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EXPERIMENTING HYDROPONIC CULTURE SYSTEMS ON DIFFERENT SUBSTRATES TO OBTAIN POTATO MINITUBERS

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ABSTRACT

The production of minitubers using the hydroponic method has become an important component in the production of seed potatoes in developed countries. The objective of the study is obtaining minitubers (Prebase), free from diseases, from the transplantation of vitroplants on industrial substrate and different hydroponic systems. The hydroponic culture involves growing plants on artificial substrates, or in a sterile, porous substrate with a high-water permeability. The parameters analyzed in this study were: average number of minitubers / plant and average weight of minitubers/plant. By using hydroponic system, a high minitubes production was obtained. The trifactor experience of the type 4 x 2 x 2 with 16 variants used the following graduations of the studied factors: experimental factor a, variety, with four graduations: a_1 - Braşovia; a_2 - Castrum; a_3 - Marvis; a_4 - Sarmis; experimental factor b, culture substrate, with two graduations: b_1 - expanded clay; b_2 - perlite; experimental factor c, the hydroponic system: c_1 - Wilma; c_2 - Nutrient Film Technique. These variants were experimented in 3 repetitions. Castrum variety was remarked by obtaining a high number of minitubers and also by weight of minitubers. Nutrient Film Technique hydroponic system had a positive influence on minituberization, for minitubers number, but also for weight of minitubers. Perlite substrate has determined obtaining high values for both studied parameters.

Key words: hydroponic system, nutrient solution, electroconductivity, perlite, expanded clay, minitubers.

INTRODUCTION

Potato is the third most important food crop in the world after rice and wheat in terms of human consumption. More than a billion people worldwide eat potato, and global total crop production exceeds 300 million metric tons (International Potato Center, CIP, http://www.cipotato.org/potato).

The production of minitubers by using the hydroponic method has become an important component in production of seed potatoes in developed countries. The introduction of this technology has reduced seed potato production period and diminished exposure of seed tubers to pathogens, thus improving their quality (Yang, 2004).

The current seed potato production program is based on the rapid multiplication method combining the procedures in laboratory, in "insect-proof" protected space and the field. This involves obtaining plantlets as a starting point in the seed potato production program followed by a second multiplication in the "insect-proof" protected space. In the last ten years, hydroponic culture technology was developed globally, thanks to the growing population in urban areas, thus being an opportunity for food production (Anda and Shear, 2017).

For potato culture it is essential to use seed tubers of high quality for obtain high yield production. Increasing the multiplication factor and producing good quality biological material have as result obtaining of valuable production.

The procedure used in hydroponic culture is to grow plants on different culture substrates (but not soil) and to apply nutritional solutions by using a complex system that automatically distributes liquid nutrients.

Minitubers production is the traditional intermediate step to make possible the use of *in vitro* plant material in the clonal field (Rolot et al., 2002). Hydroponics using for minitubers potato production is considered an alternative method to the traditional one consisting of a mixture of peat and sand. The main objectives are to ensure increasing production quality and reducing production costs (Rolot et al., 2002). Hydroponic

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systems can be used to minimize water loss (Jegathees, 1999) and to ensure a constant nutrition and irrigation.

The first studies in the production of potato minitubers by hydroponics included those of Hoagland, who in 1950 conceived a production system and nutrient solution composition. Boersig and Wagner (1988, quoted by Rolot et al., 2002) used hydroponic (NFT) and aeroponic (ARM) systems for minitubers production. Wheeler et al. (1990, quoted by Rolot et al., 2002), in a NASA study, showed that NFT technique was used to produce potato minitubers. Muro et al. (1997, quoted by Rolot et al., 2002) compared a traditional production system using a substrate composed of peat and sand with a hydroponic technique using perlite as a substrate and various types of nutritional solutions.

