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PRODUCTION OF HIGH-QUALITY SEED POTATOES IN PROTECTED AREA FOR TRUE SEED PROGENIES, WHO SHOWED TOLERANCE TO *IN VITRO* INDUCED WATER STRESS

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Abstract

Given the current climate context, the main objective of this study was to identify genotypes tolerant to water stress in true potato seed populations. The research was initiated by inoculating of true potato seeds on culture medium, regenerating viable plantlets and testing their tolerance to in vitro induced water stress. Three from nine tested genotypes obtained the best results in terms of tolerance to water stress induced in vitro. ELISA testing revealed that the three genotypes (GIL 19-03-07, ZIL 19-02-43 and GIL 19-03-29) are virus-free. In vitro true seed derived plantlets owning a very high health status were transplanted, after acclimatization, in protected area in order to produce top quality, disease-free seed potato material. Several planting variants were used in terms of biological material used, number of plants/pot and location. After minitubers were harvested, it was analyzed how the planting variants influenced number, size and weight of the minitubers obtained in the protected area. This study aimed to obtain an alternative planting material with a high phytosanitary quality and drought tolerance, to supplement the seed potato required by the market.

Key words*: in vitro plantlets, minituber, potato, true potato seed, water stress.*

INTRODUCTION

According to FAO estimates, in 2019 over 370 million tons of potatoes were produced worldwide. Potatoes are recommended as a food security crop by the United Nations. In 2019, more than 17 million hectares of potatoes were harvested worldwide (Shahbandeh, 2021). One of the pressing problems of the contemporary world is to ensure food for a continuously growing population. Also, the world economic crisis leads to necessity of finding some abundant food sources as minimal cost. Hence, enhancing the productivity of potato crops can contribute to fulfilling the nutritional requirements of the rising population (Birch et al., 2012). However, drought stress represents a major challenge to the production of potatoes worldwide. Climate change is predicted to further aggravate this challenge by intensifying potato crop exposure to increased drought severity and frequency (Gervais et al., 2021). To find solutions for these climate challenges researchers around the world are making continuous efforts to find new potato genotypes tolerant to water stress. The shallow root system of potatoes makes this crop one of the most drought-sensitive species (Gervais et al., 2021; Zarzynska et al., 2017; Iwama and Yamaguchi, 2006; Yuan et al., 2003). Drought strongly inhibits key physiological and biochemical processes, leading to poor plant performance and tuber yield loss (Evers et al., 2010; Stark et al., 2013; Obidiegwu et al., 2015; Aliche et al., 2018; Plich et al., 2020; Hill et al., 2021).

Although the potato plant is multiplied using a number of different techniques, *in vitro* nodal cuttings are probably the most common propagules used in early stages of commercial seed potato production (Pruski, 2007). The technology of production potato minitubers by direct transplantation of *in vitro* plantlets in protected areas is a frequently used technique in seed potato production system.

Seed potato production involving minituber production systems creates a bridge between the *in vitro* rapid multiplication and the field

multiplication of seed tubers and is thus a classical way to multiply or acclimatize *in vitro* material before its use in the open field (Sharma and Pandey, 2013). Producing minitubers from *in vitro* plantlets allows a faster rate of multiplication and reduces the number of field generations needed in seed production (Ranalli, 1997).

The phytosanitary condition of planting material is largely responsible for the size and quality of potato production. The real production capacity of a variety can be expressed only in healthy crops, obtained from a high quality planting material. Due to the physiological and virotic degeneration of potato, there is a progressive decrease in production. Virotic infections disrupt plant metabolism manifesting itself by shortening the growing season, accentuated decrease of production, depreciation of its quality (Donescu, 2003).

Abiotic stresses such as extreme temperature and drought often result in significant losses to the yields of economically important crops such potato (El-Magawry et al., 2015). *In vitro* screening of new genotypes represents valuable tool as alternative to field trials.

The objectives of the present study were to produce an alternative seed potato planting material with a high phytosanitary quality and drought tolerance.

