

NEWEST DEVELOPMENTS IN ROSE (*Rosa hybrida*) PROPAGATION

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RESUMEN. La productividad de las rosas cortadas puede ser mejorada mediante el uso de un material inicial bueno. Las nuevas técnicas de propagación, control de factores que influyen en el procedimiento de la propagación y selección de revigorizantes portainjertos son herramientas promisorias. "Stenting" (simultáneamente enraizamiento e injertación) e injerto de raíz son técnicas que todas pueden ser aplicadas, durante todo el año, porque los portainjertos son propagados vegetativamente, las plantas serán menos heterogéneas que cuando se injertan sobre semilleros. Sin embargo, estas técnicas son capaces de clonar selecciones para una nueva generación de vigorosos portainjertos. Algunos aspectos investigados son presentados aquí. La aplicación basal del AIB para promover la formación de raíces de los "stentings" retardaron la formación de un xilema vascular, mientras que el incremento del almidón en las partes del tallo de la yema (injerto) y portainjerto se retrasó también. Turnos de humedad con pérdidas de 9% de agua por "stenting" diario, así como una alta temperatura de 32°C durante el primer día después del "stenting", demostró ser promotor del desarrollo del xilema conectado a la unión del injerto. El tratamiento de pre- enraizamiento del portainjerto antes "stenting" incrementará el período vulnerable entre el injerto y el endurecimiento de salida en dos semanas. "Stenting" portainjertos pueden influir en el temprano desarrollo de rosas arbustivas. El efecto revigorizante estimuló el diámetro del cuello de la raíz y brotaciones basales así como las brotaciones basales y flores.

PALABRAS CLAVE: injerto, unión del injerto, propagación, injerto de raíz, portainjerto, rosa, *Rosa híbrida*, stenting, y desarrollo de herida.

SUMMARY. Productivity of cut roses can be improved by better starting material. New propagation techniques, control of factors influencing the propagation procedure and selection of invigorating rootstocks are promising tools. Stenting (simultaneously rooting and grafting) and root grafting are techniques which can be applied all around the year. Because the rootstocks are propagated vegetatively, the plants will be less heterogeneous than when grafted on seedlings. Moreover these techniques enable clonal selection for a new generation of vigorous rootstocks. Some investigated aspects are presented here. Basal application of IBA to promote root formation of stentings delayed the formation of a xylem vessel connection, while the normal increase of starch in the stem parts of scion and rootstock was delayed too. Alternation of humidity resulting in 9% water loss per stenting daily, as well as a high temperature of 32°C during the first day after stenting, proved to be promotive for development of xylem connection of the graft union. Prerooting treatment of the rootstock before stenting will decrease the vulnerable period between grafting and hardening off up to two weeks. Stented rootstocks can influence the early development of rose bushes. The invigorating effect stimulated the diameter of root collar and bottom-breaks as well as the number of bottom-breaks and flowers.

KEY WORDS: Grafting, graft union, propagation, root grafting, rootstock, rose, *Rosa hybrida*, stenting, wound development.

Abbreviations: IBA-4 (3-Indolyl) butyric acid.

INTRODUCTION

Rose is world's cut flower number one with a total greenhouse production area in the main countries of more than 5000 hectares (Pertwee, 1995). With an average life span of a rose crop of 5 years around 1000 hectares have to be planted with roughly estimated at least 70 million new rose bushes every year all over the world. Especially in Europe the rose is very popular. Alt-

hough the population of Europe is only about 5% of the world, 60% of the world's cut rose area is in Europe, while still roses are imported from other continents. Holland is the leading country in rose production with over 900 ha modern glasshouses.

Rose plants are valuable means of production. During its life-time a bush can produce a value of around U.S. \$ 50. An important problem is, that in spite of control of environmental factors to a high degree in modern

glasshouses, rose bushes are very heterogeneous. Consequently, production value may vary from U.S. \$ 15 to U.S. \$ 75 between poor and good bushes. In view of current expenses per m², the required control of quality and price per flower, this variation is unacceptable in 1995.

