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EFFECT OF NUTRITIVE MEDIA ON *SOLANACEAE* SP. REPRODUCTION *IN VITRO*

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Abstract

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It has been researched the possibility for *in vitro N. tabacum* sp., *S. tuberosum* sp., *S. lycopersicum* sp. Cultivars at a different nutritive mediums. A positive effect of liquid nutrient media on micropropagation *in vitro* in three Solanaceae species, compared to solid agar medium was established. The best for micropropagation *in vitro* in the varieties of three species - *N. tabacum, S. tuberosum* and *S. lycopersicum* is nutritive medium B. The importance of genotypein the reproduction *in vitro* has been demonstrated. The unified systeme for micropropagation *in vitro* - based on nutritive medium B of varieties of three family *Solanaceae* species was created.

Key words: nutritive media, in vitro reproduction, cultivars

Introduction

Growth and development of plants in vitro is determined by the following factors - genotype, nutrititive medium, physical factors - light, temperature, pH, concentration of O_2 and CO_2, organic substances, phytohormones, etc. The culture medium is essential for growth and development of plants (Annela and Kahn, 2010).

For gelling of the medium in the *in vitro* cultivation is most commonly used agar. It is a polysaccharide of high molecular weight and high gelling ability. It is important to know that the gelling agent is the most expensive component in solid nutrient media (Murashige and Skoog, 1962). There are studies on the influence of agar on the development of plants in vitro conditions. It was found that low concentrations of agar facilitate rooting of apple (Werner and Boc, 1980). The literature indicated to increase stem cell proliferation from the standpoint of the physical condition of the medium with agar concentration of 0.48 to 1% (Lundergan and Janick, 1980; Kassim, and al., 2010). It should be borne in mind that it contains organic and inorganic residues (Teixeira da Silva, et al., 2005).

Therefore, some authors recommend the use of liquid culture media. For example in *N. tabacum* (cv. Xanti) growth and proliferation increase in the use of liquid culture medium (Stevenson et al., 1982). The same is recommended in planting some trees (Pieik, 1987; Kassim et al., 2010) and vine (Dimanov et al., 2001).

Similar findings have been established in our previous studies (Dimitrova and et al., 1992).

The aim of this study is to trace the growth and development of varieties of tobacco, potatoes and tomatoes - representatives of the family Solanaceae cultivated in vitro in medium MS (Murashige and Skoog, 1962) and medium B (Dimanov and Atanasov, 1986) in two versions - solid (agar) and liquid.

Materials and Methods

Attempts are reproduced in laboratories of plant tissue cultures, with TTPI - Markovo and PGRI- Sadovo.

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For source material used plant explants - cultivars by Tobacco *(N. tabacum*): Virginia 89, N. alata, Krumovgrad 90 and line Ri F_{10} (Plovdiv 7 x L.2); Potatoes (*S. tuberosum*): Vivaks, Bobar, Estima, Maranka, Ponto and Nela; Tomatoes (*S. licopersicum*): Monalbo, P 2046, l 1104.

Nutritive mediums: MS - basic 5 mg / l IBA; Medium B - (Dimanov, 1987) – from microelements in Heller (1953) is removed AlCl3, sucrose is increased to 20g / l. Contains 0,1 mg / l IAA

Biometric indicators: Dynamics of growth - root system and stems were reported in 10 days in 1 month. Plant explants were plated in tubes with 10 ml of culture medium at 10 plants in four replications. The material is placed in a growth-chamber at 25-27^{0}C, photoperiod 16/8 h light / darkness, light 3000 lux and 75% humidity.