MATERIALS AND METHODS

The biological material was represented by three potato genotypes (GIL 19-03-07, ZIL 19-02-43 and GIL 19-03-29) derived from botanical seed and obtained best results regarding the tolerance to *in vitro* induced water stress. The experiment was conducted at the Plant Tissue Culture Laboratory of the National Institute of Research and Development for Potato and Sugar Beet Brasov, Romania. The *in vitro* disease-free plantlets were acclimatized and planted in protected *insect-proof* area in order to produce high quality potato seed. Genotypes were evaluated in terms of number, size and weight of minitubers, both under optimal watering conditions and thermohydric stress.

Several planting variants were used:

- biological material: plantlets and minitubers;
- number of plantlets/pot: 1 plantlet/pot (control) and 2 plantlets/pot;
- location: greenhouse with thermohydric stress conditions (control) and tunnel "*insect-proof*" with optimal watering but high temperatures.

After the minitubers were harvested, it was analyzed how the planting variants influenced the number, size and weight of the minitubers obtained in protected area.

Whereas potato plantlets obtained *in vitro* are fragile, in order to have a high percentage of survival after transplantation, the microplants were acclimatized in greenhouse, for 3 weeks. The substrate used for planting consisted of a mixture of red peat with bentonite, black peat and perlite in a 4:2:1 ratio. The substrate was distributed in plastic pots with a diameter of 7 cm, 12.9 cm height and volume of 2 l. In order to enrich the substrate with nutrients, complex NPK 15:15:15 + 6% sulfur fertilizer was applied before planting, after which the substrate was watered daily (2 hours/day). Also, to ensure the nutrients necessary for growth and development of potato plants, organic foliar fertilizer (Cropmax) treatments were applied once a week.

During the growth season daytime temperature was monitored both in tunnel *insect-proof* and greenhouse (Table 1). In tunnel *insect-proof* the plants had an optimal watering regime, while in the greenhouse the potato plants were grown under water stress conditions.

Table 1. Daytime temperature (minimum and maximum) during the growing season

Month	Minimum and maximum temperatures recorded during the growing season (°C)			
	Tunnel *insect-proof*		Greenhouse	
	Morning (9°°)	Afternoon (14°°)	Morning (9°°)	Afternoon (14°°)
June	11 – 34	14 - 53	12 - 33	16 - 47
July	20 – 32	24 - **56**	21 - **38**	25 - **50**
August	15 – **37**	28 - 54	16 - 31	25 - 46
September	8 – 19	23 - 46	13 - 18	22 - 36

Planting has been carried out on May 20 (Figure 1), watering stop on September 1, manual removal of haulms was performed on September 15 (Figure 3) and harvesting minitubers was made in September 30 (Figure 4).
Chemical treatments have been carried out periodically to control pests and diseases (Figure 2). Foliar fertilizer was also applied weekly in order to ensure the harmonious development of the potato plants.

Figure 1. Planting of microplants in tunnel *insect-proof*

Figure 2. Chemical treatments and fertilization

Figure 3. Manual removal of haulms

Figure 4. Harvesting minitubers

In order to obtain a high quality planting material, a series of preventive measures have been taken to allow the most effective control of possible viral infections: destruction of weeds around the tunnel, which could be host plants for aphids (vector for potato viruses); placement of Moericke water traps inside the tunnel, collection of insects caught in these traps and their identification by qualified personnel; performing chemical treatments to control aphids. In order to verify the effectiveness of the preventive measures of virotic control on the phytosanitary quality of the material, samples of leaves were taken for ELISA testing during the vegetation. The ideal time to harvest the leaves is before flowering, when the plant is young, or at the latest during flowering. Only the top leaflet is detached from the upper third of the plant. The ELISA results showed that all 3 genotypes tested (GIL19-03-07, ZIL19-02-43 and GIL19-03-29) are healthy.
Minitubers were harvested approximately 19 weeks after planting and true potato seed progenies were evaluated for average number, size and weight of minitubers. The obtained results were processed by the analysis of variance (Săulescu and Săulescu, 1967).

RESULTS AND DISCUSSIONS

In this study, 3 true potato seed progenies, who showed tolerance to *in vitro* induced water stress, were grown in protected areas in order to obtain a high-quality pre-basic seed from *in vitro* plantlets. After 19 weeks from planting, the minitubers were harvested and evaluated in terms of number, size and weight depending on planting variants.

