

## The potato minituber production from microtubers in aeroponic culture

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### ABSTRACT

Minituber production by traditional method is an expensive procedure due to limited productivity. In order to increase the multiplication rate of seed material *in vitro*, multiple techniques have been assayed in the last decades. The aeroponic system is a soilless culture system, where roots are kept in a dark environment saturated with aerosol of nutrient solution. Aeroponics technology is potentially efficient for specific potato cultivars. The aim of this study was to investigate the multiplication rate of microtubers of national cultivars in aeroponics while maintaining weekly intervals between harvests. The total number of harvests was 14 to 15 depending on year. The results of the experiments were analysed with ANOVA and means were separated with the Tukey's test at 5% P-value. The most important parameter of minituber production, their number, was on average 32.5–36.0 per plant and 1268–1396 per m<sup>2</sup> depending on cultivar. Number of minitubers was two to three times greater in the case of aeroponic production than by traditional method. A full economic analysis is necessary to prove that this production technique can be put into practice.

**Keywords:** differentiated systems of potato seed production; *in vitro* seed material; *Solanum tuberosum* L.

The potato (*Solanum tuberosum* L.) crop is propagated vegetatively by tubers, which implies the availability of disease-free planting materials. The introduction of microtubers and minitubers into seed production has revolutionized potato production, resulting in shortening of the field cycle to obtain an adequate number of seed potatoes and hence guaranteeing a high level of healthiness of base materials (Wróbel 2014).

Microtubers (or *in vitro* tubers) are miniature seed potatoes. They are the first generation of nuclear seed potato and their weight ranges from 24–273 mg, their diameter from 4–7 mm and their length 10–12 mm (Ranalli 2007). Microtubers are easier to transport and handle than plantlets and are less delicate, so require less aftercare during planting. Minitubers are usually defined as the progeny tubers produced on *in vitro* derived plantlets or microtubers. The term refers to their size as they are smaller than conventional seed tubers but larger than *in vitro* tubers produced under aseptic conditions on artificial media (Struik 2007). The size of minitubers may range from 5–25 mm although in current systems larger minitubers have

also become common. Minitubers from *in vitro* seed material are usually obtained in cultures growing on different substrates. However, it is an expensive procedure due to reduced productivity of the conventional seed multiplication systems (Tierno et al. 2014).

In order to increase the multiplication rate of seed material *in vitro*, multiple techniques have been assayed in the last decades, such as hydroponic systems (Chang et al. 2012) or the nutrient film technique (NFT) system (Rolot et al. 2002). However, most of these techniques have severe limitations due to an inadequate aeration of roots. Aeroponic systems for potato pre-basic seed production have been established following increased demand for more efficient, high quality seed production methods (Ritter et al. 2001, Nickols 2005). In this soilless culture system, roots are kept in dark environment saturated with aerosol of nutrient solution. Farran and Mingo-Castel (2006) found that the number and timing of non-destructive harvests were key factors in the optimization of minituber production, but according to Mateus-

Rodriguez et al. (2012) aeroponics technology is potentially efficient for specific potato cultivars.

The aim of this study was to investigate the multiplication rate of microtubers of national cultivars in aeroponics while maintaining weekly intervals between harvests.

## MATERIAL AND METHODS

The studies of minituber production were conducted using devices for aeroponic culture with a density of 36 and 42 plants per square meter and additionally by traditional method, in soil. The experiment was carried out over the course of 2 years 2012–2013 in Jadwisin (52°30'N, 21°04'E), Poland.

**Plant materials.** The microtubers of medium early cvs. Ametyst and Tajfun were obtained at the end of March from the Pomeranian-Mazurian Potato Breeding Company in Strzekęcino, Poland. The average weight of microtubers of cv. Ametyst was 246 mg and cv. Tajfun 195 mg, their transverse diameter was 7 mm and their lengths 12 mm and 8 mm. At the start of April they were placed in Petri dishes and were pre-sprouted for 5 weeks at temperatures of 18–20°C and next used for planting in devices for aeroponic culture and in the soil.

