p≤0.05)**

Height (H)	:	0.03
Time (T)	:	0.05
Wood (W)	:	0.22
W x H	:	0.09
H x T	:	0.03
W x T	:	0.11
H x T x W	:	0.14

**Table-36 : Stem diameter (mm) in walnut epicotyl grafting during the year 2012**

Height of grafting (cm)	Scion –wood used								Factor Means				
	Terminal				Mean	Basal				Stem diameter	Time		
	15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG	Mean		15 DAG	30 DAG	45 DAG
0	3.83	3.65	3.30	3.59	3.63	3.59	3.38	3.53	3.50	3.53	3.62	3.40	
2	3.79	3.51	3.41	3.57	3.50	3.52	3.29	3.43	3.56	3.64	3.51	3.35	
4	4.21	3.81	3.70	3.90	3.68	3.47	3.46	3.53	3.63	3.91	3.72	3.58	
6	3.63	3.43	3.50	3.52	3.30	3.40	3.38	3.36	3.44	3.46	3.41	3.30	
Mean	3.85	3.64	3.47	3.65	3.52	3.49	3.37	3.59		3.68	3.56	3.40	

**C.D(p≤0.05)**

Height (H)	:	0.02
Time (T)	:	0.03
Wood (W)	:	0.06
W x H	:	0.07
H x T	:	0.05
W x T	:	0.09
H x T x W	:	0.12

## Chapter – 5

### DISCUSSION

The present investigation entitled “Studies on Seed Germination and Epicotyl grafting in Walnut” was carried out at Fruit Plant Nursery Division of Fruit Science, SKUAST-Kashmir, Srinagar, J&K during the year 2011 and 2012. Salient findings of the present investigation are discussed under the relevant headings in the following pages.

#### 5.1 Studies on seed germination and seedling growth in walnut

##### 5.1.1 Germination percentage

It is clear from results that all the chemical treatments proved significant in breaking seed dormancy. Among the chemicals used highest germination was produced through use of GA<sub>3</sub> and nuts treated with 100 ppm GA<sub>3</sub> gave highest germination (58.24 and 58.57%) followed by 50 ppm GA<sub>3</sub> in which germination percentage was to the tune of 54.23 and 56.32 percent. The higher germination with GA<sub>3</sub> is due to diffusion of endogenous auxins and gibberellins like substances or might have antagonized the effects of inhibitors present in the seed (Mathur *et al.*, 1971 and Sastry and Muir, 1963). Exogenous application of GA<sub>3</sub> causes absorption of GA<sub>3</sub> and consequently growth inhibiting substances decrease thus there was an increase in catalase activity and ultimately an increase in the germination of walnut seeds (Kelly, 1969). The results of present investigation are in agreement with Dahab *et al.* (1975) who found that *Pistachio vera* seeds obtained more than 95% germination after soaking in 100 ppm GA<sub>3</sub> solution before sowing. Casini and Conticni (1979) immersed *Pistachio* seeds in 50 ppm GA<sub>3</sub> for seven days and was able to increase seed germination from 50-79 per cent. Ak *et al.* (1995) has obtained 73.30 per cent seed germination in *Pistachio* nuts when seeds were soaked for 48 hours in 125 ppm GA<sub>3</sub>. Samaan *et al.* (2000) found that 15 day chilling and soaking in GA<sub>3</sub> gave an average germination of 84.78 per cent when apricot seeds were soaked in GA<sub>3</sub> 250 ppm with stratification

period of 3 weeks. According to Mehanna *et al.* (1985) Gibberellins eliminate the chilling requirements of peach and apple seeds and thereby increase their germination.

The next higher germination (49.66 and 48.57%) was produced through use of 7.5 per cent sulphuric acid. Immersion of seed in sulphuric acid disrupts the seed coat. Sulphuric acid 7.5 per cent gave highest percentage of germination as compared to 9.5 per cent sulphuric acid (47.99 and 46.49%) indicate that more rapidly the endocarp is ruptured the faster the rate of germination. However, prolonged immersion may be injurious to the seeds as acid may rupture vital parts of embryo (Levitt, 1974). Sulphuric acid is thought to disrupt the seed coat and expose the lumens of the macrosclereid cells, permitting imbibition of water which triggers germination (Nikoleave, 1977). The results are in agreement with the findings of Nasir *et al.* (2010) who reported that in acid scarification, seeds of almond nuts scarified with 7.5 per cent concentrated H<sub>2</sub>SO<sub>4</sub> for 3 minutes gave highest germination of 50 per cent and when treated with 9.5 per cent for 3 minutes gave only 28.6 germination percentage. According to Sehgal and Singh (1990) higher concentrations of sulphuric acid were less effective compared to lower concentrations and that scarification by H<sub>2</sub>SO<sub>4</sub> and stratification at 5°C for 30 days increased seed germination in *Pistachio* nut up to 90.70 per cent. Heidari *et al.* (2008) immersed almond seeds in different concentrations of H<sub>2</sub>SO<sub>4</sub> for 10 to 60 minutes and stratified the seeds for 15, 30 and 45 days. The immersion of almond seeds in H<sub>2</sub>SO<sub>4</sub> for 10 minutes in combination with stratification for 30 days improved germination percentage of upto 90 per cent.

In the present investigation potassium nitrate (0.4%) significantly improved germination percentages (44.24 and 43.83) in comparison to potassium nitrate 0.2 per cent (41.24 and 40.66%). Potassium nitrate is thought to play role in biosynthesis of auxins and increases probability of germination (Alboresi *et al.*, 2005). The results are in agreement with Bhan and Sharma (2007) who recorded significantly higher germination (91.24%) in *Prunus armeniaca* (wild apricot)

seeds when treated with  $\text{KNO}_3$  (0.3%). Pillay (1962) reported that potassium nitrate had positive effects on the germination of seeds with and without coat. Soaking in 7,500 ppm and 10,000 ppm  $\text{KNO}_3$  gave the most significant germination rates: 64.54 per cent for seeds with coat and 74.24 per cent for seeds without coat, respectively. Moreover, all the concentrations tested increased the germination of mazzard cherry (*Prunus avium*) seeds. Due to the influence of  $\text{KNO}_3$  on seed germination it has been an important seed treatment in seed testing laboratories for many years without a good explanation for its action. (Copeland and Mc Donald, 1995 and Basra, 1994).

Ethrel used in present investigation although lesser effective in inducing good percentage of seed germination was statistically superior when compared to control. Ethrel 1500 ppm resulted higher germination percentage (38.07 and 38.82) than the ethrel at 1000 ppm (35.58 and 38.78%). The latter was also superior when compared to control. The results are in agreement to the findings of Kumar *et al.* (1988) who reported that higher concentrations of ethanol (5%) was more effective in promoting maximum seed germination in pear. Etherl possibly plays a role in enhancement of endogenous level of hormones. Applications of ethrel have been reported to induce seed germination of various plant species (Salisbury and Ross, 1986).

In the present investigation during 2011 and 2012 respectively. Cold treatment of 30 days resulted maximum germination per cent (46.51 and 46.56%) compared to ambient stored seeds (42.21 and 42.79%). Due to cold temperatures, more oxygen is soluble in water, so the oxygen requirement of embryo is better satisfied. Cold treatment of seeds is also important to meet the chilling units of walnut which is between 700-1000 hours. Seeds of temperate fruits such as peach, plum, apple and nuts require a definite period of cold treatment for uniform germination (Hartman and Kester, 1968). The results are in line with Powell (1987) who has justified it with the explanation that stratification in cold instantly stimulates the structural GA synthesis. While GA in the structure increases the

enzymatic activity, it slows the ABA activity. According to Ruduicki (1977) the percentage of seed germination in walnut increased with the increase in stratification period. The low temperature stratification probably releases the seed from dormancy and promotes germination, due to its effect on the activity of protease and lipase enzymes involves in the mobilization of stored food material in the seeds. Kester *et al.* (1977) and Grassley (1977) reported that almond seeds treated with different (30 and 60 day) stratification periods had significantly higher germination percentage whereas non-stratified seeds did not germinate. Ji and Wang (1987) reported that during cold treatment of four weeks followed by pre-soaking treatments ABA levels possibly decreased and GA levels increase.

In the present investigation seeds without soaking in water and treated with chemicals had a higher germination percentage (45.36 and 45.53%) compared to soaked ones (43.36 and 43.83%). Research work conducted by other workers also support direct application of chemicals as concentrated chemicals probably dilute the effects of inhibitors very fast. These results are in agreement with Frutos (1993) who reported that direct application of chemicals on dry nut led to the best results as for as germination is concerned. Soaking of seeds prior to chemical treatment probably produced a lesser entry of chemical solutions into the seeds. In fact, gradient of humidity between chemical solutions and tissue of seeds ought to be bigger in no water treatment, therefore the effect of this extra chemical probably increase the rate of germination.