Effect of growth conditions, number of plantlet/pot and genotypes on the number of minitubers

Both in tunnel *insect-proof* and greenhouse conditions in the case of the planting variant with 2 plantlets/pot a higher number of minitubers was obtained compared to the control (Table 2), the differences being significant both in the tunnel (2.89) and in the greenhouse (2.44).
Under the tunnel *insect-proof* growth conditions, the genotypes GIL19-03-07 and ZIL19-02-43 registered a distinctly significant positive

difference (3.25) and respectively significant positive (2.17) compared to the control. In the greenhouse conditions (thermohydric stress) it was highlighted genotype ZIL19-02-43 which obtained the highest number of minitubers/pot (ave. 8.67 minitubers/pot) with a very significant positive difference (6.33) compared to the control. By comparing the results obtained in the tunnel with those obtained in the greenhouse, regarding the number of minitubers, a distinctly significant positive difference (4.33) can be observed in the tunnel conditions for the genotype GIL19-03-07 (Table 3).

The results presented in Table 3 show an important aspect regarding the behavior in different growth conditions of tested potato genotypes about the number of minitubers. Thus, ZIL 19-02-43 showed tolerance in the conditions of thermohydric stress by obtaining a higher value of the number of minitubers in the greenhouse (8.67) compared to the tunnel (6.58).

Table 2. Effect of growth conditions and number of plantlet/pot on the number of minitubers

Tunnel	Mean	Diff.	Sign.	Greenhouse	Mean	Diff.	Sign.	a_1-a_2	Sign.
1 plantlet/pot (Ct)	4.78	-	-	1 plantlet/pot (Ct)	3.56	-	-	1.22	ns
2 plantlets/pot	7.67	2.89	*	2 plantlets/pot	6.00	2.44	*	1.67	ns
	DL 5%: 2.02; 1%: 3.34; 0.1%: 6.25				DL 5%: 1.95; 1%: 3.83; 0.1%: 9.96				

Table 3. Effect of genotypes and growth conditions on the number of minitubers

Tunnel	Mean	Diff.	Sign.	Greenhouse	Mean	Diff.	Sign.	a_1-a_2	Sign.
GIL19-03-07	7.67	3.25	**	GIL19-03-07	3.33	1.00	ns	4.33	**
ZIL19-02-43	6.58	2.17	*	ZIL19-02-43	8.67	6.33	***	-2.08	ns
GIL19-03-29 (Ct)	4.42	-	-	GIL19-03-29 (Ct)	2.33	-	-	2.08	ns
	DL 5%: 1.79; 1%: 2.46; 0.1%: 3.39				DL 5%: 2.10; 1%: 3.67; 0.1%: 8.53				

Table 4. Effect of number of plantlet/pot and genotypes on the number of minitubers

1 plantlet/pot	Mean	Diff.	Sign.	2 plantlets/pot	Mean	Diff.	Sign.	b_2-b_1	Sign.
GIL19-03-07	5.08	2.17	*	GIL19-03-07	5.92	2.08	*	0.83	ns
ZIL19-02-43	4.50	1.58	ns	ZIL19-02-43	10.75	6.92	***	6.25	***
GIL19-03-29 (Ct)	2.92	-	-	GIL19-03-29 (Ct)	3.83	-	-	0.92	ns
	DL 5%: 1.79; 1%: 2.46; 0.1%: 3.39				DL 5%: 2.02; 1%: 3.02; 0.1%: 4.86				

When used a single plantlet/pot, GIL19-03-07 registered a significant positive difference (2.17) compared to the control, and in planting variant with 2 plantlets/pot very significant differences were obtained (6.92) and significant positives (2.08) for ZIL19-02-43 and GIL19-03-07, respectively (table 4). When used 2 plantlets/pot the genotype ZIL19-02-43 recorded the highest number of minitubers (ave. 10.75), registering a very significant positive difference compared to the variant with 1 plantlet/pot (Table 4).

Effect of growth conditions, number of plantlet/pot and genotypes on the weight of minitubers

Under the tunnel *insect-proof* growth conditions, the variant with 2 plantlets/pot led to obtaining minitubers with a higher weight compared to the control variant (1 plantlet/pot), registering a distinctly significant positive difference (56.23 g). Comparing the results obtained in tunnel with those obtained in greenhouse, in terms of minitubers weight there is a distinctly significant positive difference (49.34 g) in the case of variant with 1 plantlet/pot and a very significant positive difference (111.25 g) in the case of variant with 2 plantlets/pot (Table 5).

