

STUDY ABOUT THE INFLUENCE OF GIBBERELLIC ACID AND NAPHTHYL ACETIC ACID ON *IN VITRO* DEVELOPMENT OF FOUR ROMANIAN POTATO VARIETIES

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Abstract

*In 2024 within the Research Laboratory for Plant Tissue Cultures of National Institute for Research and Development for Potato and Sugar Beet Braşov, Romania an in vitro study was performed to determine the optimum concentration of gibberellic acid and naphthyl acetic acid for growth and development of plantlets belonging of four Romanian potato varieties, created at our institute. The minicuttings were cultured in Murashige and Skoog medium (MS). Gibberellic acid and naphthyl acetic acid were applied in different concentrations. The bifactorial experience of the 4*4 type included 16 variants, on 3 repetitions. Factor a is represented by the variety with 4 graduations (Azaria, Braşovia, Cosiana and Cezarina) and factor b is the culture medium with 4 graduations (0 mg GA₃: 0.005 NAA; 3 mg GA₃: 0.005 mg NAA; 4 mg GA₃: 0.01 mg NAA; 5 mg GA₃: 0.2 mg NAA). Measurements were made for: number of leaves, vitroplants height, root length and internodes number. For the analyzed parameters, the varieties behaved differently with each hormonal treatment applied.*

Key words: potato, in vitro, variety, gibberellic acid, naphthyl acetic acid.

INTRODUCTION

In the rapid multiplication of clean material *in vitro*, the use of single nodal cutting is the most preferred method of propagation since it ensures higher propagation rates with maximum genetic uniformity in potato (Chandra and Naik, 1993). Tissue culture techniques are used worldwide to produce pre-basic, virus-free seed potatoes. Plant growth regulators play an important role in this progress and have been studied extensively but the interactions that take place between them are still being discovered (Ross and O'Neill, 2001; Zhang et al., 2002; Rafique et al., 2012; Rafique et al., 2013; Rasheed et al., 2013). The plant growth regulators (PGRs) modulate plant growth and development and mediate responses to both biotic and abiotic stress (Makau et al., 2022). They are of importance in regulating shoot and root development in seed potatoes *in vitro* (Badoni & Chauhan, 2010; Hoque, 2010).

Gibberellic acid (GA₃) is a diterpenoid carboxylic acid that belongs to the gibberellin's family and acts as a natural plant growth

hormone. Plants and some microorganisms, such as fungi and bacteria, produce it. GA₃ has promising applications in the agro-industrial sector due to its properties related to plant development. Since it was discovered, studies have been focused on enhancing its process yield, boosting its productivity, and reducing its cost, which sometimes restricts the use of this important growth hormone (Camara et al., 2018). GA₃ is involved in cell elongation and its addition in MS medium enhances shoot growth (Camara et al., 2018; Rizza et al., 2017). The phytohormone gibberellin (GA) is a key regulator of plant growth and development (Rizza et al., 2017).

The basic Murashige & Skoog (MS) medium (Murashige & Skoog, 1962) is the most widely used media in production of potato (*Solanum tuberosum* L.). Since each individual hormone has its own unique effect on regeneration, it is vital to determine the combined effects of these on the regeneration of shoots and roots (Kumlay, 2014). The aim of this study was to establish the plant growth regulators concentration for plantlets development of four potato varieties.

MATERIALS AND METHODS

In 2024 within the Research Laboratory for Plant Tissue Cultures of NIRDPSB Braşov, Romania an *in vitro* study was performed to determine the optimum concentration of gibberellic acid and naphthyl acetic acid for growth and development of plantlets belonging to four Romanian potato varieties, created at our institute. The minicuttings were cultured in Murashige and Skoog (MS) media supplemented with sucrose; gibberellic acid and naphthyl acetic acid were applied, as follow in Table 1. Medium with 5 mg GA₃ and 0.2 mg NAA significantly enhanced shoot length, with the longest shoot (14.17 cm) as a mean for all values of varieties.