All culture conditions are controlled (electroconductivity, pH, temperature, viral, fungal, infections, nematode or aphids). To protect the inside space against the ingression of aphids and the transmission of viruses, the entrances are provided with porous insulators for ventilation. In hydroponic culture systems it is essential to maintain a balance between the nutrient ratios, the electrical conductivity value (EC) and the pH required for potato culture. In hydroponic systems, the applied method for adjusting the nutrient supply in relation with their needs is the measurement of the total ionic concentration of the solution in the root area, respectively the electroconductivity (EC) (Jegathees, 1999). For hydroponic minitubers production, the electroconductivity (EC) of the nutrient solution used is in the range of 2.0-2.5 dS/ m. As the nutrient solution is more concentrated, the electroconductivity (EC) is higher, limiting the surface development of foliage and water consumption (Novella et al., 2008). Ensuring optimal pH or at least keeping it within acceptable limits contributes greatly to achieving high levels of production and it should be between 5.8 and 6.5.

The goal of the study was researching the behaviour of the varieties on two different industrial substrates and two hydroponic culture systems. The objective was obtaining minitubers (Prebase), free from diseases, from the transplantation of vitroplants on industrial substrate and different hydroponic systems.

MATERIAL AND METHODS

Plants belonging to the Brasovia, Castrum, Marvis, and Sarmis cultivars were obtained by the rapid multiplication method, starting with the meristem culture, to obtain a biologically virus free material. After 6-8 months from inoculation of meristems on Murashige-Skoog medium supplemented with vitamins, plantlets develop; these are tested by applying the DAS-ELISA technique (Murashige and Skoog, 1962). Healthy plants are multiplied at each internode, and the minicuttings obtained are inoculated on the Murashige-Skoog nutrient medium for the development of new plantlest needed for planting. Multiplication of potato plantlets by *in vitro* technique is the main technique for obtaining virus free biologic material from a valuable but infected plant stock. Plantlets obtained *in vitro* benefit from specific experimental conditions in the growth room; sterilization of test tubes is made in the oven at 180^0C and the culture medium which is put in test tubes is sterilized in an autoclave at 120^0C. The plantlets are then transferred for acclimatization to the protected area of the Vegetable Tissue Culture Laboratory, NIRDPSB Brasov, Romania, followed by planting on industrial substrates. The "insect - proof" area must be equipped with a double entry, to be providded with disposable footwear at the entrance.

The hydroponic systems were prepared where the varieties were studied. The nutritional solution was prepared from the product "Universol", which was followed in the first stage by the higher concentration of nitrogen, then in the second stage, the higher concentration of phosphorus and potassium, respecting the concentrations specified in technical prospectus of the product, in accordance with the needs of plants in N, P_2O_5, K_2O, MgO, microelements, namely: Yellow Universol (Table 1) 12 + 30 + 12 + 2 MgO + microelements (in concentrations of

0.5-1.0 g/ l); Violet Universol 9 + 9 + 27 + 3 MgO + microelements (in concentrations of 0.5-1.5 g/ l). The electroconductivity of the solution was maintained at 2.2 Siemens/cm by adding the required substances in the concentration corresponding to the solution type. The electroconductivity of the solution was weekly analysed. The nutritional solution was distributed in 12-hour cycles, from 8 AM to 8 PM. The planting was made on June 12, 2017 and minituberes were harvested on October 9, 2017. The nutritional solution was modified, after one month of culture, by decreasing in nitrogen, and increasing phosphorus, to stimulate minituberization. The pH of the solution was maintained at 5.8 by addition of 1N hydrochloric acid or 1N potassium hydroxide. For each new prepared solution, the pH was checked as well as the electroconductivity (EC).

