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# SCIENTIFIC BASIS FOR PROPAGATION OF IMPROVED VINE SEEDLINGS UNDER *IN VITRO* CONDITION

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**ABSTRACT**
*This article outlines particular features of modern healthier biotechnological method of propagation of young vine plants under in vitro condition. As a nutrient media Murasiga Skuga has been chosen and meristem tissues of black Kishmish, Saperavi, pink Taifi have been passaged to it. It has been investigated that the viability of meristem tissues depends on that from which bud of scion they are taken. During the experiments it has been observed that the cells isolated from the second bud placed below the top bud of scion have grown well.*
**KEYWORDS:** *meristem, cell, vine, in vitro, passage, bud, variety, nutrient media, culture, micropropagation.*

## INTRODUCTION

For the purpose of meeting the demand for young vine plants in order to create new vineyards with them, to renovate old ones, to replace improper vine plants with new ones in current vineyards, it is expedient to develop seedling nursery base. Seedling production is performed by farms or private plot farms specialized in seedling growing. The base of cultivation of high-grade, standard, healthy and well-rooted young vines is considered creating of intensive nurseries for regionized and perspective grapevine varieties on the farms.

At present, one of effective ways of obtaining improved seedlings is clonal micropropagation of valuable varieties, that is, growing of extracted tissue and other parts of vine shoot in an artificial nutrient media under special condition (*in vitro*) [1, 2, 4].

Supportive super-intensive nurseries are created in protected or open areas with the seedlings grown under this method. Two types of micropropagation have been used in artificial media of special condition called *in vitro.*

The first way is called microclonal propagation, that is, extracting meristem tissue from the top node apical of grapevine plant and then sowing it. Under this method tissue is sown in liquid nutrient media, but explant vegetation period occurs slowly.

The second way of propagation is performed by removing bud from the shoot, sowing it and growing in special condition.

Cultivation of high-grade (high-categorized) seedlings by in vitro micropropagation culture requires the study and developing the most effective and economical methods of fast propagation of plants.

According to the scientific researches conducted in the countries with developed agriculture, the most effective principle for cultivation of fruit plants and grapevine plants is to produce qualitative, healthy seedlings.

Creating industrial orchards and vineyards on the base of these seedlings allows to well adaptation of plants to various ecological factors, early maturation and abundant yield regularly each year. The spent costs for this kind of vineyards with healthier seedlings are

more easily and quickly covered compared to common seedlings use. The last stage of seedling production which is free from virus, is a creation of nursery for first healthy generation of seedlings grown by *in vitro* method.

## MATERIALS AND METHODS

Inserting improved vine shoot (explant) into neutralized nutrient media (culture) through apical meristem consists of two stages: apical meristem part (tissue) of grapevine shoot is separated from mother plant, then sown in nutrient media. This process stages together with nutrient media content may lead to growth, similar to *in vivo* culture. Under this condition explant joint interval may increase and leaves grow, callus or adventive shoots get formed. For neutralization and growth of plant subapical meristem is extended. Microcutting of explants involves these processes: washing cuttings under flowing simple water during 1,5-2 hours; washing under neutral detergent solution (30-40 minutes); neutralize in the solution of thimerosal 0,01%; washing by 30% ethanol solution in laminar-box conditions (2-5 min); washing by sterilised water in laminar-box conditions (5-10 min) [3].

Neutralized explants are planted in the conditions that are not hormonic. They are grown within 48-72 hours for selecting healthy material. Explants that don't contain fungus and viral disease symptoms are transplanted into other nutrient media for regeneration.

## RESULTS AND DISCUSSION

As illuminated in experiments, these technical and well-consumable varieties of grapevine have presented inclination to microclonal propagation. When explants have been transplanted into neutralized condition, at the first day of inserting (passage) into nutrient condition their inclination to this propagation made 96-98% in technical varieties, 80-83% in well-consumable varieties.

The share of explants that were not able to further glassy grow consisted 10-12% in technical variety and around 17-20% in consumable variety. The quantity of explants that were able to form callus, adventive and simple growing shoots, was 1,3-1,4 times more, that is, relatively 86% and 60%. This indication confirms that technical varieties are much more inclined to effective microclonal propagation by *in vitro* method (Table 1).

Some factors such as the age of mother plant and bud removal according to the length of growing shoot can influence on regeneration features of explants.

