Scientific Papers Series Management, Economic Engineering in Agriculture and Rural Development
Vol. 23, Issue 1, 2023
PRINT ISSN 2284-7995, E-ISSN 2285-3952

OBTAINING VITRO TUBERS OF WHITE AND PURPLE FLESH POTATOES IN ASEPTIC CULTURES OPERATED ON DOUBLE-PHASE TECHNIQUE

Alexandra-Mihaela NAGY[1,2], Monica POPA[1,3], Camelia SAVA SAND[1,2]

[1]University of Agricultural Sciences and Veterinary Medicine, Doctoral School of Agricultural Engineering, 3-5 Mănăștur, 400372, Cluj-Napoca, Romania, Mobile: +40749 4932 26, E-mail: alexandra-mihaela.nagy@usamvcluj.ro
[2]University „Lucian Blaga" of Sibiu, Faculty of Agricultural Science, Food Industry and Environmental Protection, 7 Dr. Ion Rațiu, 550012, Sibiu, Romania; E-mails: camelia.sava@ulbsibiu.ro; alexandra.nagy@ulbsibiu.ro
[3]National Institute of Research and Development for Potato and Sugar Beet, 2 Fundăturii, 500470, Brașov, Romania; E-mail: monica.popa@potato.ro

***Corresponding author*:** alexandra-mihaela.nagy@usamvcluj.ro; alexandra.nagy@ulbsibiu.ro

Abstract

Due to the interest shown in our country regarding the cultivation and marketing of purple flesh potatoes, we set ourselves the problem of examining the aspects that facilitate in vitro cultivation and obtaining micro tubers that ensure the faster micropropagation of purple potato varieties, formations that in modern food are also consumed raw. The paper aimed to present the protocol for obtaining vitro tubers or also called micro tubers from two varieties of potato: Christian with regular colour of flesh and Salad Blue with purple flesh. This protocol is based on micropropagation technique which combine an agar-solidified phase succeeded by a liquid phase in the same vessel in aseptic conditions. Using the double-phase technique gave the opportunity to reduce the costs generated by the utilities.

***Key words*:** *purple flesh potatoes, in vitro tuberization, double-phase culture medium, resources management*

INTRODUCTION

The *in vitro* cultivation of potato (*Solanum tuberosum* L.) is practiced in laboratories, not only for the purpose of carrying out scientific research, but also in an intensive regime for the delivery to agricultural farms of planting material, free of viruses, made by cultures of meristematic explants on aseptic culture media [7].

In modern nutrition, we currently consume vegetables and fruits rich in organic compounds synthesized in their secondary metabolism and which have an important and varied therapeutic influence in a series of medical treatments. This category also includes plants that synthesize anthocyanins [17].

Medicinal plants are used either fresh, respectively raw, or boiled like tea or preserved in various dried forms, in natural treatments [1].

As a rule, the compounds that fall into the category of secondary metabolism products are organic, made up of complex substances, which often define the characteristic particularities of some plant species; their location being in all plant organs, including reserve organs, but mainly in fruits, especially on blueberries and black grapes [5; 8]. This category also includes anthocyanins, compounds that stand out in various plant organs of some plants whose presence is distinguished by their red, purple or blue colour and whose colour changes when the pH varies [10], from a shade of red at an acidic pH, to purple at a pH between 6-8 and even blue at a basic pH.

The variety of these anthocyanin compounds exceeds the number of 500 varieties [21], their chemical structure being complex [6]. In potato varieties with purple flesh, the presence o six types of anthocyanins is confirmed, namely: cyanidin, petunidin, pelargonidin, delphinidin, peonidin and malvidin [13].

From a medical point of view, anthocyanins exert excellent antioxidant, antimicrobial, anticancer, antidiabetic, anti-inflammatory, antiproliferative effects [21], but also strengthen immunity, delay aging, facilitate cerebrovascular and cardiovascular circulation [4], alleviate obesity, prevent liver disorders [6], and other benefits. On the category of plants rich in anthocyanins, we can also mention purple fleshed potatoes (*Solanum tuberosum* L.), the Salad Blue variety [19], which come from Peru and are mainly grown in China, but also in many other Asian and European countries, including our country. In addition to the use of purple flesh potato tubers in the kitchen for cooking, they are also used in the food industry [4], as natural dyes.

Due to the interest shown in our country regarding the cultivation and marketing of these purple flesh potatoes, we set ourselves the problem of examining the aspects that facilitate their cultivation *in vitro* and obtaining micro tubers that ensure the faster micropropagation of purple flesh potato varieties, formations that in modern food are also consume in raw form [20].