Results and Discussion

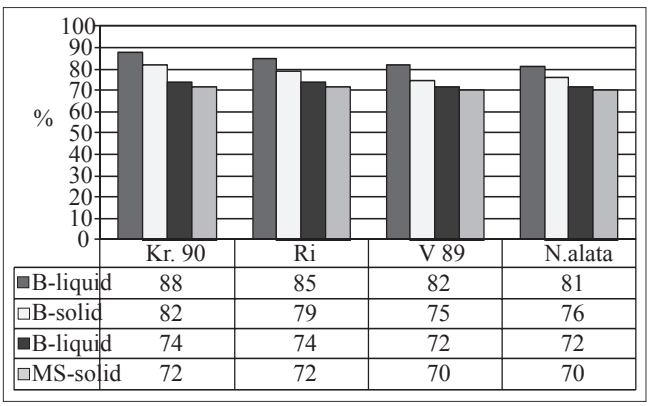
Induction of rizogenez *in vitro* and the rate at which the process goes, it is essential for overall development of plants. The cultivars rooting of study species and varieties depend on the conditions of cultivation (mentioned in Materials and methods, as it is essential from composition of the medium). In tracing the reaction of the varieties of *N. tabacum* in the initial period of cultivation experienced some varietal differences in the versions of the medium. The results presents in Figure 1 shows that most favorable to the rooting medium is B - liquid. The average percentage of rooted plants is 84%. Varietal differences were observed in the range – 88% for variety Krumovgrad 90 and 81% in explants from *N. alata*. Both versions of the medium B – liquid and solid, rooting percentage was significantly higher than the corresponding for MS. The difference between the B-liquid (MS liquid is 6.25% and 4.75% for B-solid) MS solid.

In potato varieties (*S.tuberosum*), the results presented in Figure 2 the influence of medium on the process of rooting were observed following varietal differences. The lowest percentage of rooting in Vivaks and Ponto varieties in medium MS - respectively 45 - 70% for liquid and 45-62% for the solid phase of the medium. The highest percentage of rooted plants is characterized by variety Bobar, as the best conditions for development of root system provides on medium B (liquid and solid) where the performance of 100% rooted explants.

The differences that occur between the variants tested on the respective phases - liquid (liquid and solid) solid c in the both mediums in *S. tuberosum* varieties are significantly B (MS-liquid – 13.28%) and B (MS - solid-12.15%). The results show that the most favorable in terms of rooting medium is in B-liquid, where the percentage rooted plants move in the range 90% - Vivaks and Ropta and 100% for all other varieties.

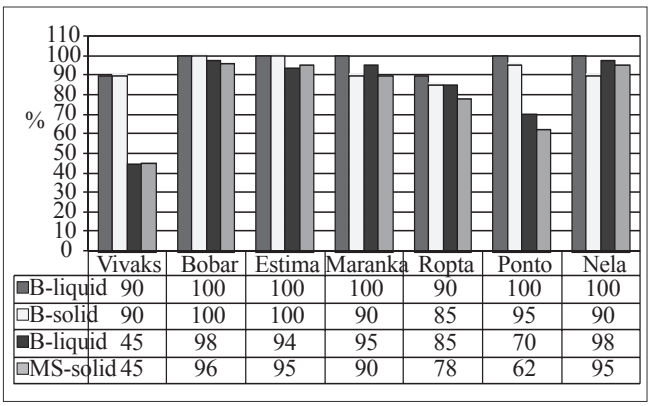
In the third representative of the family *Solanaceae*, *S. lycopersicum,* maintaining our established trend. Despite the low rate of rizogenez in all variants of the two mediums and relatively small varietal differences in medium B-liquid proved most favorable (Figure 3).

The results of comparison of both liquid and solid media are both very close to data *N. tabacum* -B liquid



	Kr. 90	Ri	V 89	N.alata
B-liquid	88	85	82	81
B-solid	82	79	75	76
B-liquid	74	74	72	72
MS-solid	72	72	70	70

Fig. 1. Effect on the nutritive medium at the rizogeneziz (%), *N. tabacum*



	Vivaks	Bobar	Estima	Maranka	Ropta	Ponto	Nela
B-liquid	90	100	100	100	90	100	100
B-solid	90	100	100	90	85	95	90
B-liquid	45	98	94	95	85	70	98
MS-solid	45	96	95	90	78	62	95

Fig. 2. Effect on the nutritive medium at the rizogeneziz (%), *S. tuberosum*

/ MS liquid – 6.25%; *S. lycopersicum* - B liquid (MS liquid - 6.66% and *N. tabacum* – B solid) MS solid – 4.75%, *S. lycopersicum* - B-solid (MS solid - 5.00%).

