



Effect of different environments, grafting methods and times on sprouting, graft success and plant growth of walnut (*Juglans regia*)

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ABSTRACT

To find out the ideal factors which affect the graft success of walnut, an experiment was carried out at experimental farm of Central Institute of Temperate Horticulture during 2008-2009 with three environmental conditions (open field, polytrench and polyhouse), two grafting methods (wedge grafting and tongue grafting) and four grafting times (15 February, 1 March, 15 March and 1 April). Polyhouse recorded highest percentage of sprouting (65.51%), graft success (54.46%) and plant growth (168.75 cm). Wedge grafting was found superior to tongue grafting in respect of sprouting percent (62.22%), graft success (52.45%) and plant height (151.5 cm). Amongst the grafting times 15 March recorded the highest graft success (58.00%) and plant growth (163.78 cm). The highest sprouting (80.00%) and graft success (64.62%) was found under polyhouse with wedge grafting performed on 15 of March under Kashmir conditions can be adopted for mass multiplication.

Key words: Environmental conditions, Grafting methods, Grafting time, *Juglans regia*

Walnut (*Juglans regia* L.) is one of the important temperate nut fruit praised for its high proteins, fibre, vitamin B, minerals and anti-oxidants such as vitamin E and omega-3 fatty acids which help in lowering the cholesterol in human body. It has high demand both in national and international market. Nation earns more than ₹ 300 crores of foreign exchange annually from 44 countries through walnut export. In India it is grown in area of 30 800 ha with 36 000 tonnes of production and 1.7 tonnes/ha of productivity. (FAO 2011). It is grown mainly in Jammu and Kashmir, Himachal Pradesh, Uttarakhand and Arunachal Pradesh. Jammu and Kashmir is major walnut producing state contributing more than 85% of total production of the country. Presently, most of the orchards are of seedling origin leading to large variability in size, shape, colour and quality. It adversely affects the market value. Besides variability, long juvenile period and low productivity is also of great concern for walnut industry in our country. The high-yielding varieties with superior quality and good nutritive value have to be propagated to make our walnut industry competitive in the international market. The clonal propagation of walnut is a difficult process due to low

rate of callus formation (Kruniyuki and Fordi 1985, Coggeshall and Beineke 1997). This is due to presence of high concentration of phenolic compounds in its tissues and their oxidation by wounding (Rongting and Pinghai 1993).

The different degree of success in walnut propagation has been achieved by various propagation techniques throughout world (Gandev 2007). The factors like, temperature, humidity, graft type and time of grafting, affect graft success (Gandev 2007). Walnut do not produce any callus below 20 °C (Erdogan 2006) and optimum temperature for maximum graft success is 26–27 °C (Lagerstedt 1979, Millikean 1984). Success of any walnut grafting method depends on the environmental factor conditions of respective regions. Thus standardization of optimum environment, grafting method and time is of most importance for maximum graft success of walnut for a particular region. Keeping in view the above facts an attempt has been made to find out the ideal environment, grafting method and grafting time for optimum graft success under Kashmir conditions.

MATERIALS AND METHODS

The experiment was conducted during 2008 and 2009 at experimental farm of Central Institute of Temperate Horticulture, Rangreth, Srinagar (J&K) in three factor factorial randomized block design with three replications. There were three environmental conditions (open field, polytrench and polyhouse), two grafting methods (wedge grafting and tongue

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grafting) and four grafting times (15 February, 1 March, 15 March and 1 April). Two year old seedling rootstock of *J. regia* with uniform growth and thickness (1.5–2.0 cm in diameter at 15 cm above the ground level) were selected for grafting. 400 gauge alkathine strips were used for tying the graft union. One year old dormant scions with four to five female buds were used for grafting. The male flower bud was removed at the time of grafting to avoid the loss of nutrient reserve from the scion wood. 50 plants were used for each treatment. The experimental area was provided uniform cultural operations. The temperature under polyhouse and polytrench was maintained at $25 \pm 2^{\circ}$ C with intermittent misting and using 50% shading nets during extreme temperature, which helps to maintain more than 80 % relative humidity. The sprouting percentage was recorded 45 days after grafting, whereas graft success percentage, plant height and number of leaves/plant were recorded when the plants started recessing their growth at the end of growing season. The pooled data of two years was analyzed as method suggested by Gomez and Gomez (1984) using R software.