Under the tunnel *insect-proof* growth conditions, regarding the weight of the minitubers, the best results were obtained by ZIL19-02-43 (ave. 100.10 g), with a significant positive difference (23.34 g) compared to the control. By comparing the results obtained in tunnel with those obtained in greenhouse conditions, in terms of the weight of the minitubers, there were very significant differences for all three studied genotypes (Table 6).

Table 5. Effect of growth conditions and number of plantlet/pot on the weight of minitubers

Tunnel	Mean	Diff.	Sign.	Greenhouse	Mean	Diff.	Sign.	a_1-a_2	Sign.
1 plantlet/pot (Ct)	59.28	-	-	1 plantlet/pot (Ct)	9.94	-	-	49.34	**
2 plantlets/pot	115.52	56.23	**	2 plantlets/pot	4.27	-5.68	ns	111.25	***

DL 5%: 28.18 g; 1%: 46.63 g; 0.1%: 87.27 g DL 5%: 22.18 g; 1%: 39.49 g; 0.1%: 86.79 g

Table 6. Effect of genotypes and growth conditions on the weight of minitubers

Tunnel	Mean	Diff.	Sign.	Greenhouse	Mean	Diff.	Sign.	a_1-a_2	Sign.
GIL19-03-07	85.34	8.58	ns	GIL19-03-07	10.15	2.38	ns	75.19	***
ZIL19-02-43	100.10	23.34	*	ZIL19-02-43	3.40	-4.37	ns	96.70	***
GIL19-03-29 (Ct)	76.76	-	-	GIL19-03-29 (Ct)	7.77	-	-	68.99	***

DL 5%: 18.18 g; 1%: 25.04 g; 0.1%: 34.47 g DL 5%: 18.27 g; 1%: 29.35 g; 0.1%: 59.15 g

Regarding the average weight of minitubers/pot in the case of 1 plantlet/pot variant, there were no significant differences between genotypes, but in the case of 2 plantlets/pot variant, the genotype GIL19-03-07 was noted, which recorded a distinctly significant positive difference (27.98 g) compared to the control, obtaining the highest value for the average weight of the minitubers/pot (ave. 74.76 g). Comparing the results obtained in the case of 2 plantlets/pot variant with those obtained in the case of 1 plantlet/pot variant in terms of the weight of minitubers, a distinctly significant positive difference (54.03 g) was observed for the GIL19-03-07 genotype (Table 7).

Table 7. Effect of number of plantlet/pot and genotypes on the weight of minitubers

1 plantlet/pot	Mean	Diff.	Sign.	2 plantlets/pot	Mean	Diff.	Sign.	b_2-b_1	Sign.
GIL19-03-07	20.73	-17.02	ns	GIL19-03-07	74.76	27.98	**	54.03	**
ZIL19-02-43	45.36	7.61	ns	ZIL19-02-43	58.14	11.37	ns	12.78	ns
GIL19-03-29 (Ct)	37.75			GIL19-03-29 (Ct)	46.78			9.02	ns

DL 5%: 18.18g; 1%: 25.04 g; 0.1%: 34.47 g DL 5%: 24.63 g; 1%: 37.87g; 0.1%: 63.81g

Calibration on size fractions of minitubers harvested from protected area

After harvesting, the minitubers were calibrated in size fractions. Depending on their size, the minitubers were distributed on several calibration classes as follows: <10 mm, 10-15 mm, 15-20 mm, 20-25 mm, 25-30 mm and >30 mm. It was followed how aspects such as: genotype, cultivation conditions (tunnel, greenhouse), number of plantlet/pot (1 or 2 plantlets/pot) and the type of material used for planting (minitubers or microplants) influenced the size of the minitubers and which was their proportion in different planting variants.