In conclusion we can summarizep that we studied species of the family *Solanaceae*, B liquid medium is most suitable for process, rizogenez and vary between 70,66% (*S. lycopersicum*) to 97.14 % (*S. tuberosum*). Proliferation of the stem depends on several factors - good rizogenez, balanced nutritional medium providing sufficient nutrients and phytohormones for the normal growth of the stem. The survey this indicator is presented in cm, as readings are for 30-day period.

The growth of the stem in all variants - varieties and growing media began almost simultaneously. In the initial period of 5 -15 days visible differences in growth, stems in both medias are not observed. Between 15-30 days of setting the explants outlines differences not only in terms of culture medium, but also in response to the variety.

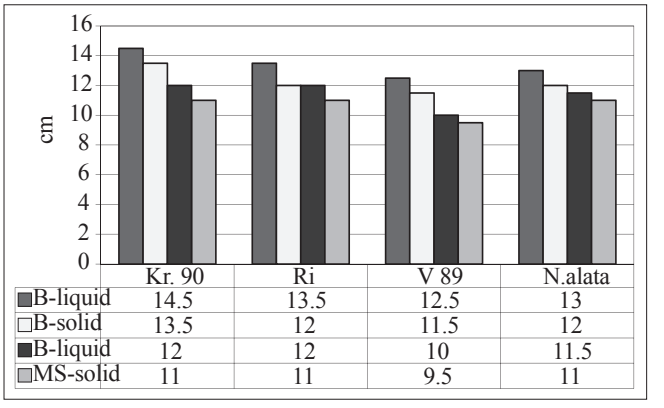
In Figure 4 is presented the influence of medium on the growth of the stem of varieties of *N. tabacum*.

When comparing the two culture medias MS (liquid and solid) and B (liquid and solid) can clearly faster growth on medium B. The average height of the stems by genotype ranged from 14.5 to 12.5 cm for varieties Krumovgrad 90 and Virginia 89 in B - liquid to 11.0 and 9.5 cm in MS-solid medium for the same varieties. For the other two options are similar results. Best proliferation of explants is recorded on the B -liquid medium.

The average height of the stems of potato varieties studied *(S. tuberosum*) is presented in Figure 5. After 15 day of explants, plating is account significant differences not only in terms of medium, but significant differences in responses of different varieties.

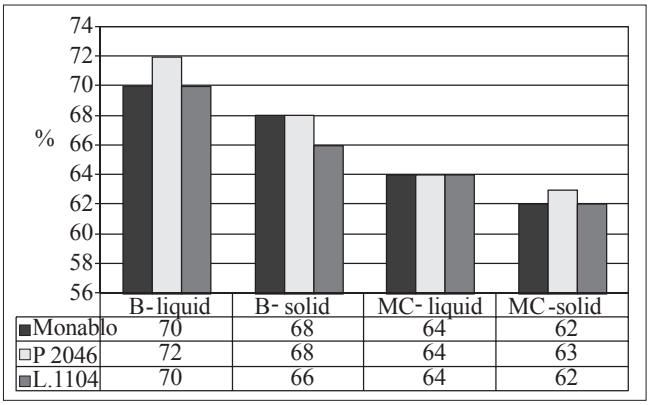
The average height of the stem varies from 7.0 cm at Nela variety of medium MS – solid to 18.0 cm in Estima, Bobar and Maranka in medim B-liquid. In all varieties, stem growth in liquid media B exceeds that of explants grown on solid medium. Although some authors recommend the use of liquid culture media **(**Stevenson et al., 1982 and Pierik, 1987) in Bobar, Estima, Maranka, Ropta and Ponto varieties explants cultured on solid medium B are in greater length than in MS-liquid. When comparing the two culture medias MS (liquid and solid) and B (liquid and solid) can clearly faster growth on medium B.