RESULTS AND DISCUSSION

Environmental conditions influenced the sprouting, graft success and plant growth significantly. Among the environmental conditions polyhouse recorded the highest sprouting (65.51 %), graft success (54.46 %), plant height (168.75 cm) and number of leaves/plant (120.06) closely followed by poly trench with the sprouting per cent (61.94 %), graft success (52.1 7%), plant height (154.57 cm) and 119.19 leaves/plant respectively (Fig 1). It may be due to congenial temperature and humidity under polyhouse which help in proliferations of callus between rootstock and scion. Callus formation in walnut is temperature specific. (Hartmann *et al.* 1997). Ideal temperature for callus formation is 26 °C (Erdogan 2006) and no callus formation takes place for the

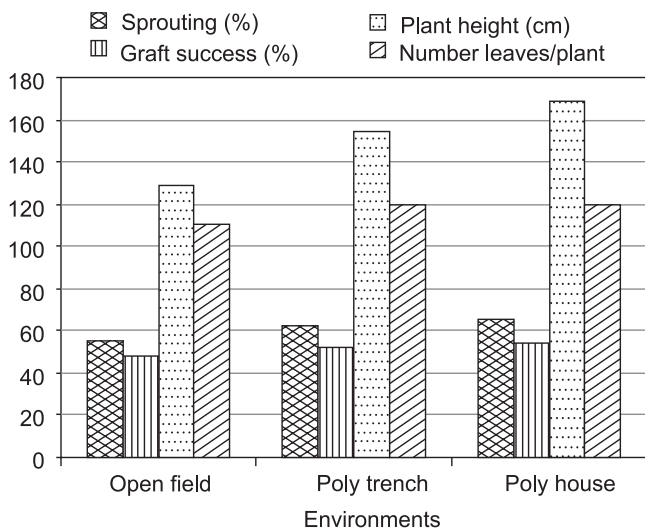


Fig 1 Effect of environment on sprouting, graft success, plant height and number of leaves

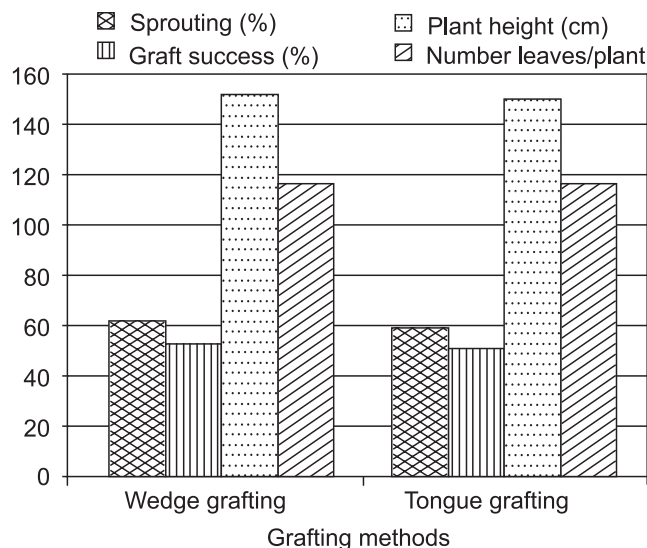


Fig 2 Effect of grafting methods on sprouting, graft success, plant height and number of leaves

healing below the 20 °C (Wilbur *et al.* 1998).

Grafting methods affected the graft sprouting, graft success and plant growth significantly. Wedge grafting recorded the maximum sprouting (62.22 %), graft success (52.45 %), plant height (151.54 cm) and number of leaves 116.73/plant, respectively (Fig 2). This may be due to better union of the space between scion and rootstock in wedge grafting as compared to tongue grafting. These results are in conformity with the findings of Singh *et al.* (2008)

Grafting time also affected the sprouting per cent, graft success and plant growth significantly. 15 March recorded the maximum sprouting (71.11 %), graft success (58.00 %), plant height (163.78 cm) and number of leaves/plant (133.24) (Fig 3) closely followed by 15 February irrespective of the other factors. This may be due to congenial temperature