The germination percentage of walnut seed increased with treatments combining of 30 day chilling plus subsequent soaking in GA<sub>3</sub> in comparison to other treatment combinations. Highest germination percentage (63.49 and 62.66) was obtained when cold treated nuts were soaked in GA<sub>3</sub> 100 ppm for 12 hours. The results are in agreement with Aslamarz *et al.* (2007) who had reported that percent of seed germination increased with the cold stratification in walnut seeds (36.8 in the control compared with 86.3% after eight weeks). Also the mean time to germinate significantly decreased with the chilling treatment. Samaan *et al.*

(2000) had recorded highest percentages of seed germination in apricot with treatment combination of 15 days chilling plus subsequent soaking in GA<sub>3</sub>. Ozguven *et al.* (1995) reported that the concentration of ABA like substances was high when Pistachio seeds were soaked for 12 hours in Gibberellic acid but decreased with the time of stratification. Dahshan *et al.* (1987) had also obtained highest germination (84.78%) with GA<sub>3</sub> at 250 ppm plus stratification for 3 weeks at 5°C.

In the present investigation Gibberellic acid, sulphuric acid, potassium nitrate and ethrel were found helpful in promoting the seed germination under 30 day cold period compared to control. It is obvious from these results that the chemical treatments helped in shortening the chilling requirement of walnut seeds. The results obtained in present investigation are also in agreement with Garcia-Gusano *et al.* (2004) who observed higher germination percentage in wild almond (*Prunus scoparia*) seeds after soaking nuts in 500 ppm of gibberellic acid. Aygun *et al.* (2008) reported that hazelnut seeds soaked in water for 24 hours and stratified into moist peat and incubated at 4°C for 100, 110 and 120 days had resulted germination of 65-67 per cent. Singh *et al.* (1966) found that GA<sub>3</sub> in agar medium broke dormancy of peach seeds and gave 35 per cent germination during 13 days at 25°C and a similar level of germination was obtained only after two months cold storage in untreated seeds. Sewal and Sharma (1992) reported that gibberellic acid helped in promoting seed germination in two cultivars of plum i.e., Santa Rosa and Green even without stratification. Sharma and Singh (1980) have revealed that release of seed dormancy was dependent on increase in quantity of acidic type of Gibberellins and on giving stratification at low temperature (7-10 °C) to dormant peach seeds, the level of basic type of gibberellins increases. Kinally (1986) reported that with increase in stratification period, GA<sub>3</sub>, IAA, soluble sugars, soluble amino acids and soluble phenol content increase in peach seeds. Change in these endogenous components are closely related to the ability of seeds to germinate.

It is essential to understand the effect of stratification, chemicals and seed treatment on germination both for practical nursery applications and conservation. From the present investigation on walnut seed germination, it is clear that chemicals can substitute chilling units of walnut seeds and can serve as alternative method. We conclude that the combination between a suitable pre-chilling period and an effective level of GA<sub>3</sub> would considerably enhance walnut seed germination.

### **5.1.2 Seedling growth of walnut**

It is clear from the results that height of seedling was increased with GA<sub>3</sub>, sulphuric acid, potassium nitrate and ethrel chemicals used in the present investigation. All concentrations of GA<sub>3</sub>, sulphuric acid, potassium nitrate and ethrel increased height of seedling in comparison to control. The maximum height (23.24 and 21.73 cm) was obtained under GA<sub>3</sub> 100 ppm. The increase in seedling growth parameters with GA might be related to the fact that GA promotes stem, and shoot elongation through both cell division and internodal elongation in higher plant. (Hartmann *et al.*, 2002; Hopkins and Huner 2004 and Harris *et al.*, 2004). The results are in agreement with Negi *et al.* (2010) who reported increased seedling height when Pistachio seeds were treated with GA<sub>3</sub> 200 ppm. Rahemi and Baninasab (2000) also reported that GA<sub>3</sub> application lead to increased height of seedlings in two wild species of pistachio. Bal *et al.* (1990) reported maximum seedling height in pear under GA<sub>3</sub> 50 ppm.

Sulphuric acid also increased the height of seedling and might be related to the fact that sulphuric acid treated seeds germinated early due to softening of endocarp as a result of which cell division and elongation of cells had taken place early and mean time growth continued for a longer period. The results are in agreement with Mabundza *et al.* (2010) who reported that sulphuric acid as pre-germination treatment, gave seedling height of 3.0 cm, while the control gave the lowest seedling height (1.6 cm). Ehiagbanare and Onyibe (2007) reported that chemical scarification of passion fruit seeds increased germination percent and

seedling growth under various conditions. Ryndin (1962) has reported that soaking seeds of plum, cherry and apple in the solution of Gibberellic acid for 12 hours increased height of seedling.

The height of seedlings was also observed to be influenced by seed storage treatments. Maximum height of seedling (21.28 and 17.93 cm) resulted from cold treatment of 30 days. Cold treatment is the process of pre-treating seeds to simulate natural processes that a seed must endure before germination. In nature, moist chilling occurs in wet soils combined with winter conditions. The results are in agreement with Ercisli and Guleryuz (1995) who reported that increasing stratification period increased seedling height in apricot. Nabil and Al-Ameen (2007) reported that the highest seedling height (18.70 cm) was observed in pistachio nut when 90 days stratification was applied. Flemion and Beardow (1963) reported that in low temperature stratification treatments of Sharabti peach seeds at 7°C, the inhibitor content decreased and growth promoting substances like auxins and gibberellins were increased which helped in raising of normal and vigorous peach seedlings.

The results of present study also propounded that GA<sub>3</sub>, sulphuric acid and potassium nitrate caused increase in stem diameter of walnut seedlings compared to control. Highest stem diameter (4.04 and 4.02 mm) was recorded in seeds treated with GA<sub>3</sub>100 ppm. Gibberellins promote cell division and cell enlargement which lead to stem growth. Better growth of seedlings might have probably helped in increasing stem diameter. The results are in agreement with Rahemi and Baninasab (2000) who reported that stem diameter was enhanced in hazelnut with application of GA<sub>3</sub>. Stratification days have also helped in increasing stem diameter of walnut seedlings. This is analogous to the findings of Ercisli and Guleryuz (1995) who have obtained highest stem diameter with GA<sub>3</sub> 250 ppm and 3 weeks stratification period.

In the present study root length was enhanced by all chemical treatments. GA<sub>3</sub>100 ppm was found more effective and recorded maximum root length

(191.57 and 194.91 mm) in comparison to other treatments and control. The results are in agreement with Tatjana Ćirković-Mitrović *et al.* (2011) who reported that the root length of the below-ground part of seedlings varied from 5.3 to 43.30 cm. Donoho and Walker (1957) also reported that Gibberellic acid increased root length in peach. Sulphuric acid was effective in promoting root length and reason might be that seeds treated with  $H_2SO_5$  germinated earlier and mobilization of photosynthates from phloem into roots take place simultaneously which leads to growth of roots. The early development of root system also leads to utilization of stored food reserved in hypo cotyledons. Potassium nitrate also enhanced root length of seedlings in comparison to control.  $KNO_3$  is helpful for reactivation of metabolic process of seeds. This compound also helps in biosynthesis of auxins which ultimately trigger growth of embryo. The present study indicates that higher concentrations of potassium nitrate were effective in producing more root length in comparison to lower concentrations. Results are in agreement with Khan *et al.* (1999) who reported that low response to nitrate might be because of the fact that some nut species may be un-responsive and concentrations tested could be sub-optimal.

The present study indicates that number of leaves was markedly influenced by application of chemical and storage treatments. Gibberellic acid,  $H_2SO_4$  and  $KNO_3$  were effective in promoting higher number of leaves. A leaf primordium is formed from the periclinal divisions of group of cells on the flank of the apex which results in broadening of apex at this region and chemical treatment represent a process involving the active participation of meristematic cells in leaf primordium. In the present study the maximum number of leaves was observed with application of 100 ppm  $GA_3$  followed by 50 ppm  $GA_3$ . The results are in agreement with Hamed and Fathi (1999) who reported that number of leaves in walnut was higher with application of  $GA_3$ . El-Nabawy *et al.* (1985) reported increased number of leaves in pecan nut with application of Gibberellic acid. Sulphuric acid also increased number of leaves in walnut seedling in the

present study. Due to early embryonic activity all the cells of embryo are capable of division.  $\text{KNO}_3$  has also significantly increased number of leaves in walnut seedlings as the seedlings exhibit property of cell division, cell enlargement and cell differentiation.  $\text{KNO}_3$  probably causes biosynthesis of auxins and therefore, might have played its role in enhancing meristematic cell division under favorable climatic conditions.

It is clear from the results that maximum leaflet size was obtained with  $\text{GA}_3$  100 ppm (74.12 and 72.70  $\text{cm}^2$ ) followed by 50 ppm (72.27 and 71.04  $\text{cm}^2$ ) in walnut seedlings. The results are in agreement with Ram Chandra and Sheo (1990) reported that in guava  $\text{GA}_3$  application increased leaf size. Muradoglu and Gundogdu (2011) have reported that average leaflet area in walnuts varied according to cultivar and leaf size ranged from 26.77 to 86.92  $\text{cm}^2$ . The present study also reveals that Sulphuric acid,  $\text{KNO}_3$  and ethrel resulted in increased leaflet size in walnut seedling compared to control which might be due to active participation of cells in growing portion.