The Department of Horticulture of the Agricultural University Wageningen investigates quality control of rose bushes during the early phases of plant development. Quality means: uniformity at a desirable number of characters. During propagation many factors exert an influence on plant quality. Our research aims at both detecting and controlling these factors. Propagation may be divided into four phases:

- **Starting material:** leaf axillary bud, shoot and rootstock, all in different qualities.

- **Propagation method:** A choice of 6 methods: cuttings, micropropagation, bench grafting, budding, stenting (= cutting-grafting), root grafting. All methods vary in technique which, in turn offer interesting possibilities of improvement and combination of advantages.

- **Conditions during propagation:** temperature, light, humidity, auxin and rooting substrate.

- **Plant treatment after propagation:** Time of planting, plant density, treatment of primary shoots and bottom-breaks.

As a most important means of production, future rose bushes should meet the highest requirements as to external and internal quality. In this paper some new developments in rose propagation will be presented.

MATERIAL AND METHODS

One of the first decisions before propagation starts, is the choice between rooted cuttings without rootstock or grafting on rootstocks. Because roses are initially selected by the breeder as seedlings on their own roots, all varieties have roots of reasonable quality. In most cases propagation, size control and tolerance for unfavourable conditions are main reasons for grafting. In Holland grafting of roses for example is mainly done to improve productivity and flower quality (Van de Pol *et al.*, 1994). Several types of rootstocks are used all around the world. In the North-Western part of Europe selections of *R. canina* seedlings were used until 1980. These seedlings were grafted by budding or bench grafting. Since that year and with the introduction of artificial substrates, scions on their own roots and clonal rootstocks were more and more used. Development of new techniques enabled selection and propagation of new clonal rootstocks. Recently the rootstock 'Natal Briar' became very popular with about 5 million propagated plants in 1994. Around the Mediterranean Sea *R. hybrida* 'Indica Major' is generally used, whereas in Japan

and South Africa *R. multiflora* and in Latin America and the U.S.A. *R. hybrida* 'Manetti' is very popular.

Stenting (Fig. 1) is a quick propagation technique based on grafting a cultivar on an unrooted cutting of a rootstock. This method is applicable the year around and therefore ideal for studying scion-rootstock interre-

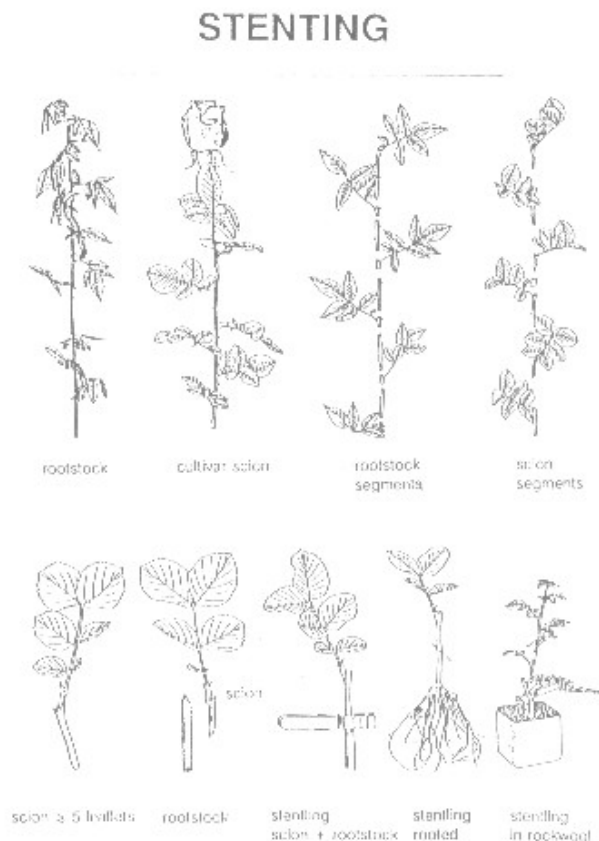


Figure 1. Schematic representation of stenting, a grafting technique based on grafting of a scion on a stem of a rootstock which has to be rooted simultaneously.

lations. The technique used is whip grafting (Hartmann *et al.*, 1990). As scions are used good five leaflet leaves with corresponding axillary bud and piece of stem. As rootstocks are used pieces of stem consisting of single internodes without buds and leaves. The top of the rootstock internodes and the basal part of the scions are cut at an angle of 30° (Photo 1).