On the experiment carried out by us in this paper, to obtain micro tubers *in vitro*, we used bud explants taken from potato tubers (*Solanum tuberosum* L.) belonging to the Christian variety (control), with white flesh tuber and the Salad Blue variety with purple flesh tuber, due to the presence of anthocyanins in their cells.

The aim of this study is to obtain vitro tubers, also called micro tubers, after initiation on a classical culture media Murashige & Skoog (1962) [12] and transferred the obtained plantlets to a double-phase culture medium.

The paper contains *material and methods* section which describe the plant material used for the experiment, succeeded by *in vitro* culture initiation for stock vegetal material and *in vitro* culture tuberization which is divided in two parts: the first *in vitro* growth of potato plantlets and the second *in vitro* tuberization induction. The second section of the article reflect the *results and discussions* of the current experiment and in the end are presented the *conclusions* of these research paper which contains the importance of purple flesh potato and micro tuberization of it.

MATERIALS AND METHODS

It is known that potato tubers are a staple food [3], especially in the cold months, during the wintertime, being kept cold, in dark spaces, such as cellars, conditions in which – towards spring – the ”eyes” present on tubers generate buds and shoots (Fig. 1 A and B).

Plant material

The plant material used consists of shoots from two disease-free varieties of potato (*Solanum tuberosum* L.), namely Christian (the control) and Salad Blue. Both varieties were obtained from the National Institute of Research and Development for Potato and Sugar Beet from Brașov.

Christian is a Romanian potato variety, developed by the National Institute of Research and Development for Potato and Sugar Beet from Brașov. This is semi-early variety, with oval tuber shapes, reddish epidermis, and yellow flesh, developing a thick bush with leaves [14].

Salad Blue is a variety originally from Scotland, preserved on the seed collection of the National Institute of Research and Development for Potato and Sugar Beet from Brașov. This is an early variety, with oval tuber shapes, bluish-purple epidermis, and purple flesh with white insertions [15].

In vitro culture initiation

From the apical area of the potato shoots from the studied varieties, after washing with sterile water, disinfecting them with 30% sodium hypochlorite solutions for 15 minutes and 3-4 time rinsing them with sterile water, upon removal of the apex coverings and proceeded to the ex-plantation of their apical zone – around 1-2 cm (Fig. 1 C (a)) – segment which then was inoculated, in a test tube with a volume of 10 ml solidified and aseptic Murashige & Skoog (1962) culture media, without growth regulators [13].