The chlorophyll content was found non-significant under both chemical as well as storage treatments which ranged from 0.81 to 0.86 mg/g of fresh weight. Days from germination to leaf fall was also counted in given treatments. It is clear from results that senescence occurred very late in  $\text{GA}_3$  treated seedlings. Further  $\text{GA}_3$  and sulphuric acid treated seeds germinated early and therefore, difference was observed in the effects of different chemical and storage treatments. Maximum number of days from germination to leaf fall was observed with application of  $\text{GA}_3$  followed by sulphuric acid,  $\text{KNO}_3$  and ethrel.  $\text{GA}_3$  helps in solute translocation in phloem and during development it helps phloem to carry more nitrogen and other nutrients to the sites where they are required. Further exogenous application of chemicals might have resulted in an increase in endogenous levels. Although leaf fall extended for only a short period but earlier germination caused due to chemical and storage treatments resulted in significant

difference in number of days from germination to leaf fall as also reported by Sinha *et al.* (1977) in walnut and Sinha *et al.* (1977) in an another study in apples.

## **5.2 Studies on epicotyl grafting in walnut**

Epicotyl grafting of walnut is a newly introduced method in few countries and the present study is the first attempt to observe the feasibility of this method in Kashmir valley under open conditions. The success of grafting in walnuts depends on different factors such as temperature, relative humidity, scion material, height of rootstock, time and method of grafting and the skill of the grafter.

### **5.2.1 Scion take percentage**

Scion take percentage was recorded in three phases. The highest scion take percentage (67.38 during 2011 and 70.38 during 2012) was recorded after 15 days in grafting performed at point of attachment of cotyledons. Levels of compatibility of rootstock and scion improve due to proliferation of callus tissue in walnut (Liu and Han, 1984). Lowest graft take (28.16 and 28.32%) under present investigation was observed in grafting performed at 6cm height. The results are in agreement with Rongting and Pinghai (1993) who reported that walnut callus quality and amount of callus formation plays an important role in grafting success. Serdar *et al.* (2005) reported chestnut swelling of union, one of the signs of graft incompatibility and lignification of cells at the junction. No such swelling was observed in the combinations of stock and scion used in present study.

Time of grafting had a significant influence on scion take percentage and maximum scion take percent (67.49 during 2011 and 70.38 during 2012) was recorded in grafting performed on 15 day old stocks. Higher activity of meristematic cells resulting in faster formation of callus and quick healing of graft union is observed in younger rootstocks (Hartmann and Kester, 1997). Minimum scion take percent was observed in grafting performed on 45 day old rootstocks.

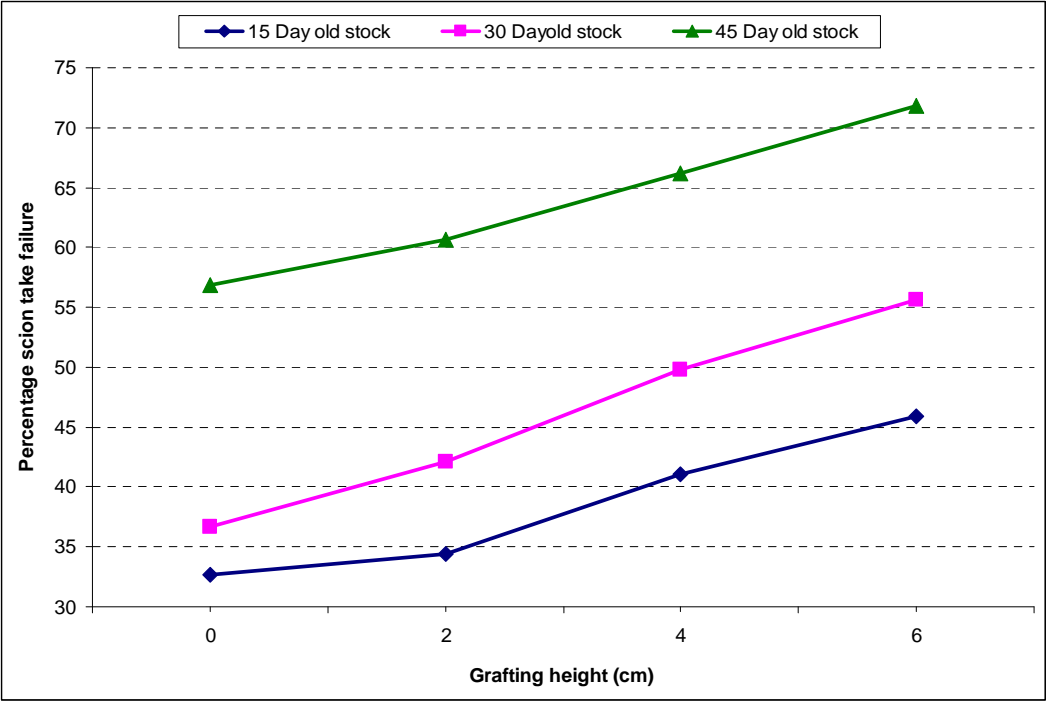
Phenolic content increases with age of rootstocks and level of compatibility of cells decrease with the result probability of graft success decreases. The results of present investigation are in line with Rongting and Pinghai (1990) who reported that variations in juglone content of walnut lead to differences in graft success. Pritiviera *et al.* (1983) who reported that accumulation of the phenolic compound like 4-hydroxynapthoquinine observed in 20-25 days old walnut and was harmful for growth of walnut callus. Dhunaga *et al.* (1989) also reported maximum graft take percentage in 5-10 days old rootstocks. Lagerstedt (1982) reported that limited exposure to higher temperatures will restrict callus formation and cause subsequent graft failure.

Scion take percent was also influenced with portion of scion taken for grafting in present investigation. Terminal scion recorded maximum scion take percent. Scion wood effect can be due to more mobilization of reserve food material and release of the same at the initiation of growth. The results are in agreement with Sul-In (2006) who observed significant graft success in terminal portion of scion used in epicotyl grafting of walnut. According to Rongting and Pinghai (1993) quality of scion affects graft take percentage. Rezaee and Vahdati (2008) have reported that time of taking the scions affects grafting success.

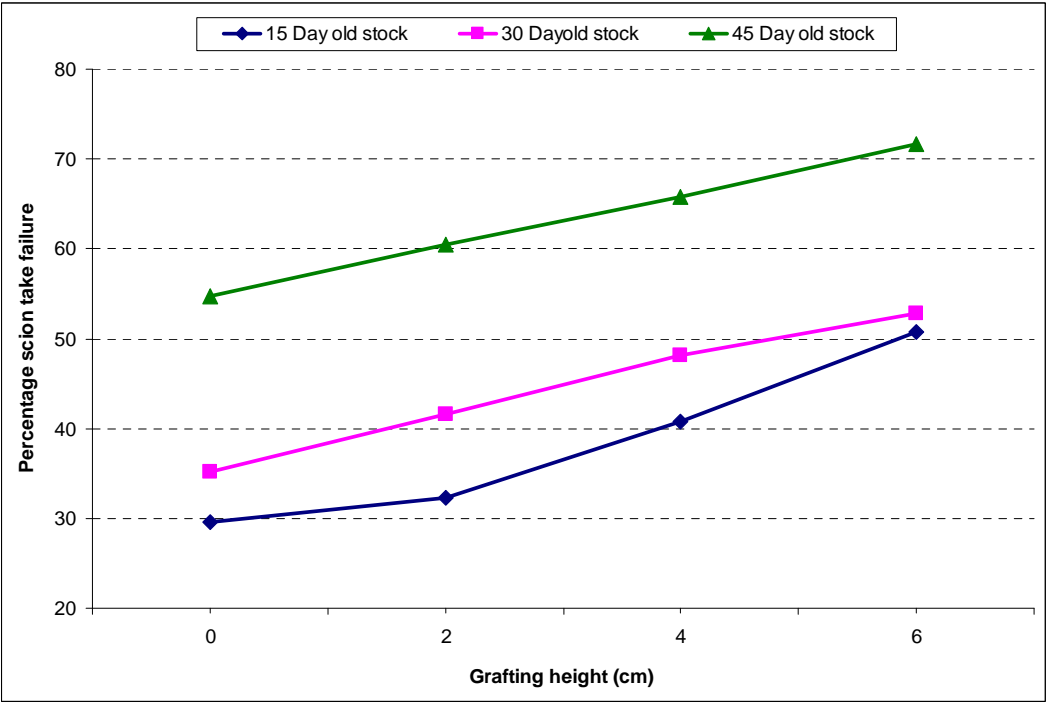
Scion take percentage was lower with the increase in height of grafting and the time of grafting. As the height of grafting increased there was corresponding increase in the scion take failure percentage (Fig. 1). Time of grafting also influenced the graft failure percentage. The later the time of grafting the more the grafting take failure was noticed (Fig. 2).

