RESEARCHES CONCERNING RASPBERRY AND BLACKBERRY MULTIPLICATION BY CLASIC AND MODERN METHODS

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ABSTRACT

Key words: multiplication, byotechnologie, raspberry, blackberry, cuttings

Raspberry and blackberry without thorns multiplication is an important activity and should be doing on scientific basis only in specialized units. Following investigations were drawn a number of conclusions, important for fruit growing practice. We studied new raspberry varieties and blackberry without thorns and objectives set at the beginning of investigations have been conducted in totality.

Propagation by suckers is practiced to species which have buds adventives on roots. During the vegetation from these buds are obtained shoots known as suckers. The suckers, because they are juvenile forms are origin in secondary meristems (periciclo). The juvenility of these suckers is expressed through a pronounced vitality, and a high capacity to form adventitious roots and meristematic tissues. Researchers from pomiculture practice have revealed certain technological measures to increase the number of adventitious buds on the roots. The number of adventives buds on the root may increase considerably, which entails a large number of suckers if the root system is slightly wounded before the vegetation.

In vitro cultivation of vegetal material (meristematic tissues, cells, somatic embryos) have many advantages, among which the most important is to ensure a rate of multiplication which cannot be reached by any of the traditional methods.

By this modality, at many crop species, especially ornamental plants, trees and shrubs and trees, ensure rapid multiplication to new varieties, which could even allow rapid change of the existing assortment.

Researches concerning extending growth of raspberry and blackberry varieties valuable by conventional and biotechnological methods have targeted a series of clear objectives, precise with real chances to be achieve.

The main objectives of this research were:

- checking the response of raspberry and blackberry varieties without thorns at multiplying in vitro

- indication of the culture medium who assure the inoculum initial multiplication and forming shoots,

- establishing the hormones who assure implants multiplication and their concentration.

- performing the rooting to shoots obtained in vitro in the same time with acclimatization,

- testing some methods for rooting of shoots produced in vitro.

- determining the rate of multiplication of new varieties.

The objectives concerning multiplying by conventional methods were:

- researches of conventional methods known and less applied in multiplication raspberry and blackberry,

- the influence of substrate rooting and rhyzogene substances on the percentage of rooting and root system development,

- determining the best moment for conducting operations vegetative multiplication at raspberry and blackberry.

- suitability to vegetative propagation of varieties studied.

- establishing of the simplest and most efficient conventional methods of multiplication.

Laboratory experiments were conducted in micropropagation laboratory of the Faculty of Biotechnology from Bucharest. Specializing in plant biotechnology, the laboratory with modern equipment is specific to this sector and has teachers and highly trained researcher.

I he investigations were carried out at SC Frasinu S.A. Buzau, which is located in the south of the city of Buzau, on the road Sloboziei at km 2.

The biological material was represented by 5 varieties of raspberries, one created in Romania and three varieties of blackberry without thorns; two newly created varieties compared a variety known long ago and unrivaled in terms of the biological and technological features.

The researches were carried in 2010-2012 and were oriented in two directions. A first line of research aimed raspberry and blackberry multiplying by laboratory methods (biotechnology) and the second direction was oriented raspberry and blackberry multiplication using conventional methods. The laboratory researches was conducted in the micropropagation laboratory of the Faculty of Biotechnology from Bucharest, and to verify the conventional multiplication methods, we used material and technical basis to SC Frasinu Buzau S.A. Located in the immediate vicinity of the city of Buzau, has profile agro-zootechnic and has experimental plots with very fertile land, and greenhouses fitted with platform rooting and tables for hydroponic culture, climate chamber, the system of drip irrigation, a grove columnar trees and skilled labor.

For the laboratory experiments to test initiation phase Murashige & Skoog we used medium of worship in three variants to determine which provides the best environment for explanls evolution. The difference between the three variants of culture medium was given the concentration of growth regulators BAP. The first variant of culture medium (VI) contained 0.1 mg / 1 BAP, the second variant (V2) contained 0.5 mg /1 BAP and the third variant (V3) contained 1 mg /1 BAP.

Multiplying known as propagation phase was phases in which the callus formed young shoots.

For the multiplication phases we used medium Murashige & Skoog. medium to which the following substances were added: -100 mg myo-inositol/1, vitamin B1 (thiamine) - 1 mg/1 nicotinic acid (vitamin B3) - 0.5 mg/1 Vitamin B6 (pyridoxine), sucrose - 30 g/1, agar - 6 g/1 benzylaminopurine (BAP) - 1 mg/1. indolyl butyric acid (IBA) - 0.1 mg/1, gibberellic acid (GA3) - 0.1 mg/1. The pH was adjusted to 5.8. This phase lasted about three weeks, and temperature from room growth was 21-24 oC. At the end of this phase the shoots occupied all the space in the vessel, we made the shoots and their rootedness in same time with acclimatization.

fhe rooting shoots. The rooting shoots can be carried and culture medium and in ex vitro. I chose to make ex vitro rooting considering that I worked with a species who make roots easily, I want to reduce the period for obtaining the material.

