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INFLUENCE OF SCION SELECTION ON GRAFTING SUCCESS AND GROWTH TRAITS IN *JUGLANS REGIA* L.: A COMPARATIVE ANALYSIS

Gazala Mansoor[1*], G. I. Hassan[1], Mahreena Farooq Rather[2] and Arifa Gulzar[2]

[1]Division of Fruit Science, Faculty of Horticulture Skuast-Kashmir Shalimar, Srinagar, India.
[2]Division of Plant Pathology, Faculty of Horticulture Skuast-Kashmir Shalimar, Srinagar, India.
*Corresponding author: gxalmanzoor707@gmail.com
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ABSTRACT

In this study, two-year-old rootstocks were wedge-grafted with various scion selections obtained from diverse locations within the Kashmir region. Grafting was performed during the first week of March under controlled polyhouse conditions. The results indicated that the earliest bud break occurred in selection WIW-33, averaging 34.7 days, whereas selection WIA-53 exhibited a delayed bud break at 43.4 days. Additionally, WIA-53 demonstrated a prolonged duration of bud break at 41 days, while bud split in the same selection was relatively brief, lasting only 43.7 days. Regarding leaf area, selection WHA-213 exhibited the largest leaf area (334.6 cm²), while WMK-16 had the smallest leaf area (277.6 cm²). The total number of leaflets per leaf was highest in WHA-213, with an average of nine leaflets. In terms of leaf nutrient content, WHA-213 also recorded the highest total chlorophyll concentration at 0.38 mg/g. Selection WIS-100 had the highest leaf nitrogen percentage at 3.19 mg/g, while WHA-213 recorded the highest levels of leaf phosphorus. Furthermore, WIS-100 exhibited the highest leaf potassium content at 1.64 mg/g. The graft success percentage was maximized in selection WIS-100, reaching 55.9%. Graft survival percentage was also highest in this selection at 90.4%, with complete graft healing quality recorded at 84.5%. The highest proportion of saleable plants was found in WHA-213 at 86.5%. Incremental shoot length was maximized in WHA-213, measuring 107.8 cm, whereas the maximum incremental shoot diameter was recorded in WIA-53 at 22.7 mm. The number of compound leaves observed at monthly intervals was highest in WHA-213, averaging 11.8 leaves. The findings of this investigation suggest that the growth parameters of walnut plants can be significantly influenced by the selection of different scions, as evidenced by variations in graft success, graft survival, graft healing, and subsequent shoot growth and diameter.

***Key words*: Walnut, selection, graft success, graft healing, graft survival.**

Introduction

Walnut (*Juglans regia*) commonly known as English or Persian walnut or "Akhrot" in Kashmir is an important deciduous tree (Mohni *et al.,* 2009). It attains height upto 25-30 m and have trunk diameter of 2 m to 4 m. The genus *Juglans* is characterized by its monoecious and heterodichogamous character. It accounts for around 21 species. *Juglansregia* L. is recognized as economically essential nut and oil species. Walnut is cultivated mostly in regions with temperate climate. It is naturally distributed from northern Pakistan, eastern Turkey, Iran, Afghanistan, mountain ranges of central Asia and Nepal. It is widely cultivated for its edible nut and for excellent wood quality. Walnut belongs to the family *Juglandaceae* with the chromosome number of 2n=2x=32.

In India, walnuts are found in Jammu and Kashmir, Himachal Pradesh, Arunachal Pradesh and Uttrakhand. Jammu and Kashmir is the chief producer of walnut in India with contribution of around 92% of the produce.

Walnuts grown in India are mainly indigenous in origin. Most of the production comes from seedling plants and no standard variety is generally grown. SKUAST-K has released two walnut cultivars (Hamdan and Sulaiman) having better nut and kernel attributes and shelling percentage. Other important selections (CITH 1-10) have been released by Central Institute of Temperate Horticulture (CITH), Srinagar, Kashmir. However, their finger prints have not been developed to protect their identity and restrict their misuse (package of practices in temperate fruit crops).

Since, the majority of the evidence currently available on the adoption of improved crop varieties comes from data on seed sales or from socio-economic household surveys and research impact assessment studies that rely on farmer self-reported data, there are a number of factors that make such methods prone to error. The limited number of morphological traits that actually distinguish closely related varieties, as demonstrated by Kosmowski *et al.,* (2016), can also have an impact on morphologically based variety assessment. These factors include environmental factors (which can affect how different morphological details are expressed), the stage of plant development at the time the variety is assessed, and the plant's developmental stage.