Scion and internode are combined and held together with a clothes-peg. For a good development of the graft union it is necessary that at least at the side where the leaf is located the cambia of the graft partners are in close contact. To prevent wilting the leaves are moistened. In case auxin is applied, the rootstock internodes are dipped in talc powder with indole butyric acid (IBA)

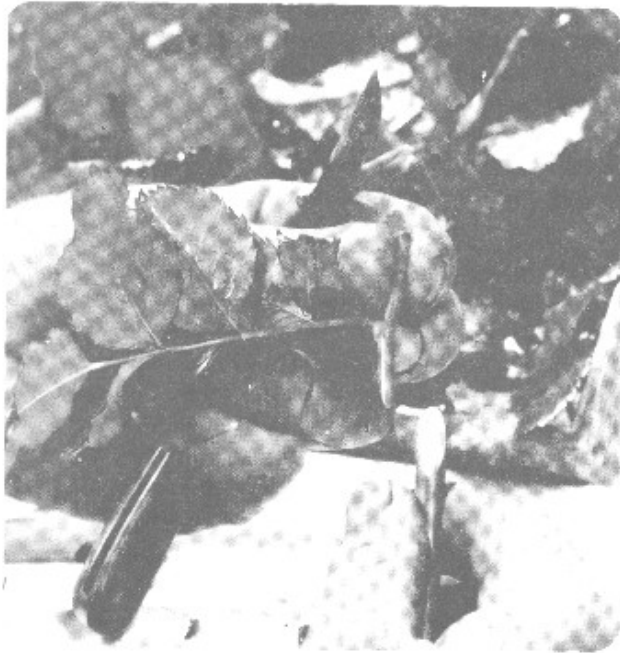


Photo 1. Stenting of rose. Combination of wounds.

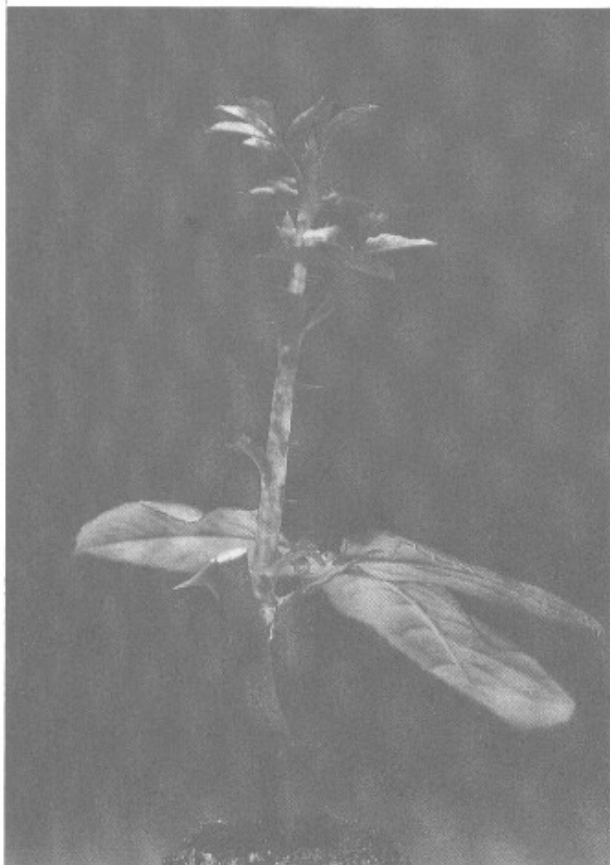


Photo 2. Propagated rose, three weeks after stenting.

0.4%, to accelerate root formation. The plants are placed in a propagation bench, covered with double layered polyacrylic. The rooting substrate is a mixture of sand and peat at a ratio of 1:1 (v/v) at pH 5.5, or cocos fibres, or Rockwool (Van de Pol et al., 1982 and 1986). Photo 2 is a picture of a young stentling.