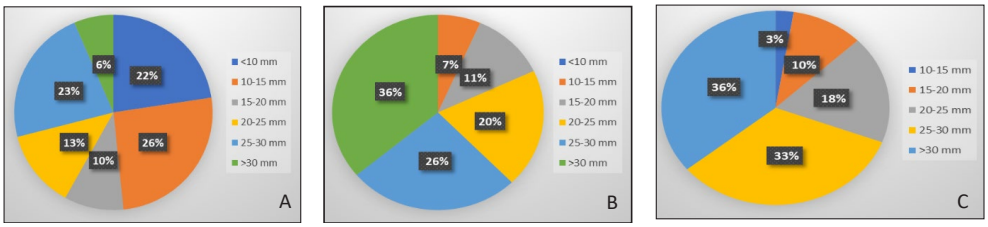

Figure 5. Percentage of minitubers by size fractions for GIL 19-03-07 in tunnel *insect-proof* depending on planting variant (A - 1 plantlet/pot; B - 2 plantlets/pot; C - minitubers)

Regarding genotype GIL 19-03-07, in tunnel *insect-proof* cultivation conditions, for planting variant in which 1 plantlet/pot was used, the highest percentage of minitubers was in 10-15 mm size fraction, and in case of variant in which minitubers and 2 plantlets/pot were used for planting, the highest percentage was registered at the size fraction > 30 mm (Figure 5).

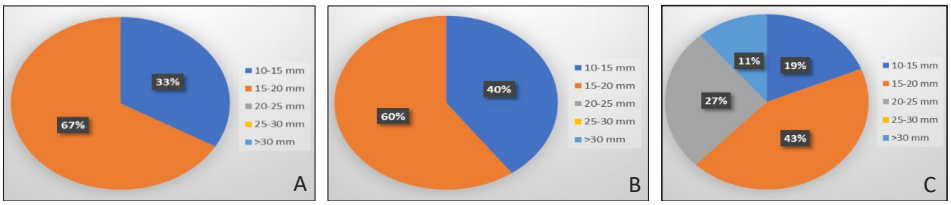

Figure 6. Percentage of minitubers by size fractions for GIL 19-03-07 under thermohydric stress conditions (greenhouse) depending on planting variant (A - 1 plantlet/pot; B - 2 plantlets/pot; C - minitubers)

In the greenhouse cultivation conditions, for GIL 19-03-07, the minitubers from the calibration class 15-20 mm had the highest frequency, followed by the size fraction 10-15 mm in the case of variants with 1 plantlet/pot and 2 plantlets/pots and the size fraction 20-25 mm when using minitubers as planting material (Figure 6).

Regarding genotype ZIL 19-02-43, under tunnel *insect-proof* conditions, for all three planting variants used (1 plantlet/pot, 2 plantlets/pot and minitubers) the highest percentage of minitubers was framed in the size fraction >30 mm, followed by size fraction 25-30 mm (Figure 7).

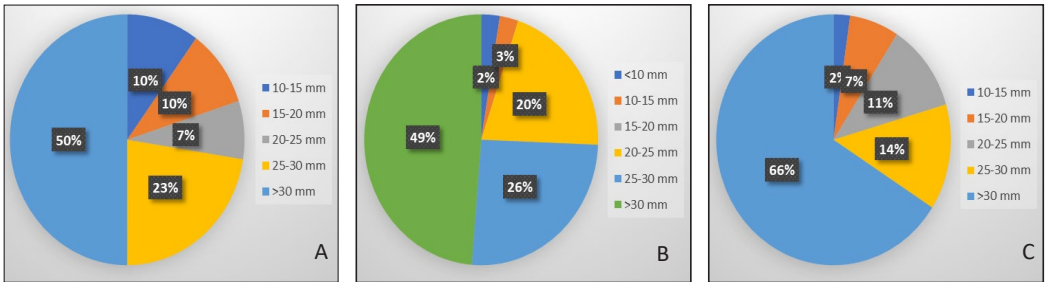

Figure 7. Percentage of minitubers by size fractions for ZIL19-02-43 in tunnel *insect-proof* depending on planting variant (A - 1 plantlet/pot; B - 2 plantlets/pot; C - minitubers)