Table 1. The chemical composition of fertilizers "Universol" (after the Scotts technical prospectus)

%, N, P, K, Mg / Nutritive elements	Yellow Universol	Violet Universol
	12+30+12+2	9+9+27+3
Total nitrogen (N):	12	9
- of which nitric N	8.8	2.4
- of which ammoniacal N	3.2	6.6
Phosphate (P_2O_5)	30	9
Potassium (K_2O)	12	27
Magnesium (MgO)	2.5	3.3
Iron (Fe) EDTA	0.06	0.06
Boron (B)	0.01	0.01
Copper (Cu) EDTA	0.01	0.01
Manganese (Mn) EDTA	0.04	0.04
Molybdenum (Mo)	0.001	0.001
Zinc (Zn) EDTA	0.01	0.01
Electroconductivity (EC) for solution with 1 g / l at 21^0C	1.2 mS/cm^2	1.3 mS/cm^2
Maximum solubility in grams/liter	250	300

The trifactorial experiment of the type 4 x 2 x 2 with 16 variants (Table 2) included the following graduations of the studied factors:

– experimental factor a, variety, with four graduations: a_1 – Braşovia; a_2 – Castrum; a_3 – Marvis; a_4 – Sarmis;

– experimental factor b, culture substrate, with two graduations: b_1 – expanded clay; b_2 – perlite;

– experimental factor c, the hydroponic system: c_1 – Wilma; c_2 – Nutrient Film Technique.

These variants were tested in 3 repetitions.

For each variety 2 variants of industrial substrates and 2 hydroponic systems were tested. Materials used in the laboratory included Murashige-Skoog culture medium; laboratory equipment and instruments: oven, autoclave, UV lamp, air purifier, electronic balance, pH-meter, laminar air flow, test tubes, forceps, scalpels, sterile filter paper, refined alcohol required for sterilization, sterile distilled water, sterilizers, hand disinfectant. Materials used in hydroponic culture in "insect-proof" protected area included hydroponic systems, industrial substrates consisting of inorganic inert materials obtained from simple industrial processes - expanded clay and perlite, fertilizers: nutritional solutions for soilless crops, preparations based on soluble fertilizer "Universol", foliar fertilizers: Cropmax, Agroleaf, calcium nitrate needed to correct the nutrient solution reaction (with a total nitrogen content 15.5%, of which ammoniacal nitrogen 1.1%, calcium 19.0% Ca), pesticides, solutions to avoid salt blockages that may be deposited in drippers, timer.

Table 2. Experimental variants on different culture substrates and different hydroponic systems

Variant	Variety (a)	Substrate (b)	Hydroponic system
V_1	Braşovia (a_1)	Expanded clay (b_1)	Wilma (c_1)
V_2			Nutrient Film Technique (c_2)
V_3		Perlite (b_2)	Wilma (c_1)
V_4			Nutrient Film Technique (c_2)
V_5	Castrum (a_2)	Expanded clay (b_1)	Wilma (c_1)
V_6			Nutrient Film Technique (c_2)
V_7		Perlite (b_2)	Wilma (c_1)
V_8			Nutrient Film Technique (c_2)
V_9	Marvis (a_3)	Expanded clay (b_1)	Wilma (c_1)
V_{10}			Nutrient Film Technique (c_2)
V_{11}		Perlite (b_2)	Wilma (c_1)
V_{12}			Nutrient Film Technique (c_2)
V_{13}	Sarmis (a_4)	Expanded clay (b_1)	Wilma (c_1)
V_{14}			Nutrient Film Technique (c_2)
V_{15}		Perlite (b_2)	Wilma (c_1)
V_{16}			Nutrient Film Technique (c_2)

The hydroponic systems studied were:

– Wilma system used as a hydroponic system entirely recirculating which can reuse the unabsorbed nutrient solution in the irrigation process (contains: basin, tray, pots, pump, power system).

A frequent feeding means an increased supply of water and nutrients. Roots do not depend on the capillarity of the growing medium.

– the NFT culture system, on thin layer, circulating of nutrient solution.

The normal growth and development of plants is possible only in the presence of all the nutrients they need. In "no-soil" crops, plants are fed with all the nutrients they need through fertilization with complete nutritional solutions (Atanasiu, 2007).

RESULTS AND DISCUSSION

The number of minitubers was higher by using the NFT system (5.79), with a statistically significant positive difference of 0.5 minitubers (Table 3).