Due to the high concentration of phenolic chemicals in its tissues and their oxidation by wounding, walnut constantly exhibits challenges in its vegetative proliferation (Rongting and Pinghai, 1993). Under normal circumstances, walnut generally does not react favourably to vegetative propagation strategies the way other temperate fruits do. Various vegetative propagation techniques for walnut have reportedly yielded differing degrees of success in India and other countries with various climate conditions. According to Awasthi *et al.,* (1982) and Qureshi and Dalal (1985), the variances are caused by the various environmental conditions that the plants are exposed to both before and after grafting and budding. The rate of success tends to vary from place to place; depending on the agroclimatic conditions, variety, health of the mother plant, tree location, tree age and management of the mother plant. Some varieties like, Franquette and Hartley have been reported as most responsive for propagation than others.

Materials and Methods

An experiment was carried at Experimental farm of the Division of Fruit Science SKUAST-K, Shalimar, Srinagar in 2022 under polyhouse conditions. The experiment was laid out in a Completely Randomized Design (CRD) having three replications and 15 treatments in each replication. Seven walnut selections namely, WMK-2, WWI-16, WIW-33, WIA-53, WAI-54, WIS-100, WHA-213 are wedge grafted on rootstock *Juglans regia* L. The scion woods were collected from different places of Kashmir. The rootstock and the scion selection were used of the same size. The rootstocks were Wedge grafted with the different scion selections under polyhouse conditions. The cultural practices are same for the experiment.

Prameters: Data was recorded on the following parameters.

- **a) Days taken to bud break:** From the date of grafting till the date of first sprouting, the number of days for each treatment was recorded**.**
- **b) Duration of bud break:** The duration of bud-break was recorded from the first bud on scion that broke into growth to last bud on the scion that broke into growth.
- **c) Graft healing quality:** It takes an average of 5-8 days after grafting for the rootstock and scion to establish vascular connection and 14 days for the graft union to heal.
- **d) Incremental shoot length(cm):** Each previously-tagged grafted plant has its incremental shoot length measured with a scale on a monthly basis (from April to September) to determine the mean length of sprouts.
- **e) Incremental shoot diameter (mm):** Incremental shoot diameter was recorded with the help of digital vernier calliper at 30 days interval (April to September),it was recorded 5 cm above the stock scion union and the mean was calculated.
- **f) Number of compound leaves at monthly intervals:** The total number of compound leaves were counted on newly emerged shoots of graft at 30 days interval and average number of compound leaves were calculated.
- **g) Total number of leaflets / leaves:** In the last week of July, when the leaflets were fully developed, a count of the number of leaflets in 10 leaves was carried out and an average was calculated.
- **h) Leaf area (cm^2):** The leaf area was recorded from fully matured 10 leaves in the month of July.A leaf area meter was used to calculate the average leaf area, and was measured in cm^2.
- **i) Total leaf chlorophyll content (mg/g fresh wt.):** To evaluate the content of chlorophyll, 0.5 g of 10 fresh leaves was homogenized in 10 ml of acetone 80%. The extracts were passed through Whatman No. 1 filter paper. Then, 8000 µl of water was added to 200 µl of the filtered extract and its absorption was measured at 663

and 645 nm by spectrophotometry (Agilent-carry 300). The concentration of chlorophyll (mg/g Fw) in the sample was calculated using the following formula (Arnon *et al.,* 1967).

$$\text{Total Chlorophyll} = \frac{[20.7(A_{645}) + 8.02(A_{663})] \times \text{vol. of extraction mg g}^{-1}}{\text{Weight of the sample}}$$

V= Volume of the sample (ml)

A= Light absorption at 663 and 645 nm

FW= Sample fresh weight

Leaf nutrient content

- **Method of leaf sampling:** In the month of July, on July 15th, plant leaf samples were collected. On sunny days, leaf sampling was carried out between 8 and 10 a.m., selecting middle leaflets as proposed by Abaev and Khvorova (1983).
- **Preparation of leaf samples:** After sample collection, the fresh leaves were washed with 0.2% liquid detergent, then washed with N/10 HCL and deionized water to remove metallic contamination and wash previous solution, respectively. Once the excess moisture was removed, the samples were placed in clean paper bags and dried in a hot air oven at 60 to 70 degrees Celsius until their weight was uniform. For the purpose of to get ready the dried leaf for further investigation, it was crushed into a powder using an electric grinder.
- **Digestion of the leaf samples:** By adding a digestion mixture which includes potassium, copper, and ferrous sulphates, the sample was digested to estimate the nitrogen content in concentrated sulfuric acid. For determining the potassium and phosphorus levels in leaves. In accordance with Bhargava and Raghupathi (1998), digestion was carried out in a di-acid mixture containing nitric acid and perchloric acid at a 9:4 ratio.