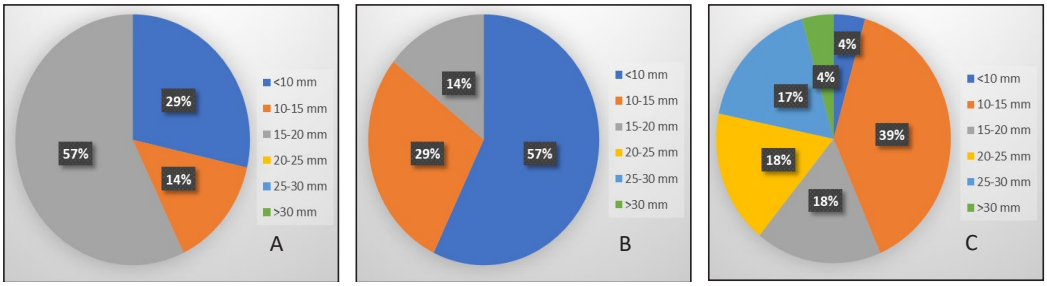

Figure 8. Percentage of minitubers by size fractions for ZIL19-02-43 under thermohydric stress conditions (greenhouse) depending on planting variant (A - 1 plantlet/pot; B - 2 plantlets/pot; C - minitubers)

Under greenhouse conditions of cultivation genotype ZIL 19-02-43 had a different behavior in terms of the size of the minitubers, depending on the material used for planting. Thus, for the variant with 1 plantlet/pot minitubers from the calibration class 15-20 mm had the highest frequency, for the variant with 2 plantlets/pot most minitubers were in the calibration class <10 mm, and in the case of the variant in which minitubers were used as planting material, the largest share of harvested minitubers was in the size fraction 10-15 mm (Figure 8).

Figure 9 shows the results obtained regarding the size of minitubers belonging to genotype GIL 19-03-29 and their distribution by calibration classes under the cultivation conditions specific to *insect-proof* tunnel. Thus,

in the case of planting variants with minitubers and 1 plantlet/pot the highest percentage was held by the minitubers from the size fraction > 30 mm, and in the case of the variant with 2 plantlets/pot, the minitubers had the highest share from the size fraction 25-30 mm.
Under greenhouse conditions for GIL 19-03-29 minitubers from the calibration class 10-15 mm had the highest frequency in the case of variants with 1 plantlet/pot and 2 plantlets/pot, and in the case of using minitubers as planting material the largest percentage of minitubers was in the size fraction 15-20 mm.
On the next place was located as frequency the minitubers from calibration class 20-25 mm (Figure 10).

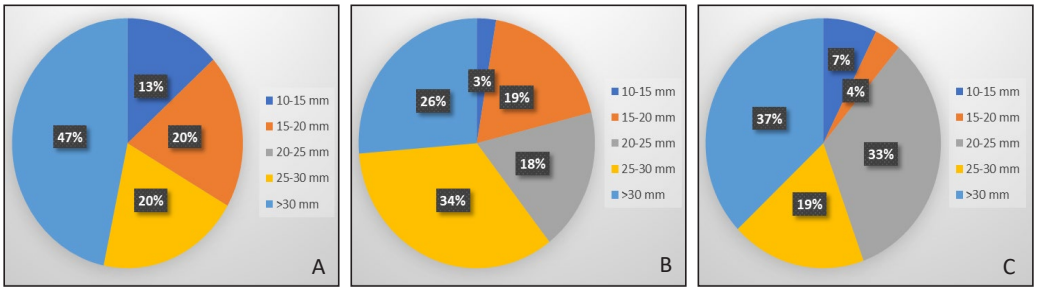

Figure 9. Percentage of minitubers by size fractions for GIL 19-03-29 in tunnel *insect-proof* depending on planting variant (A - 1 plantlet/pot; B - 2 plantlets/pot; C - minitubers)