The results of this biometric indicator of tomato varieties (S. *lycopersicu)* are presented in Figure 6. Com-



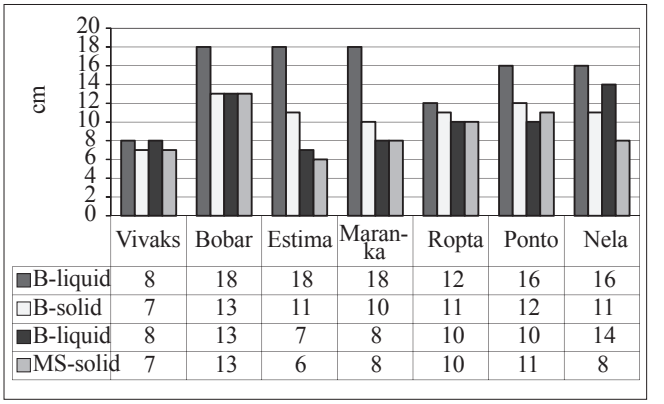
	Kr. 90	Ri	V 89	N.alata
B-liquid	14.5	13.5	12.5	13
B-solid	13.5	12	11.5	12
B-liquid	12	12	10	11.5
MS-solid	11	11	9.5	11

Fig. 4. Effect on the nutritive medium at the height on stem proliferation (cm), *N. tabacum*



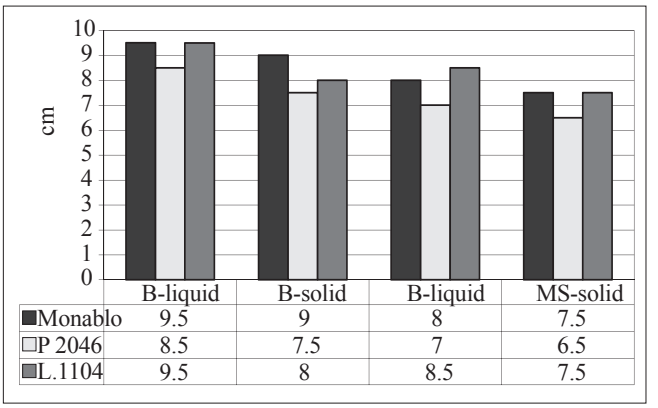
	B- liquid	B- solid	MC- liquid	MC-solid
Monablo	70	68	64	62
P 2046	72	68	64	63
L.1104	70	66	64	62

Fig. 3. Effect on the nutritive medium at the rizogeneziz (%), *L. esculentum*



	Vivaks	Bobar	Estima	Maran-ka	Ropta	Ponto	Nela
B-liquid	8	18	18	18	12	16	16
B-solid	7	13	11	10	11	12	11
B-liquid	8	13	7	8	10	10	14
MS-solid	7	13	6	8	10	11	8

Fig. 5. Effect on the nutritive medium at the height on stem proliferation (cm), *S. tuberosum*



	B-liquid	B-solid	B-liquid	MS-solid
Monablo	9.5	9	8	7.5
P 2046	8.5	7.5	7	6.5
L.1104	9.5	8	8.5	7.5

Fig. 6. Effect on the nutritive medium at the height on stem proliferation (cm), *L. esculentum*

pared with the variety differences resulting from other members of the family *Solanaceae* - tobacco and potatoes, of the stem length indicator in the options-varieties of tomatoes are less pronounced. When comparing the two culture media B and MS (liquids and solids) have a faster growth rate on medium B. Depending on the genotype variation in height was 9.5 cm for the first and third variety, and 8.5 cm for P 2046 in medium B-liquid, and 7.5 cm and 6.5 cm on medium MS-solid. Despite the slower growth of the stem with representatives of *S. lycopersicum*, confirmed our results found for tobacco and potato varieties that B -liquid media provides optimal conditions for expression of this indicator.

Conclusions

A positive effect of liquid nutrient media on micropropagation *in vitro* in three *Solanaceae* species, compared to solid agar medium was established. The best for micropropagation *in vitro* in the varieties of three species - *N. tabacum*, *S. tuberosum* and *S. lycopersicum* is nutritive medium B. The importance of genotype in the reproduction in vitro has been demonstrated.

The unified systeme for micropropagation *in vitro* - based on nutritive medium B of varieties of three family *Solanaceae* species was created.

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