### **5.2.2 Scion sprouting percentage**

The highest scion sprouting percentage (40.93 during 2011 and 41.32 during 2012) was obtained in grafting performed at point of attachment of cotyledons under open conditions. Scion sprouting percentages reveals that vascular continuity had established between rootstock and scion. This is likely due



**Fig. 1 : Scion take failure percentage as influenced by height and time of grafting during 2011**



**Fig. 2 : Scion take failure percentage as influenced by height and time of grafting during 2012**

to lining up of cambial layers of rootstock and scion, critical requirement for successful scion sprouting (Hartmann *et al.*, 2001). The temperature and relative humidity play an important role in getting higher graft success. Temperature between 24-27°C for callus formation and high relative humidity are most congenial for quick graft union initiation which are the prerequisites for quick and stout joint development. The present results are in agreement with Duman and Serdar (2006) who reported that 55 per cent scion sprouting was observed in nurse grafting of chestnut performed on 2cm long radicle after germination and under controlled temperature 72.5 per cent scion sprouting was achieved. Gandev (2009) reported that 77.50 per cent scion sprouting was recorded in cleft grafting performed at point of attachment of cotyledons. Gandev and Arnaudov (2011) reported success percentage of epicotyl grafting in walnut ranging from 56.60 to 63.35. Sawano *et al.* (1983) reported that survival rate ranging from 80 to 100 per cent in chestnut non-lignified seedlings grafted at 4-7cm heights. Greenwell (2002) reported that possible causes of nut graft failures might result from cotyledon irregularity where the petioles form at odd angles to the cotyledons in the nut; besides Fungus/mold infections and weevil infestations also contribute to the graft failures. It is, therefore necessary to protect nuts against fungus/mold infections. According to Hartmann *et al.* (2002) temperature has a pronounced effect on callus formation and optimum temperature for callusing varies between temperate zones.

Lowest percentage of scion sprouting (17.99 during 2011 and 18.21 during 2012) was recorded in grafting performed at 6cm height. One of the reason behind low percentage of scion sprouting could be increase in respiration and transpiration by actively proliferating new shoots coupled with insufficient ascent of sap. Other reasons related to low sprouting percentage may be that phenolic content increases with the age and level of compatibility of cells decreases. The results are in agreement with Pritiviera *et al.* (1983) who reported that after 15-20

days accumulation of the phenolic compound like 4-hydroxynaphthoquinine was more and harmful for growth of walnut callus.

Sprouting percentage was statistically influenced by time of grafting and maximum sprouting percentages of 36.12 during 2011 and 37.99 during 2012 were recorded in grafting performed on 15 day old rootstock. The higher sprouted grafts might be due to high meristematic activity having higher reserved food material in the cotyledons of germinating seedling. The results are in agreement with Suk-In (2006) who reported that the survival rate of epicotyl grafting in walnuts ranged from 65 to 87 per cent. Hartmann *et al.* (2002) reported that 63-75 per cent graft success was achieved on epicotyls using cleft method of grafting, generally less in other methods. Gandev (2009) also reported a good percentage of success in cleft grafting of walnuts on 10-15 day old seedling. Lowest percentages of sprouting (20.99 during 2011 and 18.21 during 2012) was recorded in grafting performed on 45 day old stocks. The reason related to the fact may be accumulation of phenolic compounds and fluctuations in temperature. Ronting and Pinghai (1990) noticed variations in Juglone content in three cultivars which lead to differences in graft sprouting percentage.

Scion sprouting was affected significantly with the portion of scion wood used in present investigation. Terminal portion of scion wood was found superior. The present results are in agreement with Anadoliev (1975) who used scions from upper, middle and lower parts of walnut shoots giving 94.59, 91.82 and 86.46 per cent success respectively.

Scion sprouting percentage was lower with the increase in height of grafting and the time of grafting. As the height of grafting increased there was corresponding increase in the scion sprouting failure percentage (Fig. 3). Time of grafting also influenced the graft failure percentage. The later the time of grafting the more the grafting failure was noticed (Fig. 3).



**Fig. 3 : Scion sprouting failure (%) as influenced by height and time of grafting during 2011 and 2012**

### 5.2.3 Seedling growth (cm) of walnut epicotyls

The present investigation indicate that scion growth was more influenced by time of grafting compared to grafting heights. Maximum scion growth of (16.22 and 16.20 cm) was observed under grafting performed on 15day old stocks. The highest scion growth of 18.59 and 17.06 cm was produced in grafting performed at point of attachment of cotyledons on 15 day old rootstocks using terminal portion of scion wood. Reasons behind this higher scion growth could be the availability and utilization of reserved food material responsible for maintaining congenial physiological conditions favourable for optimum growth and development. The results are in agreement with Ram and Bist (1982) who reported better growth rate of grafts in mango. Minimum scion growth was recorded in grafting performed at late stage either due to reduced rates of division of cambial cells or exposure of more scion surface area to atmosphere which causes higher rate of desiccation.

The present study indicates that maximum number of compound leaves (5.47and 5.20) were observed when grafting was performed on 15 days old stocks and lowest number of leaves (3.78 and 4.03) were observed when grafting was performed on 45 days old stocks. The difference in the number of leaves of grafts of different age groups might be due to variations in the meristematic activity related to cell division for development of leaf primordium.

In the present study leaflet size was influenced by both time and height of grafting and maximum leaflet sizes (38.76 and 40.23 cm<sup>2</sup>) were observed in graftings performed on 15 day old stocks. Leaf area depends upon the genotype and plant adaptations. Cell division and increase in leaflet area depends upon the relative increase in anticlinal divisions of meristematic cells. The present results are in agreement with Muradoglu and Gundogdu (2011) who reported that average leaflet area in walnut varied according to cultivar and leaflet size ranged from 26.77 to 86.92cm<sup>2</sup>.

In the present study maximum plant height (20.16 and 19.77 cm) was observed under grafting performed at the height of 6cm followed by 4cm grafting (18.88 and 18.47.15 cm) during 2011 and 2012, respectively. The reasons related could be rapid cell division, cell enlargement and shoot elongation.

Time of grafting had a significant effect on growth of grafts. Maximum plant height (19.24 and 19.14 cm) was recorded in grafting performed on 15 day old stocks. In younger rootstocks meristematic cells divide fast and food material stored in hypo cotyledons is better utilized. The present results are in agreement with Hartmann and Kester (1997) who reported that age of rootstock has relationship with regenerating ability of plant parts and higher activity of meristematic cells. The present study also indicates that the maximum stem diameter (3.67 and 3.68 mm) was recorded in grafting performed on 15 day old stocks. The higher stem diameter in younger stocks might be due to higher meristematic activity in periclinal cells (meristematic cells) which increase stem diameter owing to better water and solute absorption for new cell wall material formation.

## Chapter – 6

### SUMMARY AND CONCLUSION

Investigation entitled “Studies on Seed Germination and Epicotyl Grafting was conducted on Walnut” at Plant Nursery Division of Fruit Science SKUAST-Kashmir, Shalimar, Srinagar, J&K. The main objective was to determine the effect of various chemical treatments in breaking seed dormancy, promoting germination and growth of walnut seedling and to evaluate the Feasibility of using Epicotyl method of grafting as a Procedure for Vegetative Propagation of Walnut.

Chemical treatments each at two levels viz., GA<sub>3</sub> (50 and 100 ppm), sulphuric acid (7.5 and 9.5%), potassium nitrate (0.2 and 0.4%) and Ethrel (1000 and 1500 ppm) were used in present investigation. Storage treatments constitute main and chemical treatment constitute sub-plot treatments.

Height of grafting, time of grafting and portion of scion wood taken consists of main treatments on the basis of which feasibility of epicotyl grafting was evaluated under open conditions. Grafting heights viz., point of attachment of cotyledons, 2 cm above the point of attachment of cotyledons, 4 cm above the point of attachment of cotyledons and 6 cm above the point of attachment of cotyledons. Time of grafting viz., 15 days after germination of seedling, 30 days after germination of seedling, 45 days after germination of seedling. Scion wood viz., terminal and basal portion was taken.

The chemical treatments GA<sub>3</sub> 100 ppm followed by 50 ppm were most effective in breaking the seed dormancy and promoting germination and growth of seedling in comparison to seeds soaked in water for 12 hours. Epicotyl grafting survival rate was observed highest in grafting performed at the point of attachment of cotyledons on 15 day old seedling.

- Among the chemical treatments given to walnut seeds, GA<sub>3</sub> at 100 ppm resulted in highest seed germination percentage (58.24 and 58.57) followed by Gibberellic acid at 50 ppm (54.23 and 56.32).