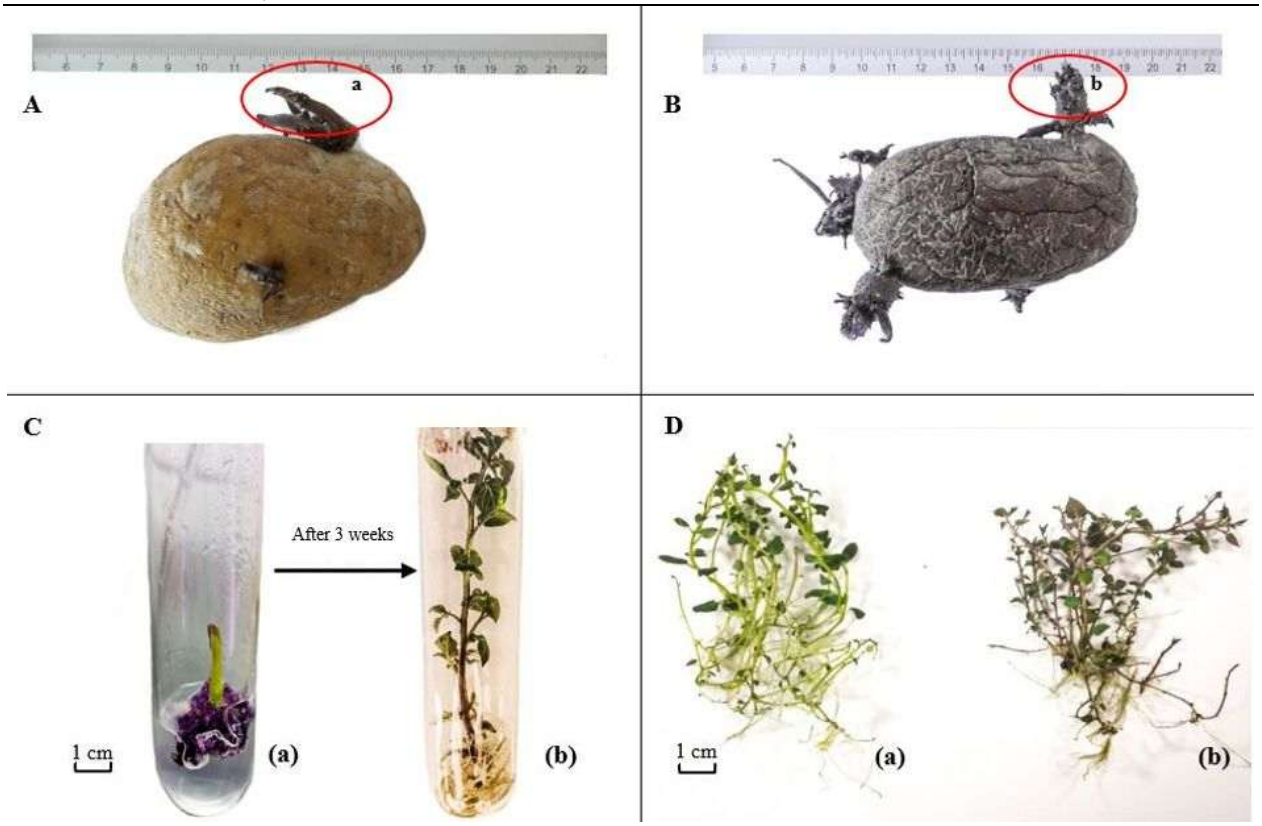

Fig. 1. The plant material used to initiate the potato *in vitro* culture; A – Christian potato variety tuber with white flesh (a – white buds); B – Buds of Salad Blue potato variety tuber with purple flesh (b – purple buds); C (a) – bud explants after 3 days of inoculation on solidified culture media; C (b) – The regeneration of potato plantlets from bud explants after 3 weeks from initiation; D – Potato ex vitro plants from variety Christian (a) and Salad Blue (b)
Source: Own determination.

These operations were carried out in a horizontal laminar flow hood of sterile air, the hood being in a sterile enclosure. The test tubes were all sealed.

After the completion of the inoculation operations, the samples (test tubes with explants) were transferred to the growth room, on shelves illuminated with neon tubes, with a photoperiod of 16 hours of light and 8 hours of darkness, at a temperature of 20°C ± 1°C.

***In vitro* culture tuberization**

From the first days after initiation, plantlets began to be generated from the inoculum, which within 3 weeks reached 7-8 cm in height (Fig. 1 C(b)).

The *in vitro* tuberization process involves two completions of two different culture phases (Table 1) on different grown condition (Table 2); first phase is the *in vitro* growth of potato plantlets succeeded by the second phase *in vitro* induction of tuberization.

Table 1. Culture media used for *in vitro* tuberization

Phase / *Elements*	1st phase: *In vitro* grown	2nd phase: *In vitro* tuberization induction
MS62	full	1/2
Sucrose	20 g/l	80 g/l
NAA	0.5 mg/l	-
Kinetin	-	0.5 mg/l
Coumarin	-	0.05 g/l
Agar	9 g/l	-

Source: Own calculation.

Table 2. Culture condition for *in vitro* tuberization

The process	Photoperiod		Temperature	Time for grown
	Light hours	Dark hours		
1st step: *In vitro* grown	16	8	20°C ± 1°C	3 weeks
2nd step: Tuberization induction *in vitro*	-	24	20°C ± 1°C	2 months

Source: Own calculation.

A. *In vitro* growth of potato plantlets

This phase aims at the optimal development of the plantlets that will later induce tuberization. These plantlets were transferred, also in aseptic mode, on solidified culture medium, MS62 supplemented with 0.5 mg/l α-naphthyl acetic acid (NAA), autoclaved at 121 ^{o}C for 20 minutes in glass containers and after that placed under sterile conditions, in the hood, in larger, single-use pots made of transparent and colorless plastic (14 cm long, 8 cm wide and 8 cm height), hermetically closed with a lid of the same material, with a green filter, from Duchefa. Each pot contains in the solidified and aseptic medium 10 potato cuttings, with a length of approximately 2 cm. Cuttings were obtained from plant material initiated in test tubes. After the cuttings transfer operation was completed, the pots were transferred to the growth room, with a photoperiod of 16 hours of light and 8 hours of darkness, for 3 weeks, in an aseptic regime.