Rooting was done concurrently with acclimatization. We experienced three ways of rooting the shoots - rooting shoots in pots Jiffy introduced in the growth chamber, rooting shoots in pots Jiffy placed in rooting platform in greenhouse and rooting shoots in hydroponic sistem. The temperature in this stage was maintained at 20-22° C, and the roots began to appear after 10-12 days in hydroponics and after 15 days Jiffy pots type. At hydroponic culture shoots were planted in cellulosic material pots with perlite, after which the pots were placed in nutritive solution. All three methods were tested version rooting hormone-free rooting and rooting hormone. The hormone product used for rooting, was Bioroots product applied as a liquid spray immediately after planting seedlings in recommended concentration of 0.5%.

The product is applied once, and the system is introduced in hydroponic nutrient solution.
In the field experiments was verify behavior raspberry and blackberry without thorns using certain methods. At raspberry was tested the behavior of 5 varieties at multiplication by mature suckers, multiplication by young suckers, propagation by cuttings green.

To evaluate the formation potential of suckers, at varieties raspberries at the end of vegetation period was carried evaluations suckers. The plants had several years of growth. The plants from my experience had 3 years old at the time of evaluation. The ability of each variety to form suckers was highlighted by manual picking of all plants formats.

Analyzing the results obtained in the initiation phase variety Citria notes that we started from a starting material (uninodal fragment) with an average weight of between 24.6 and 63.6 mg and obtained a gagging with an average weight of between 74.8 mg and 215.1 mg.

The average multiplication rate (media/rehearsal) registered a low of 170.2%, while the maximum was 218.0%. Average multiplication rate was 15.5 per variety.

At Citria variety the average multiplication rate (repetitions average) registered a value of 197.5%, which demonstrates that the culture medium used to stimulate inoculated material development.

Based on the comparative study of the values at the five varieties of raspberry it was found that the original weight of inoculum realised high growth.

The rate of multiplication of varieties studied recorded values between 196.2% (Noma) and 216.9 (Willamette). Varieties may be aranged according to the rate of multiplication follows: Noma (196.2%), Heritage (197.2%), Citria (197.5 %). Polka (205.2%) and Willamette (216.9%).
As regards the rate of multiplication, variety Orkan exceeded witness with 22.6%, while the variety Silvan witness was exceeded by 7.2%. Compared to multiplication average of varieties, Orkan had value positive of multiplication rate, 17.5% over average.

Multiplication phase began with the passing initiation callus on the multiplication stabilization medium. At the end of the initiation phase, the callus was removed from the test vessels, rinsed with distilled water and passed on multiplication medium from VI variant, with growth hormone. Benzyl aminopurine (BAP) is a plant growth regulator acting cytokinins to stimulate cell division. Indolilbutiric acid (IBA) has a role in stimulating cell division and elongation processes of cells, controls apical dominance processes, modifies the permeability of cell membranes. Gibberellic acid (GA3) also stimulate cell division, promotes regeneration of shoots, stimulates the formation of axillary shoots, delay the onset of senescence cells and stimulates the growth of chlorophyll content.

Compared with Citria variety considered witness, Heritage formed a great number of shoots for rooting percentage was 85.5% higher than the witness. Variety Willamette achieved 65.6 shoots on variety, with 44.6% more compared to the control. A variety with high capacity to form shoots

was and the variety Polka and Noma, forming more shoots compared with witness with 72.7% and 63.5% (145.19%). Variety Polka formed 78.6 shoots, with 33.1 more compared to witness variety.

Of the five varieties studied, varieties Heritage. Willamette, Noma and Polka have recorded values much higher than witness (Citria).

At blackberry without thorns the shoots number for rooting was higher than raspberry species, the minimum number of shoots beeing close to the raspberry maximum level. At Thorn Free variety, was obtained 3,3 shoots on the inoculum considered witness and 3.7 shoot at Orkan variety, while the variety Silvan had a value of 3.1 shoots. The witness was exceeded only by the variety Orkan. Variety Silvan formed compared by witness with 4.7% less shoots and the variety Orkan formed with 12.6% more shoots.

Shoots obtained in vitro were removed from culture vessels, rinse with distilled water and prepared for rooting. The following rooting methods were experimented:

- rooting in ex vitro in type Jiffy pots kept in the growth chamber.
- rooting in ex vitro in type Jiffy pots, placed in platform rooting in greenhouse.
- rooting in hydroponics system.

Shoots obtained from the culture medium were conditioned on two categories of length - 2-3 cm and 3-5 cm. In variant V2 rooting in growth room, shoots of raspberry of 2-3 cm in length had an average rooting percentage of 82.5%, and the shoots of 3-5 cm in length have an average rooting 85,3 %. This decrease in the percentage of rooting can be explained by a weaker control of vegetation factors in the greenhouse.