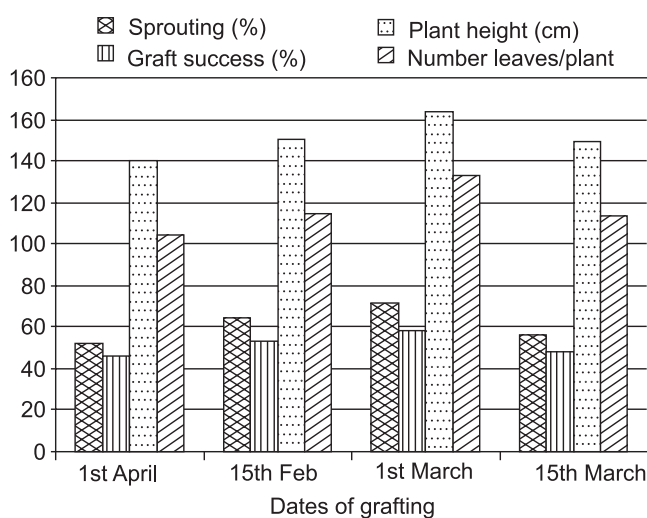


Fig 3 Effect of grafting times on sprouting, graft success, plant height and number of leaves

($25 \pm 2^{\circ}\text{C}$) which occurs from 2 fortnight of March in polyhouse and sometimes in open under Kashmir valley conditions leading to production of hormones and mobilization of energy producing nutrients after the activation of dormant of scion and rootstock cause a rapid regeneration of the cambium tissues of scion and rootstock and their intermingling. The complete graft union leads to good uptake of plant nutrients, resulting the better plant growth. These results are corroborative with the findings of Porebsiki *et al.* (2002) and Erdogan 2006.

The interaction of environment and grafting methods were also found to have significant effect on the sprouting per cent, grafting success and plant growth. The highest sprouting (69.72 %), graft success (57.16 %) was found in wedge grafting under polyhouse however, the highest plant height (169.05 cm) was observed in wedge grafting under open field and maximum number of leaves (124.139) were found in tongue grafting under open field (Table 1).

Wedge grafting performed on 15 March recorded the maximum sprouting (71.85%), graft success (58.76%), plant height (163.89 cm) and number of leaves/plant (133.67) which was statistically at par with interaction effect of tongue grafting performed on 15 March (Table 2). This may be due to the rapid regeneration of cambium tissue due to activation of scion and rootstocks coupled with ideal temperature in mid March. These findings are in conformity with findings of Porebsiki *et al.* (2002).

Interaction of environment and grafting time influenced the graft sprouting, graft success and plant growth, significantly. Highest sprouting (76.48 %), graft success (61.80%) was recorded with 15 March grafting under polyhouse condition (Table 3) however, maximum plant height (188.94 cm) and maximum number of leaves (144.33)

Table 1 Interaction effect of environmental conditions and grafting methods on sprouting (%), graft success (%), plant height (cm) and number of leaves/plant

Interaction effect of environment and grafting methods	Sprouting (%)	Graft success (%)	Plant height (cm)	Number leaves/plant
Poly trench \times Wedge grafting	63.145	52.988	128.417	120.889
Poly trench \times Tongue grafting	60.737	51.356	129.972	119.222
Open field \times Wedge grafting	53.794	47.211	169.056	114.250
Open field \times Tongue grafting	55.923	48.503	168.444	124.139
Polyhouse \times Wedge grafting	69.719	57.161	157.139	115.056
Polyhouse \times Tongue grafting	61.293	51.765	152.000	105.194
CD ($P=0.05$)	2.85	1.73	2.91	2.91

Table 2 Interaction effects of grafting methods and grafting time on sprouting (%), graft success (%), plant height (cm) and number of leaves/plant