B. *In vitro* tuberization induction

The next step consisted in inducing tuberization *in vitro* by pouring a 1.5 cm substrate of liquid culture medium (about 100 ml) without agar into the culture vessels, so that the potato plantlets were subjected to *in vitro* culture in „double layer" (Fig. 2 A and B). This process took place after about 3 weeks, at which time the plantlets present in the culture vessels reached a height of 7-8 cm. The agar-free liquid culture medium had the following composition: ½ MS62, 0.05 g/l coumarin dissolved in 2.5 ml ethyl alcohol, 0.5 mg/l kinetin and 80 g/l sucrose.

After adding the tuberization medium (in the hood), the pots were transferred again to the growth room in the dark for 2 months at a temperature of 20^{o}C ± 1^{o}C.

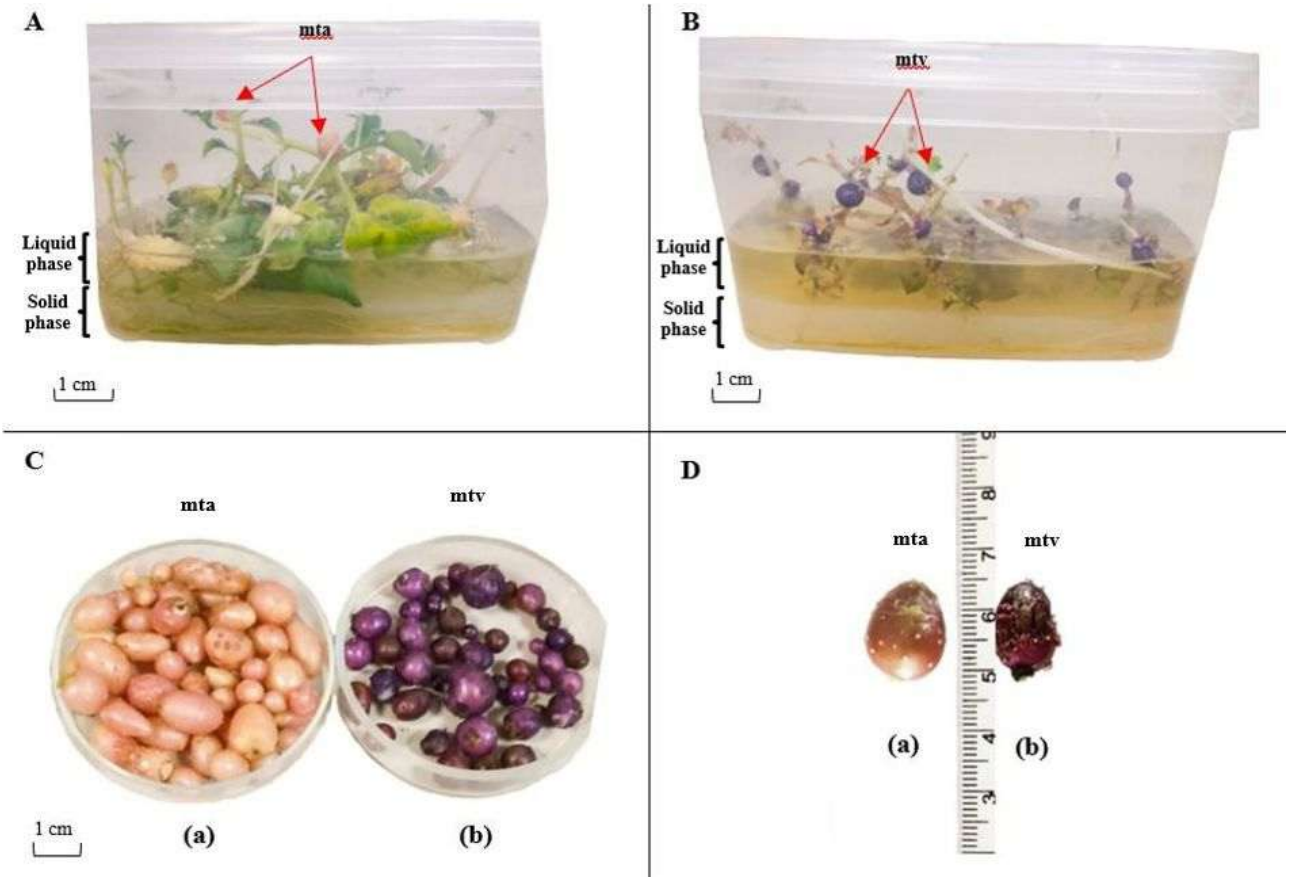

Fig.2. Appearance of Christian and Salad Blue potato micro tubers on „doble layer" culture media
A – Micro tubers of Christian variety on culture media (mta); B – Micro tubers of Salad Blue variety on culture media (mtv); C – Christian (a) and Salad Blue (b) micro tubers harvested from „double layer" culture media; D – The size of the Christian (a) and Salad Blue (b) micro tubers
Source: Own determination.