The treatment scheme followed the influence of the combination of gibberellic acid and naphthyl acetic acid on the *in vitro* development of the seedlings of the varieties under analysis. A 4 × 4 factorial design was used, in three replicates. Factor a is represented by the variety with 4 graduations (Azaria, Braşovia, Cosiana and Cezarina – control variety), and factor b is the culture medium with 4 graduations (according to treatment scheme from Table 1).

Table 1. Treatment scheme (Combination of gibberellic acid and naphthyl acetic acid)

Plant growth regulator	The amount introduced into the culture medium (mg/l)							
GA ₃	0	b ₁	3	b ₂	4	b ₃	5	b ₄
NAA	0.005		0.005		0.01		0.2	

RESULTS AND DISCUSSIONS

The results of the variance analysis showed that the effect of hormonal treatment was significant for plantlets height at the level <0.05 and for root length at the level <0.01. The influence of variety was significant for root length at the level <0.05 (Table 2).

The application in the culture medium of different concentrations of gibberellic acid and naphthyl acetic acid was favourable for Cezarina variety, which recorded the highest number values the number of leaves (8.50); the height of the seedlings (12.88 cm) and the number of internodes (6.58). About number of leaves and internodes, there were no significant differences, the values being between 8.50

(Cezarina variety) and 8.08 (Azaria, Braşovia, Cosiana) for the number of leaves and 6.58 (Cezarina variety) and 5.42 (Azaria variety) for number of internodes. Regarding the height of the plants between Braşovia (12.58 cm), Cosiana (12.83 cm) and Cezarina varieties (12.88 cm) there were no significant differences instead, the recorded values of the vitroplants height for these three previously mentioned varieties they differed significantly from Azaria variety (9.38 cm). For root length, there were significant differences between Cosiana (4.29 cm) and Azaria (2.96 cm) cultivars (Table 3).

Table 2. Analysis of variance in F values for determined parameters

Source of variation	df	Leaves number	Plantlets height	Root length	Number of internodes
Variety (a)	3	0.208 ns (4.76; 9.78)	3.401 ns	5.47 *	1.209 ns
Hormonal treatment (b)	2	0.877 ns (3.01; 4.72)	4.038 *	9.077 **	1.312 ns
Variety (a) * Hormonal treatment (b)	9	1.092 ns (2.30; 3.26)	0.980 ns	0.895 ns	1.007 ns

Table 3. The influence of variety on the studied parameters under the effect of the use of gibberellic acid and naphthyl acetic acid in the culture medium

Variety	Leaves number	Vitroplants height (cm)	Root length (cm)	Internodes number
Azaria	8.08 A	9.38 B	2.96 B	5.42 A
Braşovia	8.08 A	12.58 A	3.25 AB	6.08 A
Cosiana	8.08 A	12.83 A	4.29 A	6.25 A
Cezarina	8.50 A	12.88 A	2.54 B	6.58 A

Means of each factor designated by the same letter are not significantly different at 5% level using Duncan's Multiple Range Test.

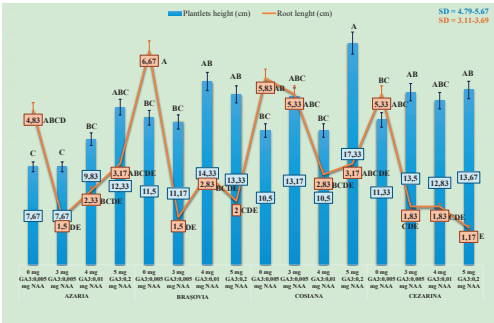
Treatment 4 (T4: 5 mg GA₃: 0.2 mg NAA) from treatment scheme was favourable for the following elements analysed in the experiment: number of leaves (8.92), but there were no significant differences between the four treatments and height seedlings (14.17 cm); for this parameter significant differences are observed between treatment 4 and treatments 2 and 3. Regarding the vitroplants root, the best result was obtained for treatment 1, with significant differences compared to the other treatments that had a negative influence, leading to a decrease in the length of the root system. Combination 3 mg GA₃: 0.005 mg NAA (T2) was beneficial for the formation of a large number of internodes (6.50), but without significant differences between treatments