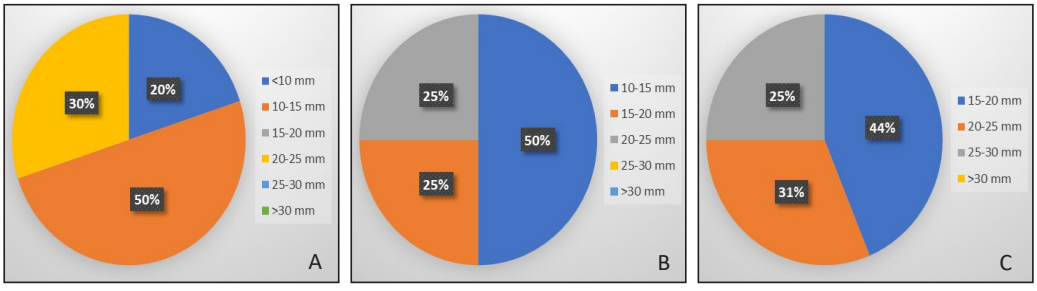

Figure 10. Percentage of minitubers by size fractions for GIL 19-03-29 under thermohydric stress conditions (greenhouse) depending on planting variant (A - 1 plantlet/pot; B - 2 plantlets/pot; C - minitubers)

As there are no official regulations regarding calibration of minitubers on size fractions, all minitubers larger than 10 mm are accepted as planting material. Also, considering the phytosanitary top quality of the biological material obtained in the *insect-proof* spaces and the high costs necessary to obtain this material, the minitubers smaller than 10 mm are kept and will be replanted in the protected area next year.

Table 8. Frequency of minitubers on size fractions depending on cultivation conditions and planting variant

Planting variant	Size fraction					
	GIL19-03-07		ZIL19-02-43		GIL19-03-29	
	Tunnel	Greenhouse	Tunnel	Greenhouse	Tunnel	Greenhouse
1 plantlet/pot	10-15 mm	15-20 mm	>30 mm	15-20 mm	>30 mm	10-15 mm
2 plantlets/pot	>30 mm	15-20 mm	>30 mm	<10 mm	25-30 mm	10-15 mm
Minitubers	>30 mm	20-25 mm	>30 mm	10-15 mm	>30 mm	15-20 mm

As can be seen in Table 8, for all three genotypes, the highest percentage of harvested minitubers was recorded in calibration classes larger than 10 mm, with one exception: genotype ZIL19-02-43 in greenhouse conditions and in case of variant with 2 plantlets/pot.

Under the tunnel *insect-proof* conditions (optimal watering but high temperatures), the obtained minitubers were mainly classified in the calibration classes > 30 mm and 25-30 mm in all genotypes and on all planting variants, except for the genotype GIL19-03-07 in which the highest frequency of minitubers obtained on variant with 1 plantlet/pot was in calibration class 10-15 mm.

Even in thermohydric stress conditions (greenhouse), the highest percentage had the minitubers from the fractions of size 10-15 mm and 15-20 mm, for all 3 potato genotypes, except for the genotype ZIL19-02-43 on variant with 2 plantlets/pot.

Regarding the production of minitubers (kg/m^2) obtained both in thermohydric stress conditions (greenhouse) and in optimal watering conditions but high temperatures (tunnel), the three genotypes obtained higher yields in tunnel compared to greenhouse (Table 9).

Table 9. The yield of true potato seed progenies depending on the cultivation conditions and the planting variant

Planting variant	**Minitubers yield (kg/m^2)**					
	GIL19-03-07		ZIL19-02-43		GIL19-03-29	
	Tunnel	Greenhouse	Tunnel	Greenhouse	Tunnel	Greenhouse
1 plantlet/pot	1.0	0.5	3.0	0.2	2.5	0.4
2 plantlets/pot	5.0	0.2	4.0	0.1	3.0	0.2
Minitubers	4.0	2.0	7.5	0.7	3.5	0.8

Regarding the planting variant, analyzing the results presented in Table 9, in tunnel *insect-proof* conditions the true potato progenies obtained higher yields in case of using 2 plantlets/pot and minitubers as initial material compared to the variant with 1 plantlet/pot.

Two important aspects can be deduced from this, namely:

- increasing productivity by optimizing the capitalization of small fraction minitubers;
- more efficient use of culture space.