By using the perlite substrate, the number of minitubers obtained was higher (6.42) and by applying as a substrate the expanded clay the number of minitubers presented a distinctly significant negative difference (-1.75) (Table 4).

Table 3. Influence of the used hydroponic system on the number of obtained minitubers

Hydroponic system	Number	%	Diff.	Signif.
Wilma	5.29	100.00	-	-
NFT	5.79	109.45	0.5	*

LSD: 5% = 0.31; 1% = 0.72; 0.1% = 2.28.

Table 4. Influence of culture substrate used on the number of obtained minitubers

Culture substrate	Number	%	Diff.	Signif.
Perlite	6.42	100.00	-	-
Expanded clay	4.67	72.73	-1.75	oo

LSD: 5% = 0.55; 1% = 0.92; 0.1% = 1.72.

Regarding the influence of variety (Table 5) on minitubers production, it appears that the Castrum variety (6.67 minitubers) was superior to Brasov variety (5.00 minitubers), with a very significant positive difference (1.67), while Marvis and Sarmis varieties presented insignificant differences, inferior to control (4.83 and 5.67 minitubers). If we examine the average number of minitubers obtained in the four varieties for two variants of the culture substrate, it is observed that by using the substrate consisting of perlite, Castrum variety had the highest number of minituberculos/plant (8.17) (Figure 1).

Table 5. The influence of variety on the number of obtained minitubers

Variety	Number	%	Diff.	Signif.
Brasovia	5,00	100.00	-	-
Castrum	6.67	133.33	1.67	***
Marvis	4.83	96.67	-0.17	ns
Sarmis	5.67	113.33	0.67	ns

LSD: 5% = 0.69; 1% = 0.94; 0.1%= 1.26.

If we examine the average number of minitubers obtained in the four varieties for two variants of the culture substrate, it is observed that by using the substrate consisting of perlite, Castrum variety had the highest number of minitubers/plant (8.17) (Figure 1).

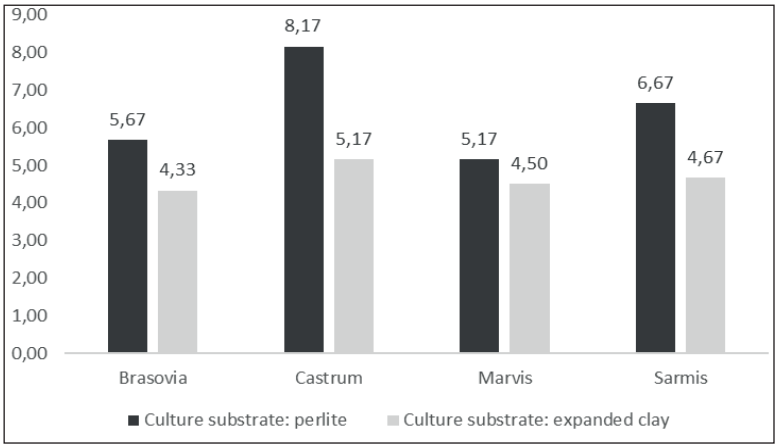

Figure 1. The average number of minitubers/plant obtained for each variety, depending on the substrate used

The combined influence of the hydroponic system and culture substrate used at planting confirmed the superiority of the perlite in the NFT hydroponic system, recording 7.5 minitubers, while by using expanded clay the number of minitubers presented a very significant negative difference (-3.42). Analyzing the two culture systems the superiority in minituberization of the NFT system when perlite is used as a culture substrate is highlighted through a distinctly positive difference (2.17 minitubers) (Table 6).

The significant superiority of the Castrum variety in the number of minitubers produced is manifested regardless of the hydroponic system used (Table 7).

Table 6. Influence of hydroponic and culture systems on number of obtained minitubers

Hydroponic system/ Culture substrate	Wilma Number	%	Diff.	Signif.	NFT Number	%	Diff.	Signif.	Diff. a_2-a_1	Signif.
Perlite	5.33	100.00	-	-	7.50	100.00	-	-	2.17	**
Expanded clay	5.25	98.44	-0.08	ns	4.08	54.44	-3.42	ooo	-1.17	o

LSD: 5% = 0.79; 1% = 1.30; 0.1% = 2.43. LSD: 5% = 1.06; 1% = 1.87; 0.1% = 4.04.