Nutrient estimation

Nitrogen: Total nitrogen content was determined by micro Kejldahl method (A.O.A.C 1980) using semi-automatic pelican equipment classic- DXVATS (E) and results were expressed in percentage on dry weight basis.

Phosphorus: Using the venedo molybdo phosphoric acid yellow color approach (Jackson, 1973), the total amount of phosphorus was determined. The results were presented as a percentage on a dry weight basis.

Potassium: By using the flame photometer approach, total potassium content has been determined (Jackson, 1973). Results were represented as a percentage of dry weight.

- **j) Graft success (%):** The percentage of sprouted graft after grafting was observed in each treatment in Juneusing the following formula to calculate:

$$\text{Graft success (\%)} = \frac{\text{Sprouted grafts}}{\text{Total grafted plant}} \times 100$$

- **k) Graft survival (%):** The survival of graft had been noted in each treatment after 8 months at the termination of experiments and the survival percentage of graftwas calculated using the following formulas:

$$\text{Graft surivival (\%)} = \frac{\text{Number of survived grafts}}{\text{Total grafted plants}} \times 100$$

- **l) Saleable plants (%):** The plants attaining good height (above 3 feet) and vigor in growth were considered as saleable plants. The number of such saleable plants were recorded at the end of growing season and expressed under:

$$\text{Saleable plants (\%)} = \frac{\text{Number of plants having} \geq 3 \times \text{height}}{\text{Number of successful grafts}} \times 100$$

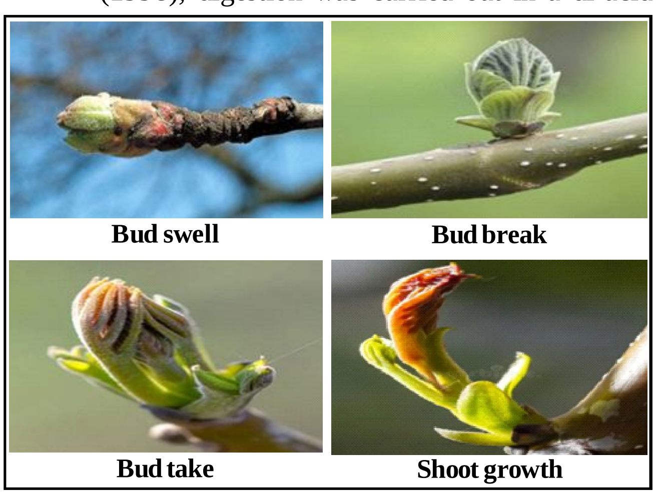

Fig. 1: Different stages from bud break to shoot growth.

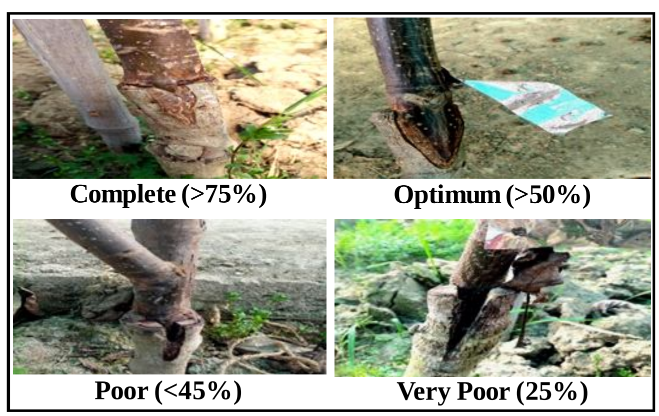

Fig. 2: Graft healing quality.

Table 1: Effect of different scion selections on days taken to bud break and duration of bud break.