Interaction effect of grafting methods and grafting time	Sprouting (%)	Graft success (%)	Plant height (cm)	No. of leaves/plant
Wedge grafting \times 15 Feb	53.82	47.22	142.33	107.26
Wedge grafting \times 1 March	65.18	54.07	150.70	115.74
Wedge grafting \times 15 March	71.85	58.76	163.89	133.67
Wedge grafting \times 1 April	58.02	49.77	149.22	110.26
Tongue grafting \times 15 Feb	49.87	44.94	137.70	101.89
Tongue grafting \times 1 March	62.96	52.61	149.30	112.52
Tongue grafting \times 15 March	70.37	57.24	163.67	132.82
Tongue grafting \times 1 April	54.07	47.38	149.89	117.52
CD ($P = 0.05$)	3.29	1.99	14.50	9.36

was found under open field grafted on 15 March and polytrench grafted on 15 March, respectively (Table 3). The higher success may be due to combined effect of congenial temperature, humidity and active state of scion and stock tissues, during this period which permits maximum regeneration of parenchyma cells in cambium region. Optimum humidity in polyhouse maintains high degree of hydration levels resulting in maximum graft success. These findings are in conformity with the findings of Ebrahimi *et al.* (2006), who obtained better success under polyhouse conditions in walnut.

The interaction effect of environment, grafting method and grafting time on sprouting per cent, graft success and plant growth was highly significant. The highest sprouting per cent (80.00 %), graft success (64.62 %) was obtained under polyhouse conditions with wedge grafting performed

Table 3 Interaction effects of environmental conditions and grafting time on sprouting (%), graft success (%), plant height (cm) and number of leaves/plant

Interaction effect of environment and grafting time	Sprouting (%)	Graft success (%)	Plant height (cm)	No. of leaves/plant
Poly trench \times 15 Feb	52.22	46.31	119.06	109.00
Poly trench \times 1 March	67.22	55.24	128.00	117.50
Poly trench \times 15 March	72.59	58.79	146.00	144.33
Poly trench \times 1 April	55.74	48.35	123.72	109.39
Open field \times 15 Feb	48.33	44.03	152.78	102.39
Open field \times 1 March	57.22	49.20	166.94	116.89
Open field \times 15 March	64.26	53.40	188.94	138.50
Open field \times 1 April	49.63	44.79	166.33	119.00
Polyhouse \times 15 Feb	55.00	47.91	148.22	102.33
Polyhouse \times 1 March	67.77	55.57	155.06	108.00
Polyhouse \times 15 March	76.48	61.80	156.39	116.89
Polyhouse \times 1 April	62.77	52.57	158.61	113.28
CD ($P=0.05$)	4.03	2.44	5.52	5.52

Table 4 Interaction effects of environmental conditions and grafting method and grafting time on sprouting (%), graft success (%), plant height (cm) and number of leaves/plant

Interaction effect of environment, grafting method and grafting time	Sprouting (%)	Graft success (%)	Plant height (cm)	Number of leaves/plant
Poly trench × wedge grafting × 15 Feb	54.81	47.80	121.22	117.00
Poly trench × wedge grafting × 1 March	70.00	57.00	127.33	118.67
Poly trench × wedge grafting × 15 March	73.70	59.75	140.67	143.44
Poly trench × wedge grafting × 1 April	54.07	47.39	124.44	104.44
Poly trench × tongue grafting × 15 Feb	49.63	44.82	116.89	101.00
Poly trench × tongue grafting × 1 March	64.44	53.47	128.67	116.33
Poly trench × tongue grafting × 15 March	71.48	57.82	151.33	145.22
Poly trench × tongue grafting × 1 April	57.40	49.31	123.00	114.33
Open field × wedge grafting × 15 Feb	47.40	43.49	155.22	97.33
Open field × wedge grafting × 1 March	55.92	48.45	169.33	117.22
Open field × wedge grafting × 15 March	61.85	51.90	189.78	140.56
Open field × wedge grafting × 1 April	50.00	45.01	161.89	101.89
Open field × tongue grafting × 15 Feb	49.26	44.57	150.33	107.44
Open field × tongue grafting × 1 March	58.52	49.96	164.56	116.56
Open field × tongue grafting × 15 March	66.66	54.91	188.11	136.44
Open field × tongue grafting × 1 April	49.26	44.57	170.78	136.11
Poly house × wedge grafting × 15 Feb	59.26	50.38	150.56	107.44
Poly house × wedge grafting × 1 March	69.63	56.75	155.44	111.33
Poly house × wedge grafting × 15 March	80.00	64.62	161.22	117.00
Poly house × wedge grafting × 1 April	70.00	56.89	161.33	124.44
Poly house × tongue grafting × 15 Feb	50.74	45.43	145.89	97.22
Poly house × tongue grafting × 1 March	65.92	54.40	154.67	104.67
Poly house × tongue grafting × 15 March	72.96	58.98	151.56	116.78
Poly house × tongue grafting × 1 April	55.55	48.25	155.89	102.11
CD ($P=0.05$)	5.71	3.45	7.82	7.82