Root grafting (Fig. 2) is a similar technique as stenting instead of an internode of a stem as a rootstock, a piece of root of around 5 cm in length and 3 mm in diameter is whip-grafted. This technique is developed for screening the root quality of seedlings after grafting, when shoots of the rootstock are not available anymore. Moreover, rootstocks with problems of rooting of their cuttings, like the *Rosa canina* group, can be successfully screened by root grafting (Van de Pol, 1986). From one root-system many pieces of root can be used, so seedlings can be cloned and selected (Vonk Noordegraaf, 1990).

ROOT GRAFTING

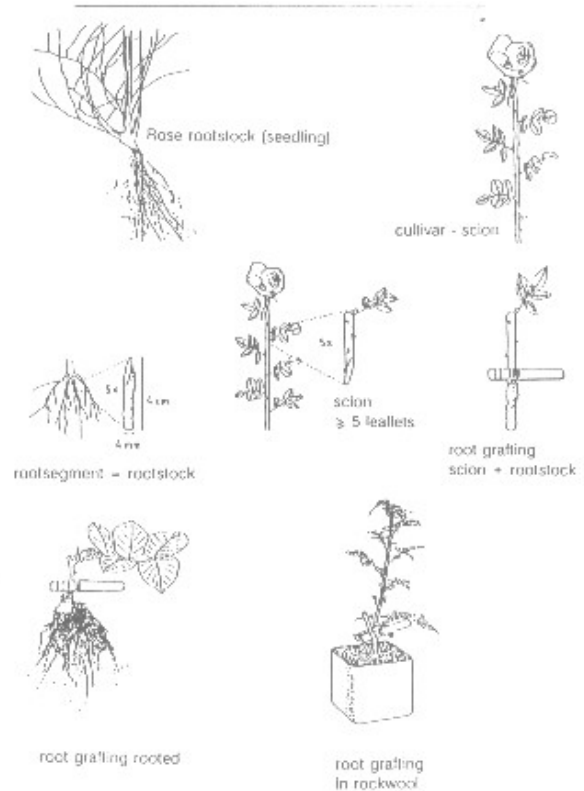


Figure 2. Schematic representation of root grafting of roses, a technique based on grafting a scion on a piece of root as rootstock.

RESULTS

To examine the changes of starch in a stentling, *R. hybrida* 'Ilona' was grafted on unrooted internodes of the same cultivar. The changes in starch level in the stem of the scion and rootstock are presented in Fig. 3. Starch level is recorded using an arbitrary scale, described by Van de Pol *et al.*, (1986). Fig. 3 shows a marked increase of the starch level of the scion, particularly during the first week after grafting. Starch in the rootstock shows a sharp decrease, directly after grafting, reaches level zero on day three and subsequently increases. It is remarkable that basal application of IBA delays the starch increase from day 3-10 in the scion as well as in the rootstock.

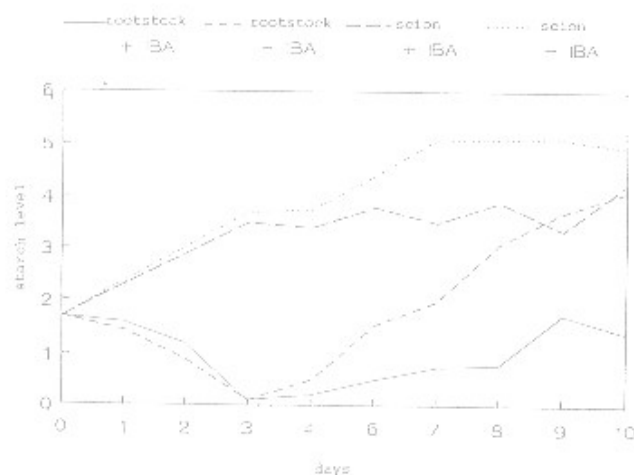


Figure 3. Changes in starch level of rootstock and scion stems during 10 days after stenting *R. hybrida* 'Ilona' on 'Ilona'. The influence of basal application of IBA (0.4% in talcum powder) on the starch level is indicated.

In Fig. 4 the development of the phloem and xylem connection after stenting *R. hybrida* 'Ilona' on 'Ilona' is presented. Labelled CO₂ was applied to the leaves and by measuring the radioactivity below the graft union, the development of the phloem could be recorded. The xylem connection was investigated by colouring the connected vessels with acid fuchsine applied to the basal wound of the rootstock (Van de Pol *et al.*, 1986). Fig. 4 shows that phloem connection begins at day five and xylem connection at day six, while after one week all the stentlings show phloem as well as xylem connection.