Selection	Days taken to bud break	Duration of bud break
WMK-2	38	36.7
WWI-16	36.4	37
WIW-33	**34.7**	37.7
WIA-53	43.4	**41**
WAI-54	40.7	38.7
WIS-100	43	35.7
WHA-213	41.4	36
Mean	39.7	37.6
C.D.($\leq$0.05)	2.9	2.5

Statistical analysis and presentation of data

The observations made over the course of the study were analyzed statistically using the "Analysis of Variance" approach (Gomez and Gomez, 1984). With the use of the software OPSTAT, treatment effect significance and non-significant were assessed. It was compared to the difference at 5% significance to determine whether there was a significant difference in the means.

Results and Discussion

Days taken to budbreak

The number of days required for bud burst was significantly affected by the different scion selections. Among all the selections, the minimum duration for bud burst (34.7 days) was observed in selection WIW-33, as shown in Table 1. This variation can be attributed to differences in chilling requirements, as scion selections with varying chilling needs may exhibit distinct bud break patterns. These findings align with those of Chalise *et al*., (2021), Mir and Kumar (2011), and Soleimani *et al*., (2010). However, our results contradict the findings of Langpoklakpam *et al*., (2017).

Duration of bud break

Our experiment revealed that the longest duration for bud break (41 days) was observed in selection WIA-53, as shown in Table 1. This variation could be attributed to differences in gene expression, with some scion selections potentially exiting dormancy more rapidly than others, leading to discrepancies in the duration of bud break. These findings are consistent with the results of Tanuja (2017) from sapota (Mehta *et al*., 2018; Hartman *et al*., 2001).

Incremental shoot length

Different scion selections significantly influenced shoot length. The longest shoots were observed in selection WHA-213, measuring 107.8 cm. This increased shoot length may be attributed to early bud burst and the subsequent rapid uptake of nutrients and food, as de-shooting was performed every alternate day (Table 2). These results are consistent with the findings of Chalise *et al*., (2021), Thapa *et al*., (2021), and C. Sharma *et al*., (2022).

Incremental shoot diameter

Different scion selections significantly influenced shoot diameter, as shown in Table 3. The highest shoot diameter (22.7 mm) was recorded in selection WIA-53. This larger shoot diameter may result from early and vigorous union formation, enhanced nutrient absorption, and an extended growing season. These findings are

Table 2: Effect of different scion selections on incremental shoot length (cm) at monthly intervals.

Selection	WMK-2	WWI-16	WIW-33	WIA-53	WAI-54	WIS-100	WHA-213	Mean A
15-April (M1)	13.4	14.6	14.4	13.8	16.4	18.5	18.8	15.7
15-May(M2)	20.1(33.5)	18.0(32.6)	18.0(32.4)	20.4(34.2)	18.0(34.4)	20.0(38.5)	23.0(41.8)	19.6
15-Jun(M3)	17.9(51.4)	20.0(52.6)	20.0(52.4)	18.0(52.2)	20.0(54.4)	20.0(58.5)	21.0(62.8)	19.6
15-Jul (M4)	**29.1(80.4)**	**29.3(81.6)**	**29.0(81.4)**	**29.2(81.4)**	**29.4(83.8)**	**29.8(88.3)**	**29.9(92.7)**	**29.4**
15-Aug(M5)	9.5 (89.9)	9.0 (90.6)	10.0(91.4)	10.0(91.4)	10.0(93.8)	9.4 (97.7)	10.0(102.7)	9.7
15-Sep (M6)	5.0 (94.9)	5.0 (95.6)	5.0 (96.4)	5.2(96.6)	6.0 (99.8)	5.3 (103)	5.1(107.8)	5.3
Mean S	15.8	15.9	16.0	16.1	16.6	17.1	17.9	

Table 3: Effect of different scion selections on incremental shoot diameter(mm).

Selection	WMK-2	WWI-16	WIW-33	WIA-53	WAI-54	WIS-100	WHA-213	Mean A
15-April(M1)	10.20	10.80	10.40	10.60	10.70	10.60	10.40	10.5
15-May(M2)	1.6 (11.8)	0.8(11.6)	1.4(11.8)	1.0(11.6)	0.86(11.5)	1.2(11.8)	1.03(11.4)	1.2
15-Jun (M3)	2.6(14.4)	2.8(14.4)	2.4(14.2)	2.8(14.4)	2.8(14.4)	2.8(14.6)	2.7(14.2)	3.1
15-Jul(M4)	**4.2(18.6)**	**3.7(18.2)**	**4.3(18.6)**	**3.9(18.3)**	**4.2(18.6)**	**4.1(18.7)**	**4.4(18.6)**	**4.2**
15-Aug(M5)	2.6(21.2)	3.0 (21.2)	2.8(21.4)	3.1(21.4)	2.6(21.2)	2.6(21.3)	2.8(21.4)	2.7
15-Sep(M6)	1.3(22.5)	1.0(22.2)	0.9(22.3)	1.3(22.7)	1.1(22.3)	1.1(22.4)	1.1(22.6)	1.2
Mean S	3.8	3.7	3.7	3.8	3.8	3.8	3.8	