Experiment design and data analysis

For the present experiment, 10 culture vessels per potato variety were used, and the experiment had 3 repetitions. For each pot were used 10 potato micro-cuttings, resulting 100 micro-cuttings per variety in a single repetition and a total of 300 micro-cuttings per variety in the whole experiment.

For each culture vessel, the number of resulting micro tubers, their length and diameter were quantified, after which weighing's were also carried out for each individual micro tuber.

After collecting the data, they were processed using the Excel tool from the Microsoft Office package, and then interpreted with the Polifact statistical program, using the Duncan test.

RESULTS AND DISCUSSIONS

The research carried out in this work aimed to obtain potato micro tubers – white (as a control) and especially purple micro tubers – in a continuous flow, for the fact that this vegetable ensures the nutrition of the Earth`s population throughout the year [9], and the tubers of the purple potato varieties (such as the Salad Blue variety that we experimented with), contain anthocyanins (Fig. 2 C and D), thus consumers benefits from the exceptional effects that these by-products of metabolism expert on the human body, they can only be consumed raw [20].

After the tuberization period, the harvested micro tubers were disinfected with Domestos which contain as active substance sodium hypochlorite, at 20% solution concentration and subsequently washed and rinsed with sterile water, to remove traces of the culture media and to avoid further infections that may occur during their storage.

Then, the micro tubers are left to dry for 1-2 days at room`s temperature. They are placed in the refrigerator at 4-5^{o}C, in the dark after dry. Micro tubers can also be stored in the freezer until planting, but no more than one year. They can later be planted in "insect proof" greenhouses and/or tunnels in a substrate consisting of a mixture of red peat with bentonite, black peat, and perlite. About 3 months after planting, the harvesting of mini tubers begins, which will later be used for planting and obtaining potato tubers by farmers.

For each pot, the micro tubers obtained were counted. Thus, for the Christian variety the average of micro tubers was 10.33 per pot, and for the Salad Blue variety the average of micro tubers was 20.67 micro tubers per pot (Fig. 3). It follows from this that following the application of the Duncan test, statistically guaranteed differences were obtained, and the culture medium in the double layer favored the obtaining of micro tubers from the Salad Blue variety, a variety of great interest, due to the rich content of anthocyanins.

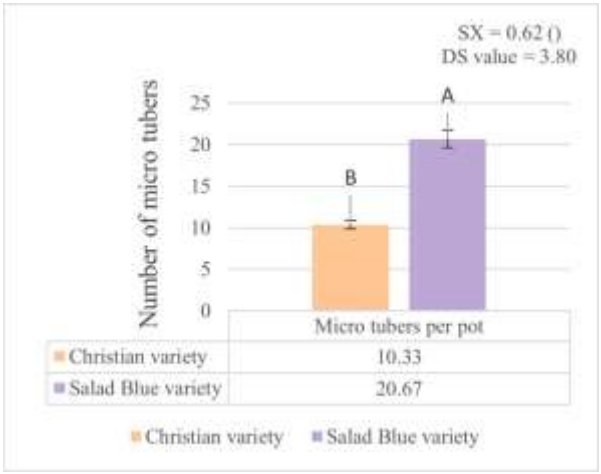

Fig.3. The potato variety influence for obtaining micro tubers on the „double layer" culture media
*A-B – significant statistical difference according Duncan test
Source: Own calculation.

According to the Duncan test, it follows that there were no statistically assured differences in weight (Fig. 4). Thus, the average weight for micro tubers obtained in double-layer culture, from the Christian variety was 0.92 g, while for those from the Salad Blue variety, the average weight was slightly higher, exactly 1.08 g, but the difference is insignificant.

Also, in the case of the length of the micro tubers, there were differences between the two varieties (Fig. 5); on the Salad Blue variety, micro tubers with a longer length (93 mm) were obtained, compared to those of the Christian variety (63 mm), and in this case, the differences not being significant from the statistical point of view.