- Amongst the storage treatments cold storage resulted in significantly higher germination percentage (46.51 and 46.56).
- Maximum germination (63.49 and 62.66%) was recorded with the treatment combination of GA<sub>3</sub> at 100 ppm and 30 days chilling. Significantly higher germination (47.73 and 47.51%) was recorded with treatment combination of un-soaked seed and cold storage.
- Interaction between chemical x seed treatment reveal that the seed treatment with GA<sub>3</sub> at 100 ppm under un-soaked conditions recorded significantly higher germination (59.33 and 59.66%).
- Higher percentage of germination was recorded in GA<sub>3</sub> 100 ppm under cold storage treatment with un-soaked seed (65.33 and 63.66%).
- GA<sub>3</sub> (100 ppm) recorded maximum height (23.24 and 23.00 cm) followed by Gibberellic acid at 50 ppm (20.24 and 20.31cm), H<sub>2</sub>SO<sub>4</sub> at (7.5%) recorded (19.62 and 19.55 cm), H<sub>2</sub>SO<sub>4</sub> (9.5%) (18.48 and 18.73 cm) and KNO<sub>3</sub> at 0.4% (18.01 and 17.94 cm).
- Amongst the chemical treatments minimum height (13.50 and 13.40 cm) was observed with the application of ethrel at 1000 ppm which was significantly higher when compared to control (12.78 and 12.90 cm).
- Among the storage treatments cold storage resulted in significantly higher stem length (21.28 and 17.93 cm) in comparison to the ambient storage (16.83 and 16.99 cm).
- Significantly higher stem length (17.78 and 17.79 cm) was also recorded under un-soaked seed treatment than the soaked seeds (16.93 and 16.96 cm).
- Interaction of chemical x storage recorded maximum height (24.17 and 24.27 cm) with the GA<sub>3</sub> 100 ppm and 30 days cold storage followed by GA<sub>3</sub> 100 ppm (22.31 and 21.73cm) under ambient storage.

- Interactions between seed treatment x storage had a significant effect on seedling height and maximum height (18.66 and 18.68 cm) was observed in un-soaked seed treatment under cold storage.
- Seed treatment and chemical interaction was found effective in promoting height and maximum height (23.83 and 23.61 cm) was observed in GA<sub>3</sub> 100 ppm with un-soaked seed treatment which was 22.16 and 22.38 cm under GA<sub>3</sub> 100 ppm in soaked seed treatment.
- The interaction between chemical x storage x seed treatment were also statistically significant and maximum stem length (25.35 and 24.91 cm) was observed in GA<sub>3</sub> 100 ppm under cold storage in un-soaked seed treatment.
- GA<sub>3</sub> at 100 ppm resulted in highest stem diameter (4.75 and 4.92 mm) followed by Gibberellic acid at 50 ppm (4.19 and 4.04 mm).
- Among the storage treatments cold storage resulted in higher stem diameter (4.09 and 3.93 mm) in comparison to the ambient storage (3.53 and 3.44 mm). Un-soaked seed treatment recorded higher stem diameter (4.01 and 3.80 mm) which was 3.83 and 3.57 mm in case of soaked seed treatment.
- Higher stem diameter was recorded in GA<sub>3</sub>100 ppm under cold storage treatment with un-soaked seed (5.71 and 5.63 mm). Among the interaction of chemical x seed treatment x storage the lowest stem diameter (2.59 and 2.14 mm) was observed in control at ambient storage under un-soaked seed treatment.
- Among the chemical treatments Gibberellic acid 100 ppm recorded maximum root length (191.57 and 194.91 mm) followed by GA<sub>3</sub> 50 ppm (181.24 and 184.16 mm).
- Cold stored seeds resulted in higher root length (148.36 and 156.69 mm) than the ambient stored seeds 134.82 and 141.55 mm. Significantly higher

root length (144.92 and 152.40 mm) was also recorded under un-soaked seed treatment than the soaked seeds (138.27 and 145.84mm).

- Maximum root length (198.49 and 200.00 mm) under interaction of chemical x storage was observed in GA<sub>3</sub> 100 ppm with 30 days chilling.
- Seed treatment x chemical interaction recorded significantly higher root length (196.16 and 199.33mm) with the application of GA<sub>3</sub> 100 ppm under un-soaked seed treatment.
- Higher root length was recorded in GA<sub>3</sub> 100 ppm under cold storage treatment with un-soaked seeds (201.66 and 203.33 mm). Among the interactions between chemical x seed treatment x storage the lowest root length (84.66 and 85.13) was recorded in control under ambient storage treatment with un-soaked seeds.
- All chemical and storage treatments were statistically significant as far as their influence on number of secondary roots of walnut seedlings is concerned. Among the chemical treatments Gibberellic acid 100 ppm recorded maximum (27.49 and 27.74) number of secondary roots followed by GA<sub>3</sub> 50 ppm (25.99 and 25.74) during 2011 and 2012 respectively.
- Among the storage treatments cold stored seeds resulted in higher (21.18 and 22.29) number of secondary roots than the ambient stored seeds (20.16 and 19.45). Significantly higher (21.02 and 21.23) number of secondary roots was also recorded under un-soaked seed treatments than the soaked seeds (20.32 and 20.51) during 2011 and 2012.
- Seed treatment x chemical interaction recorded significantly higher number of secondary roots (28.00 and 28.50) with the application of GA<sub>3</sub> 100 ppm and un-soaked seed treatment during the year 2011 and 2012.
- Higher number of secondary roots (29.33 and 29.43) was recorded in GA<sub>3</sub> 100 ppm under cold storage treatment with un-soaked seeds. Among the interactions between chemical x seed treatment x storage the lowest

number of secondary roots (14.33 and 14.16) was recorded in control under cold storage treatment with un-soaked seeds.

- Chemical treatments had a significant effect on number of leaves during 2011 and 2012. Maximum number of leaves (6.22 and 6.08) was obtained with the application of GA<sub>3</sub> 100 ppm.
- Maximum number of leaves (4.96 and 5.08) was observed under cold storage treatment which was comparatively lesser in case of ambient storage treatment 4.46 and 4.57. Seed treatment was also found non-significant, however, maximum number of leaves (4.85 and 4.89) was observed under un-soaked treatment in comparison to soaked seed treatment (4.57 and 4.76) both during 2011 and 2012.
- Highest number of leaves (6.08 and 6.53) was recorded with the application of GA<sub>3</sub> 100 ppm under cold storage with un-soaked seed treatment.
- Among the chemical treatments GA<sub>3</sub> 100 ppm recorded maximum leaflet size (74.12 and 72.70 cm<sup>2</sup>) followed by GA<sub>3</sub> 50 ppm (72.27 and 71.04 cm<sup>2</sup>).
- Among the storage treatments cold stored seeds resulted in significantly higher (65.62 and 62.77 cm<sup>2</sup>) leaflet size in comparison to ambient storage (61.30 and 61.18 cm<sup>2</sup>). Higher leaflet size (63.47 and 62.98 cm<sup>2</sup>) was also recorded under un-soaked seed treatments than the soaked seeds (62.57 and 60.97 cm<sup>2</sup>).
- Maximum (75.86 and 73.97 cm<sup>2</sup>) leaflet size at interaction of chemical x storage was observed in GA<sub>3</sub> 100 ppm with 30 day chilling.
- Under seed treatment x chemical interactions significantly higher leaflet size (75.02 and 73.74 cm<sup>2</sup>) was observed with the application of GA<sub>3</sub> 100 ppm with un-soaked seed treatment.

- Significantly higher leaflet size was recorded in GA<sub>3</sub> 100 ppm under cold storage treatment with un-soaked seeds (77.36 and 75.40 cm<sup>2</sup>) in comparison to all other treatment combinations.
- Chlorophyll content of walnut seedlings was found non-significant in all chemical and storage treatments. The chlorophyll content under different chemical treatments ranged from 0.80 to 0.86 mg/g of fresh weight, however maximum chlorophyll content (0.86 mg/g) was recorded in GA<sub>3</sub> 100 ppm during 2011 and 2012.
- Among the chemical treatments maximum number of days from germination to leaf fall (225.94 and 224.46) was recorded in GA<sub>3</sub> 100 ppm followed by GA<sub>3</sub> at 50 ppm (223.85 and 223.10).
- Among the storage treatments cold storage resulted in significantly higher number of days from germination to leaf fall (212.97 and 212.14) in comparison to the ambient storage (210.45 and 211.56).
- Interaction of chemical x storage recorded maximum number of days (226.39 and 225.44) with GA<sub>3</sub> 100 ppm with 30 days cold followed by GA<sub>3</sub> 100 ppm (225.49 and 222.26) under ambient storage.
- The interaction between chemical x storage x seed treatment was statistically significant and maximum number of days from germination to leaf fall (227.33 and 227.43) was observed in GA<sub>3</sub> 100 ppm under cold storage with un-soaked seed treatment.
- In epicotyl grafting maximum scion take percent (67.38 and 70.38) was recorded in grafting performed at the point of attachment of cotyledons followed by grafting at 2cm above the point of attachment of cotyledons (65.55 and 67.64).
- Time of grafting had a significant effect on scion take percentage. Highest scion take (67.49 and 64.79%) was recorded in grafting performed on 15

day old stocks followed by (63.37 and 63.76 %) when grafting (63.37 and 63.76 %) performed on 30 day old stocks.