Table 4: Effect of scion selections on number of compound leaves at monthly intervals.

Selection	WMK-2	WWI-16	WIW-33	WIA-53	WAI-54	WIS-100	WHA-213	Mean A
15-April(M1)	3.17	3.33	3.33	3.12	3.47	3.33	3.33	3.3
15-May(M2)	1.1(4.3)	1.0(4.33)	1.4(4.6)	1.2(4.3)	1.2(4.6)	1.0(4.33)	1.4(4.67)	1.2
15-Jun (M3)	2.0(6.33)	1.3(5.62)	1.2 (5.8)	1.4(5.67)	1.4(6.0)	2.0(6.33)	2.7(6.67)	1.7
15-Jul(M4)	**2.3(8.67)**	**3.1(8.67)**	**2.4(8.21)**	**2.6(8.33)**	**2.7(8.67)**	**2.4(8.67)**	**2.7(9.33)**	**2.6**
15-Aug(M5)	1.7(10.40)	1.6(10.24)	1.8(10.0)	2.0(10.33)	1.4(10.0)	2.0(10.67)	1.1(10.42)	1.6
15-Sep(M6)	0.9(11.33)	1.1(11.33)	1.0(11.0)	0.8(11.17)	1.0(11.2)	1.0(11.68)	1.5(11.87)	1.0
Mean S	1.8	1.9	1.8	1.9	1.9	2.0	2.1	

Table 5: Effect of different scion selections on total number of leaflets per leaf, leaf area, leaf chlorophyll content and leaf nutrient content (N, P, K).

Selection	Total number of leaflets per leaf.	Leaf area (cm^2)	Total leaf chlorophyll content (mg/ g fresh weight).	Leaf N (%)	Leaf P (%)	Leaf K (%)
WMK-2	7.4	303.4	0.35	3.13	0.24	1.55
WWI-16	8.5	277.6	0.31	3.15	0.25	1.57
WIW-33	8.6	287.2	0.34	3.12	0.27	1.63
WIA-53	7.2	296.9	0.35	3.16	0.26	1.63
WAI-54	7.6	314.9	0.36	3.15	0.27	1.55
WIS-100	8.7	295.8	**0.38**	**3.19**	**0.29**	**1.64**
WHA-213	**8.8**	**334.6**	0.34	3.15	0.28	1.63
Mean	8.2	301.5	0.35	3.15	0.27	1.60
C.D.($\leq$0.05)	0.767	32.3	0.025	0.044	0.018	0.014

consistent with those of C. Sharma *et al.*, (2022) and Chovatia and Singh (2000).

Number of compound leaves at monthly intervals

Maximum (11.8) number of compound leaves were observed in selection WHA-213. Whereas minimum number were recorded in selection WIW-33 shown in table 4. It depends upon the genetics and growth pattern. Some naturally produce more compound leaves while others may have less (Chalise *et al.*, 2021; Wani *et al.*, 2017; Mngomba *et al.*, 2010).

Total number of leaflets per leaf

The selection of scion significantly impacts the number of leaflets per leaf, as shown in Table 5. The maximum number of leaflets per leaf (8.8) was observed in selection WHA-213, which is consistent with the findings of Chalise *et al.*, (2021). This increase may be attributed to the enhanced vigor of the plant and the superior growth of the graft union, facilitating more efficient nutrient and food uptake in the grafted plant (C. Sharma *et al.*, 2022).

Leaf area (cm^2)

Different scion selections significantly influenced leaf area. The maximum leaf area (334.6 cm^2) was observed in selection WHA-213, as shown in Table 5. This variation may be attributed to the genetic makeup of the scion, as different selections possess distinct genetic traits, including differences in leaf size and shape (Chalise *et al.*, 2021). Certain genotypes may inherently produce larger leaves, resulting in a greater leaf area compared to other selections. These findings are consistent with the results of Wani *et al.*, (2017).