In this system the rooted shoots percentage was very high in both variants. In variant with nutrient solution without hormones rooted the percentage of rooting to shoots was 95.7% for the shoots of 2-3 cm and 96.4% for the shoots of 3-5 cm.

Adding Bioroots product in the nutrient solution increased the percentage of rooted shoots, but the percentage increase was reduced. In variant V2 rooting percentage had high values being close to 100%. The blackberry without thorns in hydroponic system had a rooting to shoots of 2-3 cm in an average of 97.5% (VI) and 99.2% (VI) when the shoots had 3-5 cm long. In variant V2 between the two types of length of the shoots there was no difference, the rooting percentage was 100%.

In multiplication by suckers mature varieties Polka and Noma had formed the highest number of steems, followed almost to Willamette variety and the lowest number of stems was formed to Heritage and Citria variety. However even at varieties that formed less strains the results

are good and can be taken in cosideration by producers of seedlings. The number of suckers that can be obtained per hectare is between 44 328 pes. (Heritage) and 68659 pieces (Polka). The 5 varieties had an average production potential of strains of 57.860 pes./ha. Multiplications by young suckers began by cutting the stems at the soil surface.

As a result, the number of suckers who appeared on the soil surface was big. Variety Willamette has formed 23 suckers and Heritage varieties and Polka formed on average 21 suckers. Variety Noma formed the smallest number of strains. The average number of young suckers that can be obtained per hectare amounts to 65.259 pes. After suckers datased from the mother plant was done mocirlirea their immediatly and planted in place of production, in accord with of planting distances. The comments continued to specify the percentage of new suckers. Catching was very good in 100% at all varieties. This high percentage gripping explained by the fact that each suckers presented a root portion who absorbed water immediately. The mud from around the root and watering after planting provided an appropriate and sufficient humidity to maintain turgescences suckers and foster the growth of new roots.

Propagation by cuttings green at raspberries was made in greenhouse on substrate made of sand. The river sand assured a percentage of green cuttings rooting of 73%, the minimum being recorded for variety Polka (59.4%), and the maximum at variety Heritage (89.2%). Under the influence of hormone rooting, the percent of cuttings rooted increase to 90.2%.

At the simple variant to rooting by green cuttings only Heritage variety exceeded witness with 10.8%. Following the implementation of rooting substance, three varieties exceeded witness with percentages ranging from 0.5% (Noma) and 8.5% (Willamette).

At the all 3 varieties of blackberry without thorns was experimented multiplication by cuttings, minicuttings and eye minicuttings (uninodali). Were experimented two rooting substrates. The first substrate was represented by a mixture of sand and peat in equal proportions, and the second substrate was a mixture in equal proportions between river sand and perlite.

Cuttings had size 30 cm, 15 cm minicuttings and cuttings uninodali included one bud 2-3 cm long. Minicuttings were planted in rows at 5 x 4-5 cm (400-500 cuttings / m^2).

Although not reluctant to issue adventitious roots we studied the effect of Bioroots substance in rooting.The experiments were conducted on the bed rooting in greenhouse. Not being a pretentious species to temperature and substrate conditions we proceed to the growth in solar. The results have been very encouraging and may be extended to a larger scale. The small differences between the percentages of rooting recorded between the two substrates can be attributed to mixture fertility, the peat was mixed with a nutritive substance. Thus, the percentage of cuttings rooting in mixture from

sand and peat was in averaged 88.6% and 97.1 when applied Bioroots product. The same type of cuttings placed in a mixture of sand and perlite rooted in a proportion of 86.2% when the substrate containing no hormone rooting and 95.7% when applied Bioroots.

Between varieties are very small differences concerning the percentage of rooting.

Minicuttings were planted in the substrate and had a length of 15 cm. The substrate from sand and peat assured an average rooting of the three varieties of 84.2% and 88.9%. The substrate of sand and perlite assured rooting average etween 87.2% and 98.1% (when using rooting hormone). Given the ability of forming roots I had experimented willingness to form roots when it is shaped very short. For this experiment we used the same culture substrates, cuttings had only one node (one bud). Before planting the cuttings eyes were wetted and kept in polyetilene bags for 7 days at 24 - 26°C. In this time the eye cuttings have formed roots and only after that were planted.

Planting was done by placing the cuttings on surface rooting substrate, which was covered with a thin layer of the same substrate. I made watering and temperature and humidity was optimally ensured. When the plants have reached a height of about 5 cm were made first investigations on the percentage of rooting.

In this case did not apply rooting hormone and since the time of planting all eyes cuttings were root primordia were initiated. By this method on both substrates rooting, cuttings was rooted in a very high percentage from 98.7 to 100%. The substrate from sand and peat secured a 100% rooting percentage to variety Thorn Free and Silvan. This method allows rooting almost 100%, offering the highest yield.