(Table 4). According to Pennazio and Vecchiati, (1976) higher concentration of NAA inhibited root, form Table 4 we can observe that root growth decreased with increasing NAA concentration. Increasing the concentration of GA₃ leads to plantlets development, at higher concentration of GA₃ are obtained vitroplants with the greatest height (Table 4), suitable with Salisbury and Ross (1985) study, which mentions that gibberellic acid (GA₃) is a plant growth regulator that has the unique ability to promote extensive growth of many intact plants

Table 4. The influence of hormonal treatment on the studied parameters)

Hormonal treatment applied (mg/l)		Leaves number	Vitroplants height (cm)	Root lenght (cm)	Internodes number
GA ₃	NAA				
T1	0	0.005	7.58 A	10.25 B	5.67 A
T2	3	0.005	7.83 A	11.38 B	2.54 B
T3	4	0.01	8.42 A	11.88 AB	2.46 B
T4	5	0.2	8.92 A	14.17 A	2.38 B

Means of each factor designated by the same letter are not significantly different at 5% level using Duncan's Multiple Range Test.



Means of each parameter designated by the same letter are not significantly different at 5% level using Duncan's Multiple Range Test

Figure 1. The influence of variety and hormonal treatment applied over plantlets height (cm) and root length (cm)

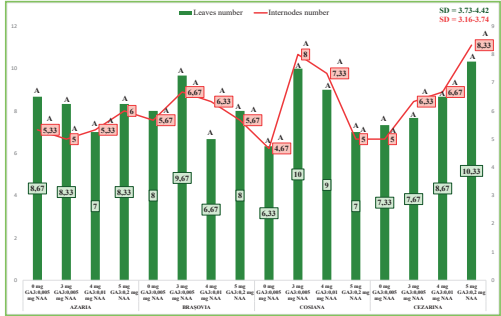
Each treatment had a different influence for the varieties studied, regarding the four parameters (Figures 1 and 2).

Combination of 5 mg GA₃: 0.2 mg NAA positively influenced Cosiana variety (Figure 1) regarding plantlets height, with the highest value being recorded for this element (17.33 cm). The lowest values of plantlets height (7.67 cm) were observed for Azaria cultivar for treatments 1 and 2.

Braşovia (6.67 cm) and Cosiana (5.83 cm) varieties presented the highest values of root

system length, for treatment 1, without significant differences (Figure 1).

From the analysis of the combined influence of the variety and the applied hormone combinations, there are no significant differences in the number of leaves and the number of internodes (Figure 2).



Means of each parameter designated by the same letter are not significantly different at 5% level using Duncan's Multiple Range Test

Figure 2. The influence of variety and hormonal treatment applied over leaves and internodes number

CONCLUSIONS

For the analysed parameters, the varieties behaved differently with each hormonal treatment applied.

The application in the culture medium of different concentrations of gibberellic acid and naphthyl acetic acid was favourable for Cezarina variety, which recorded the highest number values the number of leaves (8.50); the height of the seedlings (12.88 cm) and the number of internodes (6.58).

For root length, the highest value was recorded for Cosiana variety (4.29 cm).

Medium with 5 mg GA₃ and 0.2 mg NAA significantly enhanced shoot length, with the longest shoot (14.17 cm) as a mean for all values of varieties; this hormonal combination was beneficial in forming a high number of leaves (8.92).

Combination of 5 mg GA₃: 0.2 mg ANA positively influenced Cosiana variety (regarding plantlets height, with the highest value being recorded for this element - 17.33 cm).

Braşovia (6.67 cm) and Cosiana (5.83 cm) varieties presented the highest values of root system length, for combination 0 mg GA₃: 0.005 mg NAA, without significant differences

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