

PROPAGATION METHOD OF EPICOTYL GRAFTING IN WALNUT (*JUGLANS REGIA* L.) UNDER PRODUCTION CONDITION

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Abstract

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The experiment was carried out in 2007-2008 at Fruit-Growing Institute – Plovdiv and on a private fruit tree nursery farm. The elements of the technological process, referring to walnut epicotyl grafting under production conditions, were studied. The following practices were determined: the methods of rootstock production, choosing the cuttings, the grafting methods and the time of grafting, the conditions for callus formation, etc. The results showed that the successfully propagated walnut plants obtained in the different years were from 56.6 % to 63.35 % in average. It was established that the time of grafting exerted an effect on the percentage of survival rate, grafting at the end of March leading to obtaining a higher percentage in comparison with grafting performed at the end of April. The conclusion was drawn that the developed method of walnut epicotyl grafting under production conditions resulted in successful tree propagation and it could be applied in practice.

Key words: walnut, propagation, epicotyl grafting

Introduction

Poor callus formation in walnut made it a difficult to propagate fruit species (Kuniyuki and Forde, 1985; Coggeshall and Beineke, 1997). Out of that reason different methods of producing grafted planting material have been looked for worldwide. Gautam (1990) established that walnut propagation by cuttings was difficult to be realized. According to Rongting and Pinghai (1993a) the presence of high concentration of phenolic compounds in its tissues and their oxidation by wounding was the major reason hampering the application of micropropagation under production conditions. At present budding and grafting are the

most widely used approaches in the production of grafted walnut trees (Gandev, 2007a). As for budding, the most commonly used technique is patch budding (Kuniyuki and Forde, 1985; Germain and Prunet, 1999; Solar et al., 2001; Ozkan et al., 2001), and as far as grafting is concerned – bench grafting technique (Curkan et al., 1975; Korac, 1987; Kantarci, 1989; Tsurkan, 1990; Lantos, 1990; Flores et al., 1995; Unal, 1995). In the last years, hot callus technique has been successfully used in the production of walnut planting material. About 20 000 plants were obtained annually by that propagation method in Turkey (Erdogan, 2006). Unfortunately the grafted trees were not high enough at the end of the season – about 30-50

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cm (Erdogan, 2006; Gandev, 2007b). A principally new walnut propagation technique was announced in scientific literature called epicotyl grafting (Suk-In et al., 2006). The authors established that the survival rate was 78.9 % in average when applying that technique.

The aim of the experiment was to establish a method of walnut epicotyl grafting under production conditions, describing in details the technological process – rootstock production, scions selection, technique and time of grafting, temperature conditions, etc.

Material and Methods

The experiment was carried out in 2007 and 2008 at Fruit Growing Institute – Plovdiv and in a private fruit nursery. The methodological approach is based on own results from preliminary studies regarding different elements of the presented method of propagation.

Walnut seeds of common walnut (*Juglans regia* L.), selected for obtaining rootstocks, were immersed in water at the middle of December. After 4-5 days they were taken out of water and put for stratification in tied plastic bags at a temperature of 3-6°C. After the seeds cracked open, they were temporary planted in pots of 14 l on two floors. The substrate for covering the seeds consisted of



Fig. 2. Grafted plants with new growth

peat and perlite at 2:1 ratio. Each pot contained about 30-40 seeds. Thus processed, the seeds were left for germinating in a non-heated greenhouse. When the seeds formed a well-developed root and a juvenile epicotyl, split grafting was performed with 5-8 cm long one-year old scions (Figure 1). The scions were collected from mother trees during dormancy and they were stored in a refrigerator until the time of grafting. During the grafting procedure the vascular cambium of the scion and the rootstock were accorded with each other. The graft was fixed with plastic folio, which was not tight in order to allow the exit of surplus moisture at the place of grafting. According to Pinghai



