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GROWTH OF POTATO SHOOT CULTURES ON MEDIA WITH ANTIBIOTICS FOR ELIMINATION OF BACTERIAL CONTAMINATION

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The aim of our work was to evaluate the effect of selected antibiotics on the growth of potato shoot cultures in the Gene Bank of the Slovak Republic collection and to determine the type and dose that may be used to treat potato cultures endangered by endophytic bacteria. Antibiotics Chloramphenicol at doses 20, 50 and 100 mg/L, Gentamycin and Rifampicin in doses 20, 50, 100, 200 mg/L and the combination of Gentamycin and Rifampicin with 100 mg/L of each were used. Growth parameters – the shoot length and the number of nodal segments per shoot and rooting of ten cultivars of *Solanum tuberosum* L. of different origin were evaluated. Chloramphenicol already at the lowest dose had the strong inhibitory effect on regeneration, growth, and rooting of shoots. Gentamycin inhibited the growth of shoots gradually with increasing dose of it, rooting of shoots was negatively affected using the dose 50 mg/L or higher. Rifampicin up to 100 mg/L had the minimal effect on the shoots growth, rooting of shoots was not affected, but shoots were characterised by smaller or stunted leaves. Although the growth of shoots was affected, all ten genotypes used in the experiments were able to regenerate and grow at the highest dose of Rifampicin and Gentamycin. According to the results, it is highly probable that these antibiotics up to 100 mg/L or their combination would be suitable for culture preservation of the most genotypes in the gene-bank collection. On the other hand, Chloramphenicol cannot be recommended due to its strong detrimental effect on potato shoot cultures.

Key words: potato germplasm, *in vitro* conservation, nodal explants, Chloramphenicol, Gentamycin, Rifampicin

Cultivated potato (*Solanum tuberosum* L.) is, after cereals, one of the most important crops grown in temperate and subtropical climate throughout the world. This tuberous species belongs to family *Solanaceae* and is mainly used for human consumption and production of starch and ethanol. Cultivated potato is an annual tetraploid plant that is multiplied and maintained vegetatively through tubers to maintain their heterozygosity (Sarkar *et al*. 2011). Because propagation and conservation by tubers are laborious and associated with pathogen dissemination, several *in vitro* techniques have been developed for propagation, virus elimination, breeding including genetic modification, conservation and exchange of potato germplasm (Morais *et al*. 2018). The methods are labour- and space-saving, enable to store germplasm in controlled conditions free of pathogen attack and plant material can be multiplied in a short time (Sarkar *et al*. 2011).

In vitro conservation is the widespread method in potatoes gene banks over the world. There are several *in vitro* techniques used: cryopreservation that is suitable for long-term conservation (Kaczmarczyk *et al*. 2011; Faltus *et al*. 2011) and cultivation of shoot cultures (Westcott *et al*. 1977; Sarkar & Naik 1998) or microtubers induction and storage (Dobránszki

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et al. 2008) used for medium-term conservation. In the Gene Bank of the Slovak Republic, the potato germplasm is stored in the form of shoot cultures by a slow-growth method using growth retardant Daminozide (succinic acid 2,2-dimethylhydrazide). Slow-growth method enables unlimited storage of germplasm, but carries some risks including contamination of culture that can lead to its loss (Vinterhalter *et al*. 2008). Besides visually detectable contaminations caused by bacteria or fungi, latent infections triggered by endogenous bacteria are a serious problem in tissue cultures, because even slight changes in cultivation conditions may cause rapid proliferation of contaminants (Leifert & Cassells 2001) posing a threat to the culture vitality and growth (Orlikowska *et al*. 2017).