Table 7. The influence of hydroponic system and variety over number of obtained minitubers

Hydroponic system/ Variety	Wilma Number	%	Diff.	Signif.	NFT Number	%	Diff.	Signif.	Diff. a_2-a_1	Signif.
Brasovia	4.83	100.00	-	-	5.17	100.00	-	-	0.33	ns
Castrum	5.83	120.69	1.00	*	7.50	145.16	2.33	***	1.67	*
Marvis	5.00	103.45	0.17	ns	4.67	90.32	-0.50	ns	-0.33	ns
Sarmis	5.50	113.79	0.67	ns	5.83	112.90	0.67	ns	0.33	ns

LSD: 5% = 0.98; 1% = 1.33; 0.1% = 1.78. LSD 5% = 1.15; 1% = 1.63; 0.1% = 2.48.

Table 8. The influence of hydroponic system used on the weight of the minitubers obtained/plant

Hydroponic system	Weight of the minitubers/pl. (g)	%	Diff.	Signif.
Wilma	97.16	100.00	-	-
NFT	115.25	118.62	18.09	*

LSD: 5% = 16.62 g; 1% = 38.38 g; 0.1% = 122.15 g.

Researching the influence culture substrate used over weight of the minitubers/ plant (Table 9) shows that by using perlite, minitubers with superior weight values are obtained, compared to clay, which show a distinctly negative difference (-36.51 g).

Table 9. The influence of used culture substrate on the weight of the minitubers obtained/plant

Culture substrate	Weight of the minitubers/pl. (g)	%	Diff.	Signif.
Perlite	124.46	100.00	-	-
Expanded clay	87.95	70.67	-36.51	oo

LSD: 5% = 15.40 g; 1% = 25.48 g; 0.1% = 47.69 g.

The variety Castrum was significantly superior to all other tested cultivars in weight of minitubers per plant, while Marvis was significantly inferior (Table 10).

Table 10. Influence of variety on the weight of the minitubers obtained/ plant

Variety	Weight of the minitubers/pl. (g)	%	Diff.	Signif.
Brasovia	111.54	100.00	-	-
Castrum	130.24	116.77	18.70	**
Marvis	77.48	69.46	-34.07	ooo
Sarmis	105.56	94.64	-5.98	ns

LSD 5% = 13.56 g; 1% = 18.44 g; 0.1% = 24.69 g.

The weight of minitubers/plant was between 159.87 g (Castrum) and 83.90 g (Marvis) on perlite culture substrate; and 100.62 g (Castrum) and 71.05 g (Marvis) when using culture substrate formed by expanded clay balls (Figure 2).

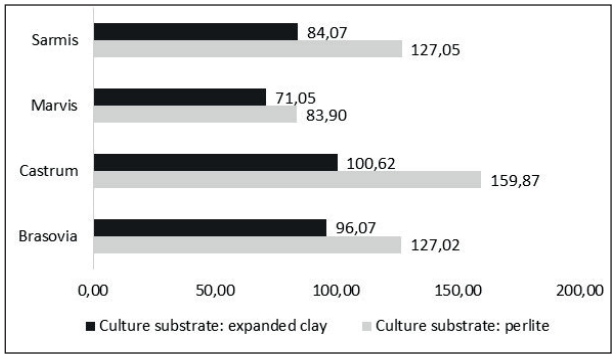

Figure 2. Weight (g) of minitubers obtained/plant for each variety, depending on the substrate used

By comparing the number of minitubers/ plant with their weight Castrum variety is remarkable in producing the biggest number of minitubers 6.67/plant with a high average weight – 130.24 g/pl. This is followed by the Sarmis variety, which records a number of 5.67 minitubers and 105.67 g/pl. Although Braşovia variety produces a lower number of minitubers/pl. their weight/pl. is 115.54 g (Figure 3).