**Minituber production in aeroponic culture.** Microtubers were planted in plastic conical baskets filled with cubes of mineral wool (Grodan, Roermond, the Netherlands). The upper diameter of the baskets was 5 cm, bottom 3 cm, and a height of 5 cm. The baskets with microtubers were placed in cuvettes filled with clean water. After about one week the emergence of plants was completed and the first roots appeared. Then the baskets with plants were transferred to devices for aeroponic cultivation, on May 16 in 2012 and on May 13 in 2013. Their number for each cultivar was 54 or 64, depending on the planting density, therefore 27 and 32 in one of two sections. In the present study two identical devices were used for aeroponic cultivation (250 cm length, 122 cm width and 60 cm depth). They were

located in an open-roofed area next to a greenhouse, where there were natural thermal and light conditions for plant growth. Weather conditions during the years of study were monitored using a Campbell weather station (Campbell Scientific Inc., Logan, USA) located adjacent to the greenhouse and additionally using a thermohygrograph placed between aeroponics. The most important meteorological factor, the air temperature was presented in Table 1. The injection time of the nutrient solution was the same for both devices: 10 s every 3 min during the day ((5:05–20:00) and 10 s every 5 min during the night ((20:05–5:00). A modified nutritive solution (pH 5.8; electrical conductivity (EC) – 2.2 mS/cm;  $\text{NO}_3^-$ ,  $\text{SO}_4^{2-}$ ,  $\text{H}_2\text{PO}_4^-$ ,  $\text{Cl}^-$ ,  $\text{K}^+$ ,  $\text{Ca}^{2+}$ ,  $\text{Mg}^{2+}$ ,  $\text{NH}_4^+$ ,  $\text{Na}^+$  and micronutrients) from Rolot et al. (2002) was prepared and supplied by fog nozzles located at the bottom of the chambers. Residual nutrition solution was recirculated. During the growing season daily control measurements of pH and EC of nutrient solution in each reservoir were performed using a portable pH meter and portable EC meter (ScanGrow, Slangerup, Denmark). At the initial stage of tuberization (July 8–9) measurements of stem length were made on 10 plants of each combination in two sections. Harvests were made at weekly intervals starting on July 10 in 2012 and on July 9 in 2013. The last harvests were just after the first ground frost (October 8–16). The total number of harvests was 15 in 2012 and 14 in 2013. All minitubers of a size not smaller than 2 cm in length were collected at each harvest. The number and weight of minitubers from each section were measured. The number of minitubers per square meter and the size of individual minitubers were calculated.

**Minituber production by the traditional method.** Sprouted microtubers were planted in boxes with an area of 1925 cm<sup>2</sup> (55 × 35 cm) and 14 cm deep, filled with universal vegetable production soil substrate (Pasłek, Poland). The number of microtubers per box was 12, according to the way the most commonly used in potato breeding

Table 1. Mean values of daily air temperature (°C) during growing season in the years of study

	May	June	July	August	September	October	Mean
2012	13.9	15.6	15.2	17.4	12.8	6.7	13.6 <sup>b</sup>
2013	15.7	17.2	18.7	18.2	10.3	10.1	15.1 <sup>a</sup>

<sup>a,b</sup>mean values followed by the same letters are not significantly different at the 0.05 level according to the Tukey's test

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practice. Number of replicates (boxes with plants) was 6. The boxes were located next to the devices for aeroponic cultivation. Throughout the growing season the plants were carefully tended daily. Recommended plant protection products were used to control diseases and pests. The harvest was performed after full maturity of plants, depending on the years of study between August 10 and 12 for cv. Tajfun and between August 25 and 29 for cv. Ametyst. After the harvests the total number and yield of minitubers in each replicate were determined. The number of minitubers per plant and per square meter and average individual minituber size were calculated.

**Data analysis.** The results of the experiments were analysed with ANOVA using a general linear model of statistics program in SAS Enterprise Guide 4 (2004). Means were separated with the Tukey's test at 5% *P*-value.

## RESULTS AND DISCUSSION

**Minituber production in aeroponic culture.** In the present study the plants of investigated cultivars did not differ in terms of stem length at the time of beginning of tuberization and thus after the end of growth and there were found only slight differences resulting from the diverse density of planting (Table 2). In the study of Ritter et al. (2001) the height of plants of cv. Nagore was much larger (150–180 cm). This could be related to higher temperatures during the growing season conducive to the development of the aboveground part of potato plants (Rykczevska 2015). Tierno et al. (2014) demonstrated that average

Table 2. Stem length (cm) of potato plants in aeroponics depending on cultivar and plant density at the time of beginning of tuberization

	Cultivar		Plant density per m <sup>2</sup>	
	Ametyst	Tajfun	36	42
2012	68.4	69.8	65.2	72.9
2013	69.1	70.5	65.8	73.8
Mean	68.8 <sup>a</sup>	70.2 <sup>a</sup>	65.5 <sup>b</sup>	73.4 <sup>a</sup>

<sup>a,b</sup>mean values followed by the same letters are not significantly different at the 0.05 level according to the Tukey's test

plant height of three tested cultivars at 60 days after planting was from 49.6–92.7 cm, so these results are similar to ours.