Fig. 5 shows the effect of basal application of IBA on development of the percentage of stentlings ('Ilona' on 'Ilona') with xylem connection. In this experiment the development of the graft union is delayed when root formation is promoted by IBA.

To examine the wound development of two graft unions in one plant double stentlings were made of the combination *R. hybrida* 'Sonia' / 'Sonia' / 'Sonia'. In this

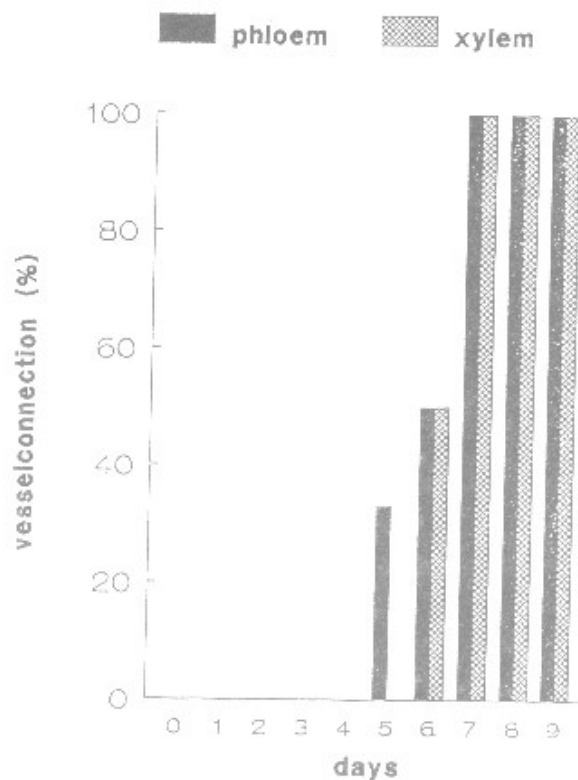


Figure 4. Development of phloem and xylem connection during 9 days after stenting *R. hybrida* 'Ilona' on 'Ilona'. Phloem connection was observed by labelled CO₂ application, xylem connection was made visible by acid fuchsine colouring.

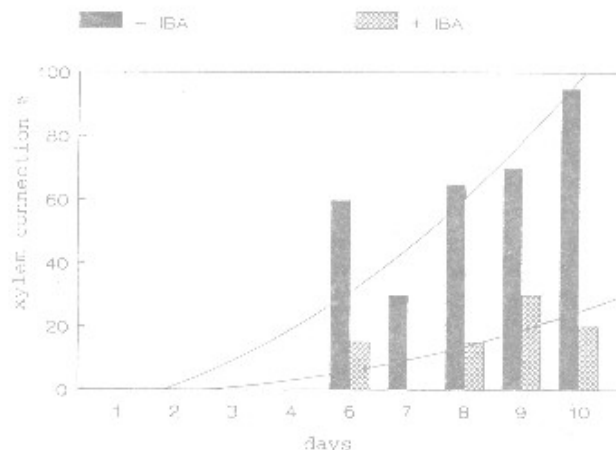


Figure 5. The effect of basal application of IBA (0.4% in talcum powder) on the percentage of plants with xylem connection during 10 days after stenting *R. hybrida* 'Ilona' on 'Ilona'.

study a leafless and budless internode of 'Sonia' was used as an interstock. The rate of healing of the two graft unions was compared by the acid fuchsine met-

hod. After 12 days the % of connected vessels was observed by estimation of the % of red coloured xylem tissue. Table 1 shows that the upper graft union of double stentlings has a much faster development of xylem connection than the lower one.

TABLE 1*. Xylem connection in double stented stentlings of Rosa Hybrida cv. Sonia/Sonia/Sonia 12 days after stenting.

Position	% of graft unions with xylem connection	% of connected vessels
Upper union	100 a	34 a
Lower union	17 b	3 b

* Different letters per column indicate significant differences (P< 0.05).