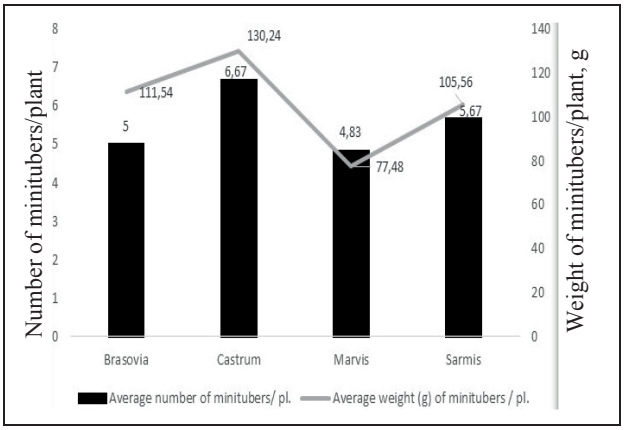

Figure 3. Number and weight (g) of minitubers/plant

The beneficial effect of substrate consisting of perlite in getting the best results regarding the weight of minitubers obtained per plant, for both hydroponic systems used (104.67 g when it is used Wilma system and 144.25 g for NFT system) can be observed (Table 11). The difference between between these hydroponic systems detaches the system NFT by using the perlite substrate, with a significant positive difference of 39.58 g.

Table 11. Influence of hydroponic system and of culture substrate over weight of minitubers obtained/plant

Hydroponic system/Culture substrate	Wilma (g)	%	Diff.	Signif.	NFT (g)	%	Diff.	Signif.	Diff. a_2-a_1	Signif.
Perlite	104.67	100.00	-	-	144.25	100.00	-	-	39.58	*
Expanded clay	89.65	85.65	-15.02	ns	86.25	59.79	-58.00	oo	-3.40	ns

LSD: 5% = 21.78 g; 1% = 36.03 g; 0.1% = 67.44 g. LSD: 5% = 22.16 g; 1% = 42.90 g; 0.1% = 109.11 g.

From the combined influence of variety and hydroponic system (Table 12) resulted a very significant negative difference for Marvis variety (-56.35g) in Wilma hydroponic system and very significant positive (35.85 g) in NFT system.

From examining the differences between these two hydroponic culture systems (NFT system and Wilma system) one can observe higher weight values/plant by appling NFT system in Marvis variety (41.52 g).

Table 12. Influence of hydroponic system and variety over weight of minitubers obtained/plant

Hydroponic system/ Variety	Wilma (g)	%	Diff.	Signif.	NFT (g)	%	Diff.	Signif.	Diff. a_2-a_1	Signif.
Brasovia	113.07	100.00	-	-	110.02	100.00	-	-	-3.05	-
Castrum	114.62	101.37	1.55	ns	145.87	132.59	35.85	***	31.25	ns
Marvis	56.72	50.16	-56.35	ooo	98.23	89.29	-11.78	ns	41.52	*
Sarmis	104.23	92.19	-8.83	ns	106.88	97.15	-3.13	ns	2.65	ns

LSD: 5% = 19.18 g; 1% = 26.07 g; 0.1% = 34.92 g. LSD 5% = 37.48 g; 1% = 62.49 g; 0.1% = 135.39 g.

CONCLUSIONS

Using the NFT hydroponic system had a positive influence on minituberization, both for number, but also as the weight of minitubers.

Regarding the influence of the culture substrate on minituberization the results showed that by using as a substrate of perlite high values for both studied parameters were obtained.

Castrum variety was remarked by obtaining a high number of minitubers and also by the weight of minitubers.

Using hydroponic systems for producing Prebasic seed material offers a high multiplying coefficient. Efficiency of using culture nutrient is the most important factor to achieve an increased yield in minituberization process.

Hydroponic systems may eliminate some of culture practices needed in the conventional process, reduce the potential contamination of plants with soil microorganisms, and lead to production of biological material with superior sanitary quality.

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