The most important parameter of minituber production in aeroponics is their number per plant. In this study it was dependent mainly on the cultivar (Table 3). Other authors also point to the problem of diversity of cultivar reaction. Rolot and Seutin (1999) reported a cultivar response of 8 to 13 minitubers in each plant. Then Chang et al. (2011) found that the average number of tubers per plant was higher in the cv. Superior than in cvs. Haryeong and Jayoung. The results in this study are slightly similar to those described by Chang et al. (2012), who found that the number of minitubers in cv. Superior was 37.6 and in cv. Atlantic 22.7. However, according to Mateus-Rodriguez et al. (2013) a multiplication rate of 1:45 is possible and could be based on individual management by cultivar. In this study a significant positive impact of lower planting density on the number of minitubers per plant was found. The direction of reaction of the tested cultivars was consistent

Table 3. Parameters of microtubers (MT) multiplication in aeroponics depending on cultivar and plant density – in total for all dates of harvest

		Cv. Ametyst			Cv. Tajfun			Mean		
		36/m <sup>2</sup>	42/m <sup>2</sup>	mean	36/m <sup>2</sup>	42/m <sup>2</sup>	mean	36/m <sup>2</sup>	42/m <sup>2</sup>	mean
Number of MT per plant	2012	44.2	38.0	41.0 <sup>a</sup>	35.1	34.9	35.0 <sup>b</sup>	39.7	36.5	38.1 <sup>a</sup>
	2013	33.8	28.1	31.0 <sup>b</sup>	30.8	29.3	30.1 <sup>b</sup>	32.3	28.7	30.5 <sup>b</sup>
	mean	39.0 <sup>a</sup>	33.1 <sup>b</sup>	36.0 <sup>a</sup>	33.0 <sup>a</sup>	32.1 <sup>a</sup>	32.5 <sup>b</sup>	36.0 <sup>a</sup>	32.6 <sup>b</sup>	34.3
Number of MT/m <sup>2</sup>	mean	1404	1388	1396 <sup>a</sup>	1187	1349	1268 <sup>b</sup>	1296 <sup>a</sup>	1369 <sup>a</sup>	1333
Yield of MT (kg/m <sup>2</sup> )	2012	12.60	11.77	12.19	9.82	11.55	10.68	11.21	11.66	11.44
	2013	17.18	13.99	15.58	12.54	13.00	12.77	14.86	13.50	14.18
	mean	14.89	12.88	13.89 <sup>a</sup>	11.18	12.28	11.73 <sup>a</sup>	13.04 <sup>a</sup>	12.58 <sup>a</sup>	12.81
Size of individual MT (g)	mean	11.00	9.62	10.31 <sup>a</sup>	9.54	9.22	9.38 <sup>a</sup>	10.27 <sup>a</sup>	9.42 <sup>a</sup>	9.85

<sup>a,b</sup>mean values followed by the same letters are not significantly different at the 0.05 level according to the Tukey's test

Table 4. Correlation coefficients between the mean values of daily temperature during growing season in the years of study and number and yield of minitubers (MT) ( $n = 8$ )

Tested factor	Mean daily temperature
Number of MT per m <sup>2</sup>	-0.8750**
Yield of MT per m <sup>2</sup>	+0.7720**

\*\* $P \leq 0.01$ ; \* $P \leq 0.05$

with the results obtained by Farran and Mingo-Castel (2006), who also found a reduction in the number of minitubers at higher planting density.

In the present study the number of minitubers was significantly differentiated depending on year (Table 3). This was the result of different temperatures during the growing seasons, which was evidenced by a highly significant negative correlation between the mean values of daily temperature in the years of study and the number of minitubers (Table 4). However, the response of the tested cultivars was diverse and statistically significant only in the case of cv. Ametyst. The results are consistent with those reported by Oraby et al. (2015), who pointed out the negative impact of higher temperature in the production of minitubers by aeroponic method. In fact, temperature is a very important factor influencing photosynthesis and also the number of tubers and yield of potato plants even in years with weather conditions generally favourable for potato development (Rykaczewska 2015, Rykaczewska and Mańkowski 2015). However, Oraby et al. (2015) suggest that it