Recently a large scale facility was built in Holland to propagate roses under completely controlled environmental conditions of light, temperature, CO₂ as well as humidity of air and substrate. The facility contains four units of six layers in which around 100.000 roses per three weeks can be stented. Important question now is, what are the optimal conditions per period for the total duration of 500 hours (= three weeks) of the stenting process.

Hypothetically we supposed that the first phases of wound development with cell division can be promoted by high temperature. Table 2 shows the effect of temperature during the early development; it can be concluded that one day at 32°C promotes the xylem vessel connection without negative effect, while two days at 32°C is promotive for development of the graft union but causes damage to the leaf of the scion.

TABLE 2*. The influence of 0, 1, 2, 3 or 4 days at 32°C on the xylem connection of Rosa hybrida cv. 'Madelon' on 'Ludiek', 9 days after grafting (N= 14).

Days at 32°C	%of plants with xylem connection	% of connected xylem vessels (mean)	% of plants with leaf damage
0	100	25 b	0
1	100	40 c	0
2	100	67 c	29**
3	65	10 b	64**
4	0	0 a	42**

*Different letters per column indicate significant differences (P< 0.05).

**Brown colouring of the cambium of the scion.

An example of the effect of humidity is presented in Table 3. Here continuous 100% humidity of the air during the early development was compared with daily waterlosses of the stentlings. The waterloss was measured by weighing the plants during dry conditions varying from 0 to 2.5 hours. Subsequently the plants were replaced to the wet conditions. Daily waterloss up to 9% proved to be promotive for the % of connected xylem vessels after 10 days, while 13% was clearly negative.

TABLE 3. The influence of daily waterloss on xylem connection, 10 days after stenting Rosa hybrida cv. 'Sonia' on 'Indica Mayor'. The % of connected vessels are presented as scores: 1= 1- 25%; 2= 26-50%.

Daily waterloss (%)	Xylem development % of plants with connection	Score
0	60	0.95
3	75	1.05
5	70	1.50
7	75	1.30
9	75	1.90
11	50	0.90
13	35	0.55



Photo 3. Rootstock internode after 9 days of prerooting treatment. At the base root initials are just appearing.

In some cases, pretreatment of the rootstock before stenting can be useful. Rootstock internodes with leaves were prerooted as cuttings until the stage of formation of root primordia, indicated by a swollen base (Photo 3). Dependent from the conditions and rootstock type this pretreatment will last around 10 days. When this material is grafted its leaf with axillary bud is removed. Advantages of this method are that immature rootstock material can be used and that the vulnerable period between grafting and hardening off can be shortened up to two weeks (Photo 4).

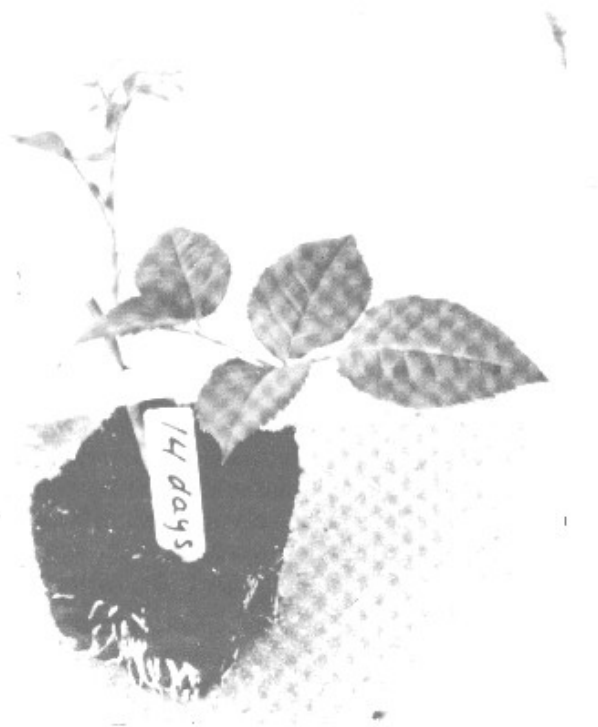


Photo 4. Propagated rose, two weeks after stenting *R. hybrida* 'Frisco' on a rootstock internode which was prerooted as a cutting (see also Photo 3).