- Minimum scion take (45.36 and 55.73 %) was observed when grafting was performed on 45 day old stocks.
- Higher percentage of scion take (62.99 and 64.52) was recorded in terminal portion of scion wood as compared to basal portion which was to the tune of 59.01 and 59.42 per cent.
- Maximum scion take percentage (69.10 and 72.77) was observed under interaction of scion wood x height in grafting performed at the point of attachment of cotyledons using terminal portion of scion wood.
- Height x time interaction produced maximum scion take percentage (71.39 and 75.83) when grafting was performed at the height of 2cm on 15 day old stocks.
- Scion take per cent of 67.16 and 68.25 was obtained under interaction of scion wood x time (W x T) in grafting performed on 15 day old stocks using terminal portion of scion wood.
- Highest scion take percent (75.33 and 78.33) was observed at interaction under grafting was performed on 15 day old stocks at the point of attachment of cotyledons using terminal portion of scion wood.
- Maximum scion take percent (63.32 and 64.84) was recorded after 30 days in grafting performed at the point of attachment of cotyledons followed by grafting at 2cm above the point of attachment of cotyledons (57.90 and 58.60).
- Maximum scion take percent (43.16 and 45.32) was recorded after 45 days when grafting was performed at the point of attachment of cotyledons followed by grafting at 2cm above the point of attachment of cotyledons (39.32 and 39.54).

- Time of grafting had a significant effect on scion take percentage. Highest scion take (42.53 and 43.78%) was recorded in grafting performed on 15 day old stocks followed by (38.78 and 39.54%) in grafting performed on 30 day old stocks.
- Maximum scion sprouting percentage (40.93 and 41.32) was recorded in grafting performed at the point of attachment of cotyledons followed by grafting at 2cm above the point of attachment of cotyledons (35.88 and 33.99%).
- The time of grafting was effective in promoting scion sprouting percentage. Highest scion sprouting percentage (36.12 and 37.99) was observed under grafting conducted on 15 day old stocks followed by grafting on 30 days old stock (33.74 %).
- The scion sprouting percentage got significantly lowered in grafting performed on 45 day old stocks and was to the tune of 22.24 and 19.82 per cent.
- Significantly higher scion sprouting percent (32.18 and 31.85) was recorded in terminal portion of scion wood as compared to basal portion which was to the tune of (29.21 and 29.63 per cent).
- Maximum scion sprouting percentage (43.88 and 43.77) was recorded under interaction scion wood x height when grafting was performed at the point of attachment of cotyledons using terminal portion of scion wood scion.
- Height x time interaction was statistically significant. Maximum scion sprouting (47.49 and 49.99 %) was obtained on 15 day old stocks grafted at the point of attachment of cotyledons.
- Scion wood x time interaction was found effective which records significantly higher scion sprouting per cent (37.91 and 39.99) when

grafting was performed on 15 day old stock using terminal portion of scion wood.

- The interactions between height x time x scion wood indicate that significantly higher scion sprouting (51.66 and 53.33 %) under obtained grafting was performed at the point of attachment of cotyledons on 15 day old stocks using terminal portion of scion wood.
- Maximum scion growth (15.75 and 15.78 cm) was recorded grafting performed just at the point of attachment of cotyledons.
- Among grafting heights the minimum scion growth (15.15 and 15.23 cm) was obtained in grafting performed at the height of 6cm.
- Time of grafting had a significant effect on scion growth. Maximum scion growth (16.22 and 16.20 cm) was recorded in grafting performed on 15 day old stocks.
- The portion of scion wood used also affected scion growth and significantly higher scion growth (15.87 and 15.71 cm) was recorded in terminal portion than in basal portion of scion wood (15.42 and 15.41cm).
- The interaction between height of grafting x time of grafting x scion wood used reveal that highest scion growth (18.59 and 17.06 cm) was obtained in grafting performed at the point of attachment of cotyledons on 15 day old stocks using terminal portion of scion wood.
- Height of grafting, time of grafting and scion wood used had a significant effect on number of leaves in walnut epicotyls grafting during 2011 and 2012. Maximum number of leaves (4.88 and 5.27) was recorded when grafting was performed at the point of attachment of cotyledons.
- Lowest number of leaves (4.10 and 4.20) was recorded under grafting performed at a height of 6cm.

- Higher number of leaves (4.69 and 4.69) was recorded in terminal portion of scion wood as compared to basal portion which (4.46 and 4.63).
- Maximum number of leaves (5.33 and 5.33) was recorded under interaction of wood x height in grafting performed at the point of attachment of cotyledons using terminal portion of scion wood.
- The interactions between height of grafting x scion wood used x time of grafting were significant during 2011 and 2102. Highest number of leaves (6.23 and 6.33) were observed at interaction of height x time x scion wood when grafting was performed on 15 day old stocks at the point of attachment of cotyledons using terminal portion of scion wood.
- Height of grafting, scion wood used and time of grafting had a significant effect on leaflet size in walnut epicotyl grafting during the year 2011 and 2012. Maximum leaflet size (38.52 and 38.78 cm<sup>2</sup>) was recorded in grafting performed at the point of attachment of cotyledons.
- Among the grafting heights minimum scion leaflet size (36.29 and 37.13 cm<sup>2</sup>) was obtained in grafting performed at the height of 6cm.
- Significantly higher leaflet size (38.76 and 40.23 cm<sup>2</sup>) was recorded in grafting performed on 15 day old stocks.
- Maximum leaflet size (39.08 and 38.93 cm<sup>2</sup>) was recorded under interaction of wood x height when grafting was performed at the point of attachment of cotyledons using terminal portion of scion wood.
- The interaction between height of grafting x time of grafting x scion wood used were significant and highest leaflet size (40.33 and 41.59 cm<sup>2</sup>) was observed in grafting performed at the point of attachment of cotyledons on 15 day old stocks using terminal portion of scion wood.
- Height of grafting, scion wood used and time of grafting had a significant influence on plant height during 2011 and 2012. Maximum plant height

(20.16 and 19.77 cm) was observed in grafting performed at the height of 6cm.

- Among grafting heights lowest plant height (17.84 and 18.00 cm) was recorded in grafting performed at the point of attachment of cotyledons.
- Time of grafting had a significant effect on plant height. Highest plant height (19.24 and 19.14 cm) was recorded in grafting performed on 15 day old stocks.
- Significantly higher plant height (19.14 and 18.89 cm) was recorded in terminal portion of scion wood used which was 18.41 and 18.34 cm in case of basal portion of scion wood used.
- Maximum plant height (20.43 and 20.07 cm) was observed at interaction of scion wood x height under grafting performed at the height of 6cm using terminal portion of scion wood.
- The interactions between height of grafting x scion wood used x time of grafting were significant. Highest plant height (20.90 and 20.35 cm) was recorded in grafting performed at 6cm height on 15 day old stocks using terminal portion of scion wood.
- Height of grafting, time of grafting and scion wood had a significant effect on stem diameter during 2011 and 2012. Maximum stem diameter (3.73 and 3.63mm) was recorded under grafting performed at the height of 4cm followed by stem diameter (3.59 and 3.56 mm) in grafting performed at 2cm height.
- Time of grafting had a significant effect on stem diameter and higher stem diameter (3.67 and 3.68 mm) was recorded in grafting performed on 15 day old stocks.

- Under interaction of height x scion wood maximum stem diameter (3.95 and 3.90 mm) was recorded under grafting performed at the height of 4cm using terminal portion of scion wood.
- Highest stem diameter (4.13 and 4.21 mm) was recorded under interaction of height x time x scion wood under grafting performed at 4cm height on 15 day old stocks using terminal portion of scion wood.

## **CONCLUSION**

Use of chemicals and storage treatments can be employed to attain maximum percentage germination and subsequent seedling growth. The results obtained in the present study indicate that seed germination of walnut at late stage can be possible with the use of GA<sub>3</sub> at 100 ppm concentration without pre-soaking treatment. Chemical treatments can substitute chilling required for walnut seed germination. Epicotyl grafting under Kashmir valley conditions is a viable option for mass propagation of walnuts. Epicotyl grafting can be performed on 15 day old walnut rootstocks at height of point of attachment of cotyledons. The scion wood needs to be stored under moist conditions at 3-4°C to retain the optimum moisture level. Skill development of nurserymen with respect to performing of grafting technique as well as management of propagated material is of vital importance.