Table 5. Parameters of microtubers (MT) multiplication in soil depending on cultivar

Cultivar	Number of MTs per plant	Number of MTs per m <sup>2</sup>	Yield of MTs (kg per m <sup>2</sup> )	Individual MT size (g)
Ametyst	11.27 <sup>a</sup>	679 <sup>a</sup>	9.80 <sup>a</sup>	14.5 <sup>b</sup>
Tajfun	7.16 <sup>b</sup>	428 <sup>b</sup>	9.26 <sup>a</sup>	21.6 <sup>a</sup>
Mean	9.21	553	9.53	18.1

<sup>a,b</sup>mean values followed by the same letters are not significantly different at the 0.05 level according to the Tukey's test

is possible to increase potato minituber production in aeroponic systems under high summer temperatures by cooling the nutrient solution to 18°C. The number of minitubers per square metre is dependent not only on cultivar, but also on the number of plants per surface area (Rolot and Seutin 1999, Faran and Mingo-Castel 2006). Most authors, however, do not provide data concerning the number of minitubers per unit area.

In this study the yield of minitubers was also significantly differentiated depending on year (Table 3). Between the mean values of daily temperature in the years of study and the yield, a highly significant positive correlation was found (Table 4). Thus, the thermal conditions in 2013 did not allow for the formation of a larger number of minitubers, but positively influenced their size. It is confirmed by the highly significant positive correlation between the mean values of daily temperature in the years of study and the size of individual minitubers (Table 4). The size of individual minitubers pro-

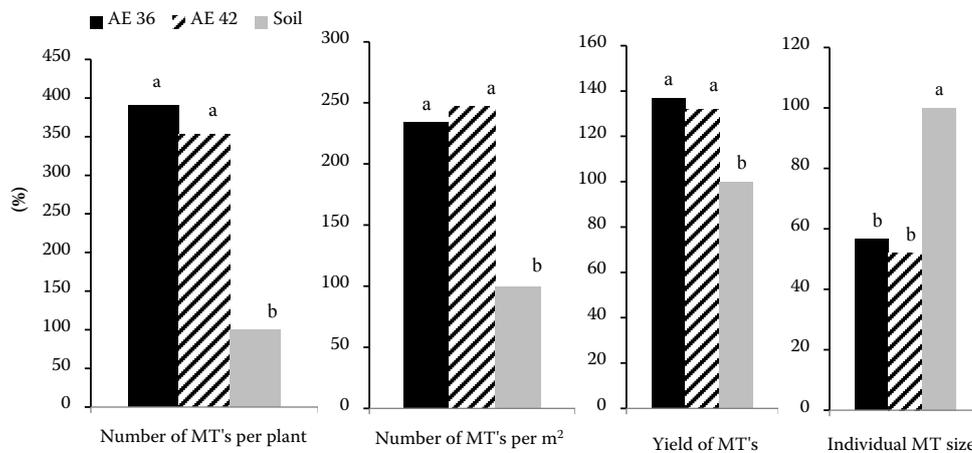


Figure 1. Comparison of parameters of microtuber (MT) multiplication in aeroponics in relation to these multiplication parameters in soil – mean values for cultivars and years; values of traditional cultivar represent 100%. AE 36 – density of microtubers planting 36/m<sup>2</sup>; AE 42 – density of microtubers planting 42/m<sup>2</sup>; <sup>a,b</sup>mean values followed by the same letters are not significantly different at the 0.05 level according to the Tukey's test

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duced in aeroponics, presented in the literature is quite varied and is dependent on cultivar and harvesting intervals (Chang et al. 2011, 2012). In this experiment all minitubers of a size not smaller than 2 cm in length were collected.

**Minituber production by the traditional method.** Significant differences between the studied cultivars in terms of number and size of minitubers were found (Table 5). For this reason there were no significant differences between cultivars in minituber yield.

**Comparison of parameters of microtuber multiplication in aeroponics and in the soil.** There were significant differences between all examined parameters of minitubers produced by aeroponic and traditional method (Figure 1). Number of minitubers produced per plant and per square meter was two to three times greater in aeroponic production than by traditional method. However, differences between the total yields were lower, due to much larger size of minitubers produced in the soil.

In conclusion, the results of this study showed that the aeroponics system is a viable technological alternative for the potato minituber production component within a potato tuber seed system. The cultivar played a significant role in the number of tubers formed. A full economic analysis including energy cost, labour costs and amortization of material specific to aeroponics is necessary to prove that this production technique can be put into practice.

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