The effect of rootstocks on the early development of rose bushes is presented in Table 4. In this experiment *R. hybrida* 'First Red' was stented on 26 rootstocks compared with 'First Red' on own roots. The plants were grown in Rockwool. The experimental set up was groups of 3 plants of the same combination, planted in a row, with replication in 6 blocs. Here the invigorating effect of the rootstocks 18-31 (a new rootstock candidate) and 'Natal Briar' is demonstrated by the diameter of the root collar, the number of bottom breaks and the average diameter of the bottom breaks. As a result the flower production was positively correlated with the plant architecture. These differences in flower production remained stable for several years.

TABLE 4*. The influence of roots on the early development of *R. hybrida* cv. 'First Red', 6 months after propagation, R.C= root collar, B.B= bottom break, H.S= harvest shoot, (N= 6 X3).

Stock name or code	Diameter of R.C.	Number of B.B	Diameter of B.B	Number of H.S
18-31	18.85 a	2.06 a	10.30 a	5.61 a
'Natal Briar'	16.20 b	1.89 ab	10.19 b	5.33 a
Own roots	12.53 c	1.61 b	8.55 b	3.61 b

*Different letters per column indicate significant differences ($P < 0.05$).

DISCUSSION

Productivity of cut roses can be improved by better starting material. New propagation techniques, control of factors influencing the propagation procedure, and selection of invigorating rootstocks are promising tools. In this paper some examples of these factors are presented.

Stenting and root grafting are techniques which can be applied all around the year. Because the rootstocks are propagated vegetatively the plants will be less heterogeneous than when grafted on seedlings. Moreover these techniques enable clonal selection for a new generation of vigorous rootstocks.

A stentling is a complicated plant. During the early development several physiological processes occur: photosynthesis in the leaf, demonstrated by the pattern of the starch level, development of the graft union, and root formation at the basal part of the rootstock internode. The faster development of the upper graft union of a double stenting indicates the influence of the scion leaf on the vascularization. When IBA is applied to promote root formation, the increase of the starch level in the stem is delayed. This can be explained by energy consumption due to stimulated cell activity. The delayed formation of xylem connection after basal application of IBA points to competition between the development of the two wound regions. Ypema *et al.* (1987) found that young rootstocks can be sensitive to black rot, caused by fungi. This effect proved to be stronger when IBA was applied. Low energy level due to immaturity and IBA effect can be the explanation for this effect. When rootstocks are prerooted as a cutting, the starch pattern as well as the competition between the wounds will be changed. The experiment with humidity is an indication that a certain degree of activation of the plant by water loss is stimulating for the xylem vessel development.

The influence of propagation techniques on plant quality is demonstrated in several woody crops. Coles



Photo 5. *R. hybrida* 'Vivaldi' grown on Rock-wool, 12 weeks after propagation.
Left: stented on *R. multiflora* Cathayensis cv. 'Multic'.
Right: 'Vivaldi' on own roots.
 The stented plants were much earlier with formation of bottom- breaks and flower Production.

and Campbell (1969) reported better apple trees after inverted 'T' budding in stead of normal 'T' budding. Skene *et al.* (1983) showed better plant development of fruit and ornamental trees, when propagated by chip-budding in stead of T-budding. Howard (1992) reported that the ratio between leaf area and stem diameter of softwood cuttings of *Syringa spp.* was positively correlated with the rooting response. One of the advantages of the stenting technique is that the leaf area of the scion and the diameter of the rootstock can be chosen in an optimal ratio.

A major reason for grafting roses is the use of an invigorating rootstock. For certain of the major tree fruit crops a wide assortment of size controlling rootstocks has been developed now (Brase and Way, 1959, Hartmann *et al.*, 1990). In apples even a complete range of tree size, from very dwarfed to very large, has been obtained, with a given scion cultivar grafted to different rootstocks (Preston, 1966). A remarkable effect of a rootstock can be the promotion of the formation of bottom-breaks (Photo 5). The rootstock effects presented in this paper, are in line with other experiments (Kool and Van de Pol, 1992). They reported that differences between rootstocks, expressed in quality of bottom-breaks and branches were clearly pronounced 100 days after pinching the bottom-breaks. Quality of bottom-breaks and laterals was highly positively correlated. It seems possible to predict the quality of a certain rose crop from plant parameters during the early stage of development.

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