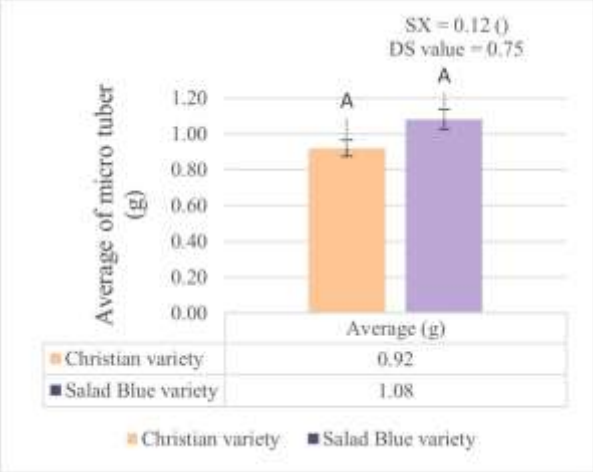

Fig. 4. The potato variety influence on weight of micro tubers obtained on the „double layer" culture media
*A-A – no significant statistical difference according Duncan test
Source: Own calculation.

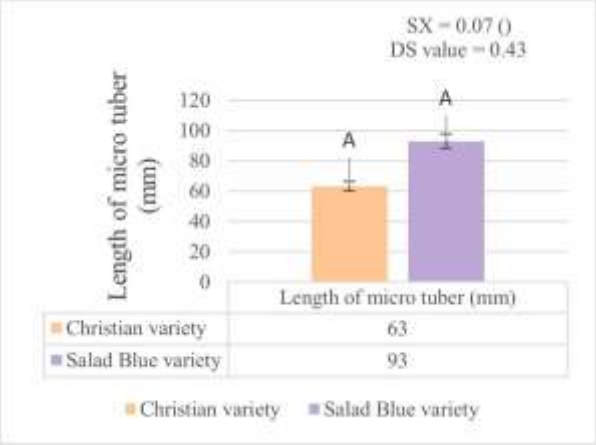

Fig. 5. The potato variety influence on micro tubers length obtained on the „double layer" culture media
*A-A – no significant statistical difference according Duncan test
Source: Own calculation.

Since the potato is propagated vegetatively [11], through tubers, and varieties with purple flesh tubers are still more difficult to access for European countries, and especially for the inhabitants of our country, we considered that the *in vitro* micropropagation procedures of the purple flesh potato are beneficial, with even more so as foods with anthocyanins increase immunity and mitigate the harmful consequences of many diseases.

In addition, we emphasize the advantages of purple flesh potato tubers consumption, respectively micro tubers produced by *in vitro* culture procedures, in a fresh state, without being cooked. Moreover, we are of the opinion that purple flesh potato ex vitro plants can also be eaten raw, in salads after are eating for decades in the European states bundles of seedlings from germinated seeds from different plant species such as red radishes, red cabbage, mustard, sunflower, onion, pepper, etc. (Fig. 6 A–E). Currently, they are called "sprouts" by traders (Fig. 6 A), being seedlings with cut roots (Fig. 6 B-E) sold in our country, following the model of other states.

The plants used for this purpose benefit from the fact that they contain meristems, which in their apexes hold stem cells, whose therapeutic and fortifying effects are highly valued. But seedlings from seed embryos (Fig. 6 A-E) have a short growth to the young seedling stage.

In contrast to the vitro plants, plants derived from seeds, respectively from zygotic embryos, cannot ensure the transmission of the characters of a specific genotype selected as a result of genetic engineering operations; instead, *in vitro* culture procedures ensure the continuous cloning of a certain genotype, as well as the preservation of these representatives in gene banks (in liquid nitrogen) and upon thawing of such plantlets, the continuous micropropagation of these genotypes is ensured.

A similar experiment was successfully carried out by Cachiță-Cosma and Zăpârțan (1991) [2], in which the pieces of potato micro tubers bearing buds were used in test tubes as plant material on *in vitro* culture process, on a two-layered aseptic substrate - agarized basal over which - after cooling - the inoculums and the liquid culture medium were placed, in a 2 cm layer. But, in the present work, the use - in stages of the bi-layered medium - of pots with a larger capacity, in which a volume of 100 ml of solid medium (1.5 cm layer height) and 100 ml of liquid medium (1.5 cm layer height) proved to be beneficial as it facilitated the implantation operation of the potato explants and favored both the growth of newly formed shoots at the phytoinoculs level and tuberization (Fig. 2 A-C).

Fig. 6. Seedlings from different plant species called „sprouts", of zygotic origin, without roots, sold and consumed raw in Romania; A – Closed pack with „sprouts"; B – Pack with „sprouts" of *Allium cepa*; C – Pack with „sprouts" of *Helianthus annuus*; D – Pack with „sprouts" of *Brassica oleracea*; E – Pack with „sprouts" of *Raphanus sativus* Source: Own determination.