on 15 March, which was statistically at par with polytrench conditions with wedge grafting executed on 15 March, closely followed by polyhouse conditions with tongue grafting executed on 15 March and polytrench conditions with tongue grafting performed on 15 March (Table 4). This may be attributed to the firm placement of scion and rootstock component with maximum union surface of cambium region under wedge grafting. Active cell regeneration stage in mid-March, conducive temperature and humidity under polyhouse conditions which helps in maximum parenchyma cell regeneration in cambial layer, resulting better union in scion and rootstock and higher growth (Bayazit *et al.* 2005). These results are in conformity with findings of (Ozakean and Giimmiis 2001).

REFERENCES

- Bayazit S, Imrak B and Kiden A. 2005. Determination of grafting time and methods of walnut under Adana ecological conditions. *Bance Ceviz* **34** (1): 231–4.
- Coggeshall M V and Beineke W F. 1997. Black walnut vegetative propagation; the challenge continues (*in*) *Knowledge for future of walnut*. J.W.V.Sambeek (Ed.), *Proceedings of 5th Black Walnut Symposium*. Gen Tech. rep. NC-191. St. Paul, MN: U.S. Dept. of Agriculture, Forest Services, North Cental Forest, Experiment Station, pp 70–7.
- Erdogan V. 2006. Effect of hot callusing cable on graft success in walnut (*Juglans regia*) propagation: nursery results. *Indian Journal of Agricultural Sciences* **76**(9): 544–6.
- Ebrahimi A, Vahdali K and Fallahi E. 2006. Walnut grafting under environmentally controlled conditions. *International Journal of Fruit Science* **6** (4): 8–12.
- FAO. 2011. FAO statistical databases and data sets 2011. *Cited from* <http://faostat.fao.org/site/13339/default.aspx>.
- Gandev S. 2007. Propagation of walnut under controlled temperature by the method of omega bench grafting, hot callus and epicotyl grafting. *Propagation of Ornamental Plants* **41**: 105–11.
- Gomez K A and Gomez A. 1984. *Statistical Procedure for Agricultural Research*, edn 2. John Wiley and Sons, Inc., New York.
- Hartmann H P, Kester D E, Daviees F T and Geneve R L . 1997. Theoretical aspects of grafting and budding. (*in*) *Plant Propagation-Principals and Practices*, edn 6. Prentic Hall of India Pvt, Ltd, New Delhi.
- Kruniyuki A and Fordi H. 1985. Walnut propagation. (*in*) *Walnut Orchard Management*, pp 38-46. D. Ronaas (Ed), University of California, USA.
- Lagerstedt H B. 1979. Propagation seed, grafting, budding. (*in*)

- Nut Trees Culture in North America*, pp 240-71. Jaynes R A (Ed). NNGA Broken Arrow Road Hamdan Conn.
- Millikean D F. 1984. Propagation of *Juglans* species by fall grafting. *Annual Report Nut Grower Association* **61**: 41-4.
- Ozakean Y and Giimmiis A. 2001. Effect of different application of grafting under controlled conditions of walnut (*Juglans regia* L.). *Acta Horticulturae* **412**: 187-99.
- Porebsiki S, Rzeznicka B and Poniedzialek W. 2002. Comparison of two methods of walnut grafting. *Journal of Fruit and Ornamental Plant Research* **10**: 55-62.
- Rongting X I and Pinghai D. 1993. A study on the uniting process of walnut grafting and factors affecting. *Acta Horticulturae* **311**: 160-70.
- Singh S R, Srivastava K K and Sharma M K. 2008. Hot cable callusing and zero energy polyhouse propagation techniques of walnut. (in) *Advances in Temperate Fruit Production*, pp 228-31. Banday F A and Sharma M K (Eds). Kalyani Publishers, Ludhiana.
- Wilbur O R, Leslie C A, Forde H I and Mc Kenna J R. 1998. Propagation. (in) *Walnut Production Manual*. Ramos D E (Ed). Division of Agriculture and Natural Resources, University of California.