Fig. 1. Grafted plant



Fig. 3. Epicotyl grafted plants in pots

and Rongting (1993b) excess bleeding caused the formation of anaerobic conditions at the graft juncture resulting in a decrease of graft survival. The root tip of each rootstock was cut in order to stimulate the formation of new lateral rootlets. The grafted plants were dipped into fungicide solution to prevent from walnut bacteriosis (*Xanthomonas campestris* p.v. *juglandis* Pierce) and walnut anthracnose (*Gnomonia leptostyla* Fries) and then placed horizontally in plastic basins. The roots were covered with moist substrate of pine sawdust and perlite at 1:1 ratio. The place of grafting was not covered with substrate, just the opposite it was isolated from the moist substrate with a polyethylene band. Each plastic basin with grafted plants was put in a nylon bag which was tied to provide air humidity to the plants. Thus handled, the basins were placed on shelves in a room with controlled temperature of 25-27°C. After 14-20 days the plants formed callus and started growing (Figure 2). When the new growth reached a length of 4-6 cm, the plants were temporarily planted in plastic bags of 15/20 cm size and left for adaptation. After adaptation they were planted in pots of 14 l (Figure 3).

In 2007 grafting was performed on 27 March and 30 April and in 2008 – on 25 March and 29 April. Izvor 10 cultivar was grafted on common walnut (*Juglans regia* L.) rootstock. In the first experimental year by 60 plants were grafted on each date and in the second one – by 250 plants.

In 2008, 2180 common walnut (*Juglans regia*

L.) rootstocks were grafted with Izvor 10 cultivar in a commercial walnut nursery. Grafting was performed every 4th – 5th day – from 25 March until 29 April 2008.

When the plants were adapted, the percentage of survival rate was reported. The data were statistically analyzed by Duncan's test (Steele and Torrie, 1980).

Results and Discussion

The results presented in Table 1 showed that in 2007 the percentage of successfully propagated plants was 63.35 in average and in 2008 – 56.6 %. It should be mentioned that in both years of the experiment the percentage of the survival rate was higher for the first period of grafting, the differences being statistically significant. In 2007 from the grafting performed on 27 March the successfully propagated plants were 75.0 % versus 51.7 % from grafting performed on 30 April. A similar result was obtained in 2008 – 71.2 % survival rates from grafting performed on 25 March versus 42.0 % from grafting on 29 April. Obviously the time of grafting influenced the survival rate and the results were in favour of the earlier grafting date. We suppose one of the reasons for the lower percentage of successfully propagated plants at the later date of grafting was the prolonged storage of the scions in the refrigerating chamber (about two months), which decreased their vitality. A proof of that was the fact that in the second date of grafting

Table 1

Percentage of successfully propagated walnut plants of cultivar Izvor 10 during 2007 and 2008

Cultivar	Date of grafting	Number of grafted seedlings, n	Number of successfully grafted plants, n	Percentage of survival rate, %
Izvor 10	27 Mar. 2007	60	45	75 a
	30 Apr. 2007	60	31	51.7 b
				average 63.35
Izvor 10	25 Mar. 2008	250	178	71.2 a
	29 Apr. 2008	250	105	42b
				average 56.6

Table 2**Percentage of survival rate in a nursery under production conditions during 2008 year**

Cultivar	Period of grafting	Number of grafted seedlings, n	Number of successfully grafted plants, n	Percentage of survival rate, %
Izvor 10	25 Mar. - 29 Apr. 2008	2180	1285	58.9

about 15% of the scions were obviously unfit for grafting.

Data presented in Table 2 showed that in commercial production when grafting in the nursery was more time consuming and it could not be performed only on the two dates mentioned, but it took a whole month, the successfully propagated plants were 58.9 %. That percentage was close to the average percentage of the survival rate during the two experimental years presented above in Table 1.

Conclusions

The described method of walnut epicotyl grafting under production conditions resulted in successful propagation.

The time of grafting influenced the success of propagation. Performing grafting at the end of March led to obtaining a bigger percentage of grafted plants compared to grafting performed at the end of April.

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