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## Appendix – I

### Weather data for the year 2011

Date	Maximum Temperature (°C)	Minimum Temperature (°C)	Rainfall (mm)	Relative Humidity (%)	
				RH1	RH2
1/3/2011	11	2	1.4	91	52
2/3/2011	6.5	2	0	91	71
3/3/2011	7.5	2	6.7	94	71
4/3/2011	8	1	12.3	94	52
5/3/2011	11	1.4	1.2	91	37
6/3/2011	14	-0.5	0	90	37
7/3/2011	12	1.4	0	87	45
8/3/2011	15	3	0	87	55
9/3/2011	16.5	0	0	80	39
10/3/2011	18	0.4	0	80	39
11/3/2011	17.5	0	0	58	31
12/3/2011	19	0.4	0	74	23
13/3/2011	19.6	1.4	0	77	33
14/3/2011	22.5	1.5	0	68	21
15/3/2011	22.5	2.5	0	73	25
16/3/2011	22	3.5	0	69	25
17/3/2011	21	5.2	0	76	30
18/3/2011	23	6	0	79	37
19/3/2011	9.6	8.8	2.8	88	85
20/3/2011	10.5	1	34.7	80	45
21/3/2011	14	2.5	0	82	50
22/3/2011	17.5	1.6	0	91	46
23/3/2011	20	3	0	91	30
24/3/2011	21.5	3.5	0	86	31
25/3/2011	22	3	0	84	33
26/3/2011	21	4	0	87	30
27/3/2011	21.5	6.5	0	80	35
28/3/2011	9.5	7.5	2.6	98	92

29/3/2011	14	5	13	95	55
30/3/2011	16	7	8.6	92	43
31/3/2011	18	3.5	0	92	46
1/4/2011	13.5	5	0	85	73
2/4/2011	13.5	4.6	4	87	51
3/4/2011	11.5	2	2.4	94	77
4/4/2011	8	3	7.6	89	86
5/4/2011	6	3	9.4	94	86
6/4/2011	9.5	4	0	92	87
7/4/2011	15	4.5	2.6	92	52
8/4/2011	17.5	1.5	0	87	40
9/4/2011	18	3.5	0	72	40
10/4/2011	14	9.5	0	54	67
11/4/2011	13	7	20	93	92
12/4/2011	15.5	7.8	9.6	78	62
13/4/2011	20.5	6.5	0	80	36
14/4/2011	23	4.5	0	77	36
15/4/2011	22	6	0	63	38
16/4/2011	18	10	0	83	52
17/4/2011	14	8.4	18.1	93	68
18/4/2011	12.5	6.5	18.8	88	74
19/4/2011	11.5	6.6	0	90	88
20/4/2011	16.6	3.5	0	95	62
21/4/2011	19	4	0	82	49
22/4/2011	23.5	4.6	0	68	42
23/4/2011	26.5	5.5	0	68	36
24/4/2011	27	5.4	0	67	33
25/4/2011	28	6	0	65	27
26/4/2011	29	7	0	64	28
27/4/2011	29	7	0	65	28
28/4/2011	25.5	8	0	66	59
29/4/2011	22	9.6	0.6	71	65
30/4/2011	20.3	8	0	83	51

1/5/2011	24.4	8	0	72	29
2/5/2011	27	8	0	73	35
3/5/2011	29.5	9	0	68	27
4/5/2011	28	8.5	0	71	35
5/5/2011	26	7.6	0	71	52
6/5/2011	18	11.5	3.6	85	90
7/5/2011	23.5	8	2	87	42
8/5/2011	25	7.6	0	89	45
9/5/2011	24.5	8.5	0	91	53
10/5/2011	24	12	1.6	82	44
11/5/2011	24.5	10	0	81	59
12/5/2011	23.5	7.6	1	80	59
13/5/2011	24.5	7.4	1.8	70	42
14/5/2011	27.5	8	0	75	31
15/5/2011	31.5	7.6	0	69	27
16/5/2011	31	9	0	81	27
17/5/2011	32	11	0	73	36
18/5/2011	32	13	0	64	27
19/5/2011	31.5	12.8	0	67	26
20/5/2011	30	13	0	66	32
21/5/2011	26	13	0	58	41
22/5/2011	27.5	8.6	0	67	28
23/5/2011	30.5	9	0	67	28
24/5/2011	29	11	0	63	30
25/5/2011	29	14.3	0	70	30
26/5/2011	23.5	11.2	0	68	49
27/5/2011	25.5	14.1	5.8	94	32
28/5/2011	30	12	0	77	28
29/5/2011	30	13.4	0	64	28
30/5/2011	31	13	4	69	32
31/5/2011	31.5	11.5	0	66	26
1/6/2011	18	12.8	0	63	64
2/6/2011	24.5	10.4	1	83	51

3/6/2011	27	9.5	0	64	38
4/6/2011	30	9	0	65	30
5/6/2011	31	11.7	0	73	42
6/6/2011	29	15	0	54	22
7/6/2011	33.5	10.5	0	59	23
8/6/2011	30	12.9	0	61	24
9/6/2011	31	13.4	0	69	37
10/6/2011	30.5	11.4	1.1	69	33
11/6/2011	31.5	15.5	0	74	39
12/6/2011	26.5	12.6	37.4	87	43
13/6/2011	30.5	15	0	75	45
14/6/2011	32.5	13.7	0	68	43
15/6/2011	32.5	13.5	0	61	54
16/6/2011	33.5	16	0	65	66
17/6/2011	31.5	16	0	72	60
18/6/2011	29.5	16.5	0	82	47
19/6/2011	29.5	14.5	0	77	50
20/6/2011	30.5	15.5	0	82	52
21/6/2011	33	14	0	64	37
22/6/2011	34.5	16.1	0	69	45
23/6/2011	33	16	3.4	75	39
24/6/2011	33.5	16.7	0	73	43
25/6/2011	35	18.6	0	76	39
26/6/2011	33.5	20.5	0	69	47
27/6/2011	33.6	18.7	0	70	44
28/6/2011	29	18.4	0	68	50
29/6/2011	30	17.4	0	68	57
30/6/2011	29.5	16.6	0	59	57
1/7/2011	29	17.5	0	75	76
2/7/2011	29	15.4	0	73	50
3/7/2011	27	15.5	0	71	81
4/7/2011	29.5	13.2	7.2	79	46
5/7/2011	32	14.4	0	72	36

6/7/2011	33.5	15.5	0	64	32
7/7/2011	26.5	17.8	0	69	75
8/7/2011	25.5	16.4	0	91	56
9/7/2011	26.5	16	5	89	48
10/7/2011	26	15.5	0	78	51
11/7/2011	30	14	0	78	40
12/7/2011	32.5	13.7	0	68	52
13/7/2011	34.5	16.5	0	62	41
14/7/2011	25.5	20	0	81	69
15/7/2011	29	17.5	6	91	48
16/7/2011	23	17	3	91	73
17/7/2011	29.5	13.4	0	86	44
18/7/2011	30	15	0	68	48
19/7/2011	30.5	18	0	80	45
20/7/2011	33	16.8	0	73	48
21/7/2011	31	17.7	0	68	53
22/7/2011	33	18	0	67	65
23/7/2011	34.5	18.6	0	69	41
24/7/2011	31	21	0	76	59
25/7/2011	31.5	21.4	0	76	59
26/7/2011	30.5	20.8	0	89	64
27/7/2011	30.5	19.5	1.8	84	50
28/7/2011	30.5	17.4	7.2	88	53
29/7/2011	29.5	17	0.8	83	48
30/7/2011	32	16.4	0	77	58
31/7/2011	32.5	17	0	74	48
1/8/2011	33	14	0	61	48
2/8/2011	33.5	14	0	78	44
3/8/2011	33	16.4	0	68	42
4/8/2011	31	19.1	0	73	52
5/8/2011	33.5	16.5	0	80	40
6/8/2011	34.5	18.5	0	65	39
7/8/2011	26.6	19	2.2	93	69

8/8/2011	31	17	0	83	53
9/8/2011	32.5	16.9	5	76	44
10/8/2011	32.6	18.6	0	70	40
11/8/2011	31.5	20.1	0	76	48
12/8/2011	20	18	9.8	91	87
13/8/2011	28	15.5	6	85	57
14/8/2011	27.5	15.4	0	80	64
15/8/2011	27	15	0	87	76
16/8/2011	27	15	0	74	60
17/8/2011	31	13.6	0	75	44
18/8/2011	30.5	13.5	0	82	40
19/8/2011	30.6	14.2	0	75	44
20/8/2011	31.5	15.6	0	80	44
21/8/2011	33.5	16	0	70	35
22/8/2011	34	19.5	0	72	35
23/8/2011	35	20.2	0	66	33
24/8/2011	34.5	21.5	0	72	84
25/8/2011	28.5	16.5	11.2	78	57
26/8/2011	32	17	0	87	49
27/8/2011	29	18.4	2.4	88	57
28/8/2011	23.5	17.4	8	96	64
29/8/2011	22.5	15.4	0.6	91	60
30/8/2011	29.5	17	0	91	52
31/8/2011	32.5	17	0	94	39

**Source :** Department of Agronomy, SKUAST-Kashmir, Srinagar, Jammu and Kashmir

## Appendix – II

### Weather data for the year 2012

Date	Maximum Temperature (°C)	Minimum Temperature (°C)	Rainfall (mm)	Relative Humidity (%)	
				RH1	RH2
01/03/2012	13	1	0	80	36
2/03/2012	14	-0.6	0	87	49
3/03/2012	17	1.4	0	79	29
4/03/2012	5	4	6	94	91
5/03/2012	10	2.5	23.4	97	72
6/03/2012	13.5	2.5	0	89	58
7/03/2012	14.5	1.8	0	86	43
8/03/2012	10	1.8	0	94	47
9/03/2012	10	-0.7	2.2	97	54
10/03/2012	12.5	1	0	85	41
11/03/2012	11.5	-1	0	84	43
12/03/2012	11.5	1.5	0	72	45
13/03/2012	13.5	2.3	0	88	40
14/03/2012	15.5	4.1	1	65	52
15/03/2012	16	0.5	0	82	45
16/03/2012	15	3	0	67	35
17/03/2012	16	4.4	0	81	45
18/03/2012	19	5	0	84	40
19/03/2012	20	6.2	0	64	30
20/03/2012	20.5	17	0	38	26
21/03/2012	17	2.5	0	69	31
22/03/2012	19.5	2	0	73	25
23/03/2012	22	3.5	0	73	21
24/03/2012	22	5.4	0	64	21
25/03/2012	17	7.5	0	73	53
26/03/2012	12.5	6.4	1.2	87	87
27/03/2012	13.5	6	4	87	68