At the same time, we would like to underline the fact that micro tubers, but also potato plantlet shoots, resulting from *in vitro* cultures, as well as plantlets resulting from the seeds of other species germinated under aseptic regime, should be integrated within the terminology integrated into the gemmotherapy compendium (respectively of meristem therapy), field introduced in 1991, in naturist therapy procedures [16].

Such biological material - especially purple micro tubers - contain anthocyanins (valuable by-products of metabolism, with recognized therapeutic effects), which during autumn, winter and the first months of spring are absent in nature, and the products used in dry form do not they still have a therapeutic value like that of living organs.

In addition, the shoots, and plantlets that we have all year round in a fresh state contain not only meristems, but also stem cells, tissues with strong therapeutic and antioxidant value.

In the other hand, periodically, the vitro-cultured biological material can be fragmented - micro tubers or stalks - into mini-cuttings which - for a period of time - can be sub-cultivated, possibly even in the culture substrate from which they came, until the compounds it contained are exhausted, provided that the regime of guaranteed asepsis of the operations is ensured. Such vitro plants can be fragmented into uninodal micro cuttings that can be reinoculated *in vitro* and give rise to new vitro plants in continuous flow. Such procedures would expand the methods of obtaining new varieties of natural medical treatments, such as meristem therapy or gemmotherapy, which are modern natural medicine methodologies [18]. A diversification of gemmotherapy procedures can also be achieved by consuming purple micro tubers (Fig. 2 C) that hold in their reserve parenchyma cells, a large amount of anthocyanins. Such biomass, in modern biotechnologies, could be obtained in bioreactors, in continuous flow, and the procedure can serve to extract and capitalize the respective anthocyanins in bioindustry and in commerce, a procedure carried out in continuous flow.

CONCLUSIONS

In vitro tuberization in potatoes species, especially on the purple flesh variety Salad Blue, from explants sized at a waist of about 2 cm, taken from the buds formed on the surface of mature tubers kept in the cool and dark condition, which were inoculated on the culture medium solidified, and after that subcultured on a double-layer culture medium, with the addition of phytohormones (NAA in the solid medium, and kinetin, to which coumarin is added, reducing the MS

concentration to ½ and increasing the sucrose concentration in the liquid substrate), placed in pots made of colorless and transparent plastic. The culture pots were kept in the dark for 2 months after the liquid medium substrate was added. This procedure facilitates the management of energy resources, by saving electricity. Through this procedure, we managed to obtain an average of 21 micro tubers of a size about 90 mm from the Salad Blue variety, while in the Christian variety, we obtained an average of 10 micro tubers of a size about 60 mm.

The novelty of the experiment consists in the use of larger culture vessels/pots, in which the vitro plants regenerated from the sprouted buds on normal tubers, kept in the dark initially on agar medium and later in a double layer regime (the first agar medium layer and then the plantlets of about 7-8 cm were flooded with the second layer of the liquid culture media). If we aim to obtain seed material, these micro tubers can be planted in "insect proof" greenhouses and/or greenhouses in a substrate consisting of a mixture of red peat with bentonite, black peat and perlite. About 3 months after planting, the harvesting of mini tubers begins, which will later be used for planting and obtaining potato tubers. Another aspect of the originality of the present paper consists in suggesting the use of purple flesh potato plantlets, the Salad Blue variety, of about 3-10 cm, in a fresh (raw) state, as a natural food (as the seedlings generated from the embryos are used in nutrition seeds), as the ex vitro plants are rich in stem cells, and the anthocyanins held in the tissue cells of the potato vitro plants and especially in the reserve parenchyma of the purple potato micro tubers are very beneficial in strengthening the human body.

Obtaining purple micro tubers in bioreactors could constitute usable biomass in the production of anthocyanin extracts, marketable and usable in naturopathic medicine.

ACKNOWLEDGEMENTS

This research work was carried out with the support of National Institute of Research and Development for Potato and Sugar Beet from Braşov, during the PhD training program of the first author.

Also, we would like to thankfully to PhD. Dorina Cachiţă-Cosma for her support in this research paper.

REFERENCES

[1]Alamgir, A. N. M., 2017, Herbal drugs: their collection, preservation, and preparation; evaluation, quality control, and standardization of herbal drugs. Therapeutic Use of Medicinal Plants and Their Extracts: Volume 1: Pharmacognosy, 453-495.

[2]Cachiţa-Cosma, D., Zăpârţan, M., 1991, Potato tuberogenesis using in vitro bi-layer tehnique. In The IVth National Symposium on plant cel land Tissue Culture (pp. 7-9).