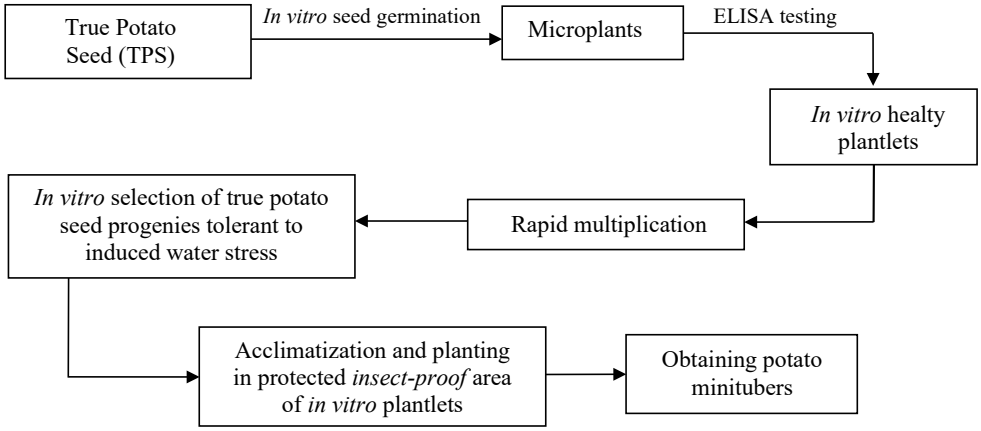

Figure 11. Scheme for production in protected area of potato minitubers (Prebase) starting from *in vitro* germination of true potato seeds

Figure 11 shows the steps that were followed to obtain potato minitubers in protected *insect-proof* area starting from true potato seed (TPS) cultivated under *in vitro* conditions. This scheme includes both the stage of testing tolerance to *in vitro* induced water stress of potato genotypes derived from true seed, and the acclimatization of potato microplants obtained *in vitro* before planting them in protected *insect-proof* area.

CONCLUSIONS

Based on *in vitro* screening for drought stress three of nine true potato seed progenies were selected as tolerant.

Regarding the average number of minitubers/pot obtained under *insect-proof* conditions the genotypes GIL19-03-07 (7.67) and ZIL19-02-43 (6.58) were highlighted. Under thermohydric stress conditions the genotype ZIL19-02-43 obtained the highest number of minitubers (ave. 8.67). For variant with 2 plantlets/pot a higher number of minitubers was obtained both in tunnel (ave. 7.67) and in the greenhouse (ave. 6.00), compared to variant in which 1 plantlet/pot was used (ave. 4.78, respectively ave. 3.56).

Regarding the average weight of the minitubers, in tunnel the variant with 2 plantlets/pot led to obtaining minitubers with a higher weight (ave. 115.52 g) compared to 1 plantlet/pot variant (ave. 59.28 g), but in conditions of thermohydric stress the weight of minitubers was higher on the control variant (ave. 9.94 g) compared to 2 plantlets/pot variant (ave. 4.27 g).

In tunnel the best results regarding the weight of the minitubers were recorded at genotype ZIL19-02-43 (ave. 100.10 g), and under thermohydric stress conditions genotype GIL19-03-07 (ave. 10.15 g) was noted with a positive difference of 2.38 g compared to the control. On the variant in which 1 plantlet/pot was used, the genotype ZIL19-02-43 obtained the best results regarding the average weight of the minitubers (45.36 g), and on the variant with 2 plantlets/pot, genotype GIL19 03-07 (74.76 g) was highlighted.

In tunnel cultivation conditions, the obtained minitubers were mainly classified in the calibration classes > 30 mm and 25-30 mm for all genotypes and all planting variants. Under thermohydric stress conditions (greenhouse) the highest percentage of minitubers was noticed mainly in fractions of size 10-15 mm and 15-20 mm, for all 3 potato genotypes.

The larger size of minitubers that will be planted next year in field conditions, the more vigorous potato plants will be, and the higher production of tubers in the clonal field will be obtained.

The size of the minitubers influences duration of the germination period, the vigor of the seed tubers, number of stems, percentage of emergence, number of surviving plants, the vigor of the stems and their production capacity.

Regarding the production of minitubers obtained both in thermohydric stress conditions (greenhouse) and in optimal watering conditions but high temperatures (tunnel), all three genotypes obtained higher yields in tunnel compared to greenhouse. In potato cultivation, this aspect is similar to what happens in field conditions. If it is a year with high temperatures during the growing season, but the crop is irrigated, higher yields will be obtained than if the potato plants in addition to high temperatures, will be subjected also to water stress.

Minitubers (pre-basic seed) from *insect-proof* protected area will be used as planting material to monitor the behavior of potato genotypes in open field conditions.

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