**Table-1**

**Reaction of various varieties of grapevine explants to inserting to in vitro condition (Murasiga and Skuga), in 2017-2018**

| Grape varieties | Inclination to microclonal propagation, % | | |
|---|---|---|---|
| | live explants | rhizogenesis of explants | |
| | the first passage | glassy | callus |
| black Kishmish | 96 | 10 | 86 |
| Saperavi | 98 | 12 | 86 |
| pink Taifi | 83 | 17 | 66 |

**Explanation:** *content of Murasiga and Skuga condition = 6-benzylaminopurine of 0,3 mg/l, indolyl acid of 0,5 mg/l water.*

As it was identified in our researches that in spite of mother bush age, the second and third buds placed below half-wooden cane tip had high regeneration features in the propagation by *in vitro* method. The buds placed more down of the shoot had less of this feature. The highest inclination to micropropagation was observed in young plants at the most productive period (juvenile period of growth). Particularly, when the buds of the tip shoots of three-year old mother plant were used, the viability of explants consisted 45%, while the buds of below parts of shoots were used viability made 20-30%, in middle part – 47-65%.

The buds from older aged mother bushes (six year old) cause to reducing inclination of vine explants to micropropagation 1,8-2,0 times. General growth and development of grapevine seedlings grown by micropropagation method depend on the age of mother plant and the position of the used bud in the shoot. Consequently, in the best variant of experiment , the use of above-placed buds provided leaves formation of 6,4-7,1 mm length and 3,3-3,9 leaf pieces in annual growth range, while the used of below-placed buds allowed to decreasing of these indication as 6,3-6,8 mm and 2,8-3,0 pieces. When the explants of buds of old aged (six year old) plants were used this growth indication became much lower, by 1,4-1,6 times (Table 2).

**Table 2**
**The impact of the position of used bud in the shoot and the age of mother bush on regeneration feature of vine explants, 2017-2018**

| Bud position from the tip of shoot | Three year old mother plants | | | Six year old mother plants | | |
|---|---|---|---|---|---|---|
| | bud regeneration, % | leaf quantity, pcs | shoot length, mm | bud regeneration, % | leaf quantity, pcs | shoot length, mm |
| Tip | 45 | 2,8 | 6,8 | 23 | 1,8 | 4,3 |
| Second | 65 | 3,9 | 7,1 | 32 | 2,9 | 5,0 |
| Third | 47 | 3,3 | 6,4 | 24 | 2,2 | 4,3 |
| Forth | 30 | 3,0 | 6,3 | 16 | 2,0 | 4,2 |
| Fifth | 20 | 3,0 | 6,3 | 11 | 1,8 | 4,2 |

## CONCLUSION

When technical and well-consumable grape explants have been transplanted into Murasiga and Skuga nutrient media, at the first day of inserting (passage) into nutrient condition their inclination to this propagation was observed, the share of live explants made 86-88% in technical varieties, 80-83% in well-consumable varieties, while the explants that were not glassy viable made 10-12% and 17-20% relatively.

The quantity of explants that are able to form callus, further adentive and simple growing shoots is 1,3-1,4 times more in technical grape variety compared to well-consumed varieties, that is, relatively 86% and 60%. This indication confirms that technical varieties are much more inclined to effective microclonal propagation by *in vitro* method.

The bud explants of grapevine shoots have high regeneration feature and this is observed in juvenile growing period of mother plants (up to three years). At the productive period of mother plants this indication decreases by 1,9 times. Relatively general growing and developing process: leaf formation and shoot growing also decrease by 1,5 times.

The highest regeneration feature (to 65%) of grapevine varieties in micropropagation is observed in the buds that are taken from the middle part of three-year old mother plant's shoot. This indication shows 45% in the top part buds and 20-30% in down part buds. The more increases age of mother plants up to six years, the more decreases regeneration feature of buds, relatively to 33, 22 and 9%.

## REFERENCES


1. *Gauthere P., Cossio F. Risultati di osservazioni su un clone di Corvina Veronse (Vitis vinifera) moltiplicato attraverso la coltura "in vitro" – Riv. vitie. enol., 1932.-41.-p.110-117.*
2. *Haydu Z. In vitro cultures of grape Vitis sp.//International symposium plant tissue and cell culture application, the crop improvement. Olomouc, Chekhoslovakia, 24-29 September, 1984. -Prague: Chechoslovak Academy of Sciences, 1984. P.531-532.*
3. *Kutsenko A.A., Kulinsev L.G. Scientifically based preparation of improved transplants from valuable, native and perspective varieties of grapes by applying modern biotechnology. Methodological recommendation. - Mikhaylovskiy, 2011. P.8-20.*
4. *Zatyko Z.M., Simon I. In vitro culture of ovaries of Rubus species// Acta Agronom.cad.scien. hung. – 1975. –V.24. № 3/4. – p.277-281.*