[3]Devaux, A., Goffart, J. P., Kromann, P., Andrade-Piedra, J., Polar, V., Hareau, G., 2021, The potato of the future: opportunities and challenges in sustainable agri-food systems. Potato Research, 64(4), 681-720.

[4]Gong, S., Yang, C., Zhang, J., Yu, Y., Gu, X., Li, W., Wang, Z., 2021, Study on the interaction mechanism of purple potato anthocyanins with casein and whey protein. Food Hydrocolloids, 111, 106223.

[5]Gordillo, B., Sigurdson, G. T., Lao, F., González-Miret, M. L., Heredia, F. J., Giusti, M. M., 2018, Assessment of the color modulation and stability of naturally copigmented anthocyanin-grape colorants with different levels of purification. Food research international, 106, 791-799.

[6]Huang, Y., Zhou, S., Zhao, G., Ye, F., 2021, Destabilisation and stabilisation of anthocyanins in purple-fleshed sweet potatoes: A review. Trends in Food Science & Technology, 116, 1141-1154.

[7]Hussey, G., Stacey, N. J., 1981, In vitro propagation of potato (Solanum tuberosum L.). Annals of Botany, 48(6), 787-796.

[8]Kalt, W., Cassidy, A., Howard, L. R., Krikorian, R., Stull, A. J., Tremblay, F., Zamora-Ros, R., 2020, Recent research on the health benefits of blueberries and their anthocyanins. Advances in Nutrition, 11(2), 224-236.

[9]Lachman, J., Hamouz, K., Šulc, M., Orsák, M., Pivec, V., Hejtmánková, A., ... Čepl, J., 2009, Cultivar differences of total anthocyanins and anthocyanidins in red and purple-fleshed potatoes and their relation to antioxidant activity. Food chemistry, 114(3), 836-843.

[10]Mattioli, R., Francioso, A., Mosca, L., Silva, P., 2020, Anthocyanins: A comprehensive review of their chemical properties and health effects on cardiovascular and neurodegenerative diseases. Molecules, 25(17), 3809.

[11]Mohapatra, P. P., Batra, V. K., 2017, Tissue culture of potato (Solanum tuberosum L.): A review. International Journal of Current Microbiology and Applied Sciences, 6(4), 489-495.

[12]Murashige, T., Skoog, F., 1962, A revised medium for rapid growth and bioassays with tobacco tissue cultures. Physiologia plantarum, 15(3), 473-497.
[13]Nagy, A. M., Boboc (Oros), P., Cătană, C., Antofie, M. M., Sava Sand, C., 2022, Comparative study of two varieties of purple flash potato grown in vitro. Scientific Bulletin. Series F. Biotechnologies, 26(1), 43-48.
[14]Nistor, A., Campeanu, G., Atanasiu, N., Chiru, N., Karacsonyi, D., 2010, Influence of potato genotypes on "in vitro" production of microtubers. Romanian Biotechnological Letters, 15(3), 5317-5324.
[15]Pęksa, A., Kita, A., Kułakowska, K., Aniołowska, M., Hamouz, K., Nemś, A., 2013, The quality of protein of coloured fleshed potatoes. Food Chemistry, 141(3), 2960-2966.
[16]Piterà, F., 2000, Compendio di gemoterapia clinica (Meristemoterapia) con indice clinic. Genova.
[17]Pojer, E., Mattivi, F., Johnson, D., Stockley, C. S., 2013, The case for anthocyanin consumption to promote human health: a review. Comprehensive reviews in food science and food safety, 12(5), 483-508.
[18]Soescu, S., Nițu, S., Ponoran, C., Olah, N., 2008, Gemoterapia de la A la Z. Eikon, 2008.
[19]Vaitkevičienė, N., Kulaitienė, J., Jarienė, E., Levickienė, D., Danillčenko, H., Średnicka-Tober, D., ... Hallmann, E., 2020, Characterization of bioactive compounds in colored potato (Solanum tuberosum L.) cultivars grown with conventional, organic, and biodynamic methods. Sustainability, 12(7), 2701.
[20]Xu, D., Liu, H., Jin, C. Y., Cao, C. M., Li, W. G., Zeng, F. K., ... Liu, G., 2018, A new potato variety grown in China suitable for raw eating. European Food Research and Technology, 244(5), 851-860.
[21]Yong, H., Liu, J., 2020, Recent advances in the preparation, physical and functional properties, and applications of anthocyanins-based active and intelligent packaging films. Food Packaging and Shelf Life, 26, 100550.