28/03/2012	18.5	6.5	0.8	88	40
29/03/2012	21	3.5	0	87	38
30/03/2012	23	3.5	0	77	29
31/03/2012	21.5	4.5	0	87	40
1/4/2012	21	4	0	77	56
2/4/2012	22	3	0	74	51
3/4/2012	21.5	7.4	30.4	72	51
4/4/2012	23	6	0	77	40
5/4/2012	21.5	6.5	0	75	43
6/4/2012	21	3.8	0	77	35
7/4/2012	23	5.2	0	80	35
8/4/2012	24.5	6	0	74	30
9/4/2012	19.5	10.5	0	70	60
10/4/2012	11.5	9	13.4	91	88
11/4/2012	13	8.5	8	95	78
12/4/2012	17	7	20	88	54
13/4/2012	10	5.9	2.1	92	87
14/4/2012	16.5	6.5	3	87	53
15/4/2012	16.5	4.5	0	87	78
16/4/2012	18.5	4	0	90	49
17/4/2012	22	4.4	0	81	38
18/4/2012	23.5	4.5	0	73	30
19/4/2012	23.5	5.5	0	88	30
20/4/2012	21.5	10.5	0	79	43
21/4/2012	21.5	11	9.2	89	40
22/4/2012	19	8.5	1	83	68
23/4/2012	18.5	9.5	3	89	55
24/4/2012	20	9.5	9	85	49
25/4/2012	21	7.6	0	83	72
26/4/2012	17.5	10	5	89	56
27/4/2012	20	9	20	89	53
28/4/2012	17.5	7	0	84	70
29/4/2012	17	7.5	1.6	89	69

30/4/2012	17.5	8	4.8	89	66
1/5/2012	13.5	3.5	1.6	92	68
2/5/2012	19.5	5	0.2	83	49
3/5/2012	23	4.6	0	84	41
4/5/2012	25	5.5	0	69	38
5/5/2012	25	6.5	1.5	69	50
6/5/2012	24	11	0	68	52
7/5/2012	22	8.6	0	82	59
8/5/2012	27	8.4	0	92	59
9/5/2012	28.5	9.4	0	72	46
10/5/2012	25	13	0	80	67
11/5/2012	18.5	10	1.6	83	81
12/5/2012	19	10	6.6	85	72
13/5/2012	22	7.9	3.5	91	53
14/5/2012	23	8	0	80	59
15/5/2012	22	6.8	0	81	63
16/5/2012	25	9.5	0	76	56
17/5/2012	20.5	9.5	0	75	62
18/5/2012	19.5	11.2	10	79	66
19/5/2012	24	9	2	89	54
20/5/2012	22.5	8.5	0	79	51
21/5/2012	18.5	10.6	1.6	93	69
22/5/2012	17.5	10	9.2	89	80
23/5/2012	21	8	0	89	58
24/5/2012	26	7.6	0	76	44
25/5/2012	21.5	10	0	66	80
26/5/2012	21	12	3.2	90	62
27/5/2012	25.5	8.4	2.6	84	45
28/5/2012	27	9.5	0	93	38
29/5/2012	29	9	0	85	37
30/5/2012	31	10	0	69	39
31/5/2012	30	10	0	64	71
1/6/2012	30.5	9	3.4	63	70

2/6/2012	31	10	0	56	35
3/6/2012	31	10	0	64	43
4/6/2012	29.5	9.4	0	74	47
5/6/2012	27.5	10	0	73	54
6/6/2012	24.5	12	0	73	57
7/6/2012	20	11.5	0	86	90
8/6/2012	17	11	0	94	86
9/6/2012	23	10.4	8.4	89	47
10/6/2012	26	11.5	0	80	38
11/6/2012	25	11.5	0	77	50
12/6/2012	24.5	10.6	0.6	90	39
13/6/2012	25	13.8	0	77	53
14/6/2012	27	11.4	1	90	48
15/6/2012	29.5	11.4	0	83	45
16/6/2012	30	11	0	73	44
17/6/2012	29.5	11.5	0	71	45
18/6/2012	27.5	12	0	88	50
19/6/2012	29	15.5	0	81	37
20/6/2012	30.5	13	0	82	32
21/6/2012	33	13.5	0	79	40
22/6/2012	32.5	16.5	0	75	40
23/6/2012	30	18	0	52	42
24/6/2012	29.5	15	0	86	51
25/6/2012	24	16.4	0	84	62
26/6/2012	27	14.6	0.8	73	51
27/6/2012	26.5	12	10	70	51
28/6/2012	29.5	14	0	78	51
29/6/2012	29.5	14.5	0	77	39
30/6/2012	32.5	14	0	84	36
1/7/2012	31.5	13	0	87	43
2/7/2012	34	15	0	68	36
3/7/2012	35.5	16	0	67	36
4/7/2012	35.5	20	0	69	36

5/7/2012	28.5	18.7	1	91	50
6/7/2012	33.5	17	0	79	40
7/7/2012	31	20	0	76	53
8/7/2012	31.5	16	0	75	48
9/7/2012	31	16.5	0	70	48
10/7/2012	34.5	15.6	0	74	41
11/7/2012	28	18.5	0	76	64
12/7/2012	29	17.5	0	88	72
13/7/2012	21.5	16.4	0	89	91
14/7/2012	28	14.5	6.2	89	58
15/7/2012	26.5	14.5	12.4	91	64
16/7/2012	30.5	14.5	0	82	43
17/7/2012	32	16	0	75	43
18/7/2012	34	16	0	72	38
19/7/2012	28	16.5	2.2	68	67
20/7/2012	29	15.5	1.2	96	56
21/7/2012	29	15.5	0	96	48
22/7/2012	33.5	15.5	0	87	40
23/7/2012	34.5	17.4	0	91	41
24/7/2012	34.5	18.6	0	76	41
25/7/2012	29	18	7	79	48
26/7/2012	28.5	18.4	0	89	53
27/7/2012	32.5	15.2	2.2	68	40
28/7/2012	33.5	15.4	0	91	39
29/7/2012	34.5	17	0	86	41
30/7/2012	33	20	0	79	40
31/7/2012	34.5	18.5	0	84	45
1/8/2012	33	20	0	76	45
2/8/2012	34	21.5	0	83	45
3/8/2012	33.5	19.6	0	83	45
4/8/2012	21	18.5	2.6	91	91
5/8/2012	22.5	14.4	26.4	90	75
6/8/2012	29.5	14.6	0	87	48

7/8/2012	31	15	0	84	48
8/8/2012	33.5	15.2	0	75	44
9/8/2012	31	17	0	76	48
10/8/2012	28.5	18	0	80	55
11/8/2012	23.5	17.5	0	91	84
12/8/2012	28.5	18	4.8	91	64
13/8/2012	29	16	0	86	64
14/8/2012	20	18.6	1.6	91	54
15/8/2012	28.5	19	21	83	55
16/8/2012	29	19	1	91	64
17/8/2012	32	17.5	0	84	54
18/8/2012	33.5	18	0	83	49
19/8/2012	33	20.5	0	80	49
20/8/2012	34	20	0	76	46
21/8/2012	34.5	21.5	0	79	45
22/8/2012	33	19.5	1.8	85	45
23/8/2012	32.5	19.3	0	84	54
24/8/2012	33.5	18.7	0	76	45
25/8/2012	30.5	19.1	0	83	53
26/8/2012	26.5	17	0	93	45
27/8/2012	30	15.5	0	88	57
28/8/2012	30	17	1	75	58
29/8/2012	32	15	0	75	51
30/8/2012	30.5	16	0	67	36
31/8/2012	33	14.6	0	71	49

**Source :** Department of Agronomy, SKUAST-Kashmir, Srinagar, Jammu and Kashmir

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## **CERTIFICATE**

Certified that all the corrections/amendments as suggested by External Examiner Prof. D.B. Singh, Principal Scientist, Central Institute of Temperate Horticulture (CITH), Srinagar during Viva-Voce examination held on 28<sup>th</sup> of March, 2014 have been incorporated in the manuscript entitled “**Studies on Seed Germination and Epicotyl Grafting in Walnut (*Juglans regia* L.)**” submitted by **Mr. Suja Nabi Qureshi (Regd. No. 2010-310-D)**.

*(Prof. M.S. Wani)*  
*Chairman*  
*Advisory Committee*