Total leaf chlorophyll content (mg/g fresh weight)

The highest leaf chlorophyll content (0.38 mg) was observed in selection WIS-100, while the lowest (0.31 mg) was recorded in selection WWI-16, as shown in Table 5. This variation may be attributed to genetic differences, as scion selections can exhibit distinct genetic traits, including differences in chlorophyll production. Certain scion selections may produce higher chlorophyll content, which enhances photosynthetic efficiency and contributes to healthier leaf development (Roza Gnolami, 2020).

Table 6: Effect of different scion selections on graft success, graft survival, graft healing quality and saleable plants percentage.

Selection	Graft success (%)	Graft survival (%)	Graft healing quality (%)	Saleable plants (%)
WMK-2	32.5	85.4	44.0	60.2
WWI-16	37.9	90.3	60.8	47.1
WIW-33	45.4	57.3	48.4	65.3
WIA-53	45.7	73.5	58.4	41.4
WAI-54	52.4	62.3	30.8	60.3
WIS-100	**55.9**	**90.4**	74.5	80.1
WHA-213	23.3	66.4	**84.5**	**86.5**

Leaf nutrient content

Among all the scion selections, the highest leaf nutrient content of nitrogen (N), phosphorus (P), and potassium (K) was observed in selection WIS-100, as shown in Table 5. The variations in leaf nutrient concentrations across the different selections can be attributed to the inherent capacity of each selection to absorb and translocate these nutrients within the plant (Melana, 2010).

Graft healing quality

The highest graft healing quality (84.5%) was observed in selection WIS-100. This may be attributed to genetic factors, as different walnut genotypes possess varying genetic characteristics. Some genotypes produce higher levels of phenolic compounds, which can inhibit callus formation, while others produce lower levels of phenols, thereby promoting more efficient callus development (Table 6).

Graft success (%)

Different scion selections significantly influenced graft success (%). Among all the selections, the highest graft success (55.9%) was observed in selection WIS-100, as shown in Table 6. This can be attributed to genetic compatibility, as successful grafting requires a close genetic match between the scion and rootstock (Akyuz, 2018). Walnut species exhibit varying genetic traits and growth patterns, and selecting a scion that is genetically compatible with the rootstock can enhance the likelihood of a successful graft union and subsequent growth. These findings are consistent with those reported by Chalise *et al*., (2021). Grafting success was evaluated using the formula outlined by Soleimani *et al*., (2010).

Graft survival per cent

The highest graft survival rate (90.4%) was observed in selection WIS-100 (Table 6). This may be attributed to the age and size of the scion, as younger and healthier scions typically have higher survival rates and better compatibility with the rootstock compared to older scions (Rezaee and Vahdati, 2008).

Saleable plants per cent

The highest percentage of saleable plants (86.5%) was observed in selection WHA-213, which aligns with the findings of Singh *et al*., (2019). This may be attributed to the grafting success rate, with healthy and well-matured grafted plants leading to a higher number of saleable plants (Wani *et al*., 2017; Dwivedi, 2000).

Conclusion

It is concluded that selections WIS-100, WHA-213, WAI-54 and WIW-33 showed significantly high graft success, graft survival, graft healing, number of leaflets per leaf, shoot length, shoot diameter, number of compound leaves. Whereas, the minimum graft success, graft survival, number of leaflets per leaf, shoot diameter, shoot length, number of compound leaves, graft healing was observed in selection WMK-2 and WIA-53. Thus, the selection of scion varieties is a critical determinant of the growth characteristics in walnut trees, influencing key growth parameters such as graft success, leaf area, trunk diameter, tree height, and overall vigor. Scion compatibility with rootstocks is a fundamental factor in ensuring grafting success, as genetic and physiological congruence between the scion and rootstock is essential for successful union and subsequent growth. Scions exhibiting larger leaf area and greater physiological vigor typically result in trees with accelerated growth rates, enhanced productivity, and improved overall structural development. Furthermore, the selection of disease-resistant scions contributes to improved health and resilience, potentially reducing susceptibility to pathogens and enhancing the longevity of the trees. Thus, judicious scion selection is integral to optimizing walnut tree growth and productivity, fostering both high grafting success and sustainable, long-term orchard performance.

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