To control both types of bacterial contamination, the use of antibiotics may be required. But, besides the inhibition of bacterial growth, antibiotics can also affect explants survival and regeneration (Morais *et al*. 2018). β-lactam antibiotics that inhibit cell wall synthesis (e.g. Carbenicillin, Cefotaxime, and Ticarcillin) used to have a neutral or positive impact on explant regeneration of many plants species, including potato (Mahadev *et al.* 2014; Rákosy-Tican *et al.* 2011; Venkatasalam *et al.* 2013). They are most commonly used to eliminate *Agrobacterium tumefaciens* after genetic transformation, but have relatively weak bactericidal activities – bacterial growth was only suppressed and bacteria restarted proliferation after transfer to fresh medium without antibiotics (Ogawa & Mii 2005).

Therefore we tried to select antibiotics from other groups: Gentamycin that belongs to aminoglycosides inhibiting protein synthesis and Rifampicin that belongs to rifampin group inhibiting RNA synthesis. They were selected according to published papers with respect to their ability to inhibit the growth of bacteria in plant tissue cultures.

Ali *et al*. (2018) isolated nine bacterial strains (7 of them belong to species *Bacillus*) from contaminated explant cultures of potato and tea and performed sensitivity test to seven antibiotics. All bacteria were sensitive to Gentamycin and Streptomycin, eight of them to a low dose of Rifampicin. Rahman *et al*. (2017) treated contamination of potato culture in the bioreactor with three antibiotics in different dose and they obtained the best results with 50 mg/L of Gentamycin. Jena and Samal (2011) isolated bacterial strains from *in vitro* culture of another tuberous plant – sweet potato, and found out that all bacterial strains were susceptible to Gentamycin. Msogoya *et al*. (2012) confirmed the susceptibility of bacteria (*Klebsiella* spp., *Erwinia* spp., *Proteus* spp. and *Staphylococcus* spp.) isolated from explant culture of banana to 150 – 200 mg/L of Chloramphenicol, Gentamycin, and Rifampicin. Eziashi *et al*. (2014) also confirm the sensitivity of different bacterial species isolated from oil palm explants to Gentamycin and Rifampicin. Horáčková and Domkářová (1998) also mentioned about application of Gentamycin and Rifampicin during establishment of potato *in vitro* culture.

The aim of our work was to evaluate the effect of selected antibiotics on the growth of potato shoot cultures and to determine the type and dose that can be recommended for most genotypes in gene-bank collection. Growth parameters of potato shoot cultures grown on media with different concentrations or combination of these antibiotics were compared with chloramphenicol (belonging to macrolides) which had been used in our laboratory previously but used to have an adverse effect on the most of conserved genotypes. Our hypothesis was that Rifampicin or Gentamycin would not seriously inhibit the growth of shoots of potato and at least one of them may be used to treat potato *in vitro* cultures.

MATERIAL AND METHODS

For experiments, shoot cultures of potato genetic resources collection were used. Ten genotypes of different origin, to ensure genetic variability among them, were randomly selected: Linzer Delicates (AUT), Imperia (SWE), Sázava (CZ), Breza (SK), King Edward (GB), Lady Florina (NL), Vesna (SVN), Inovator (NL), Fanchette (FR), Lyra (DEU).

To ensure homogenity of initial material, the 3rd and 4th nodal segment from shoots with 6 – 8 nodal segments were inoculated to testing media with or without (control variant) antibiotics. Each nodal segment was cultivated individually in a test tube 17 × 160 mm. Antibiotics were added to modified MS medium containing MS salts (Murashige & Skoog 1962), 192.2 mg/L $NaH_2PO_4.2H_2O$, 100 mg/L inosi-

tol, 30 g/L sucrose, 0.4 mg/L thiamine HCl, 2 mg/L calcium pantothenate and 8 g/L agar. In the first experiment, the antibiotics Gentamycin, Rifampicin and Chloramphenicol in doses 20, 50, and 100 mg/L, and in the second experiment, 200 mg/L of Gentamycin or Rifampicin and the combination of Gentamycin and Rifampicin at 100 mg of both were used. Tubes (40 tubes per each antibiotic variant) were cultivated under the photoperiod 16 h light/8 h dark with the light intensity 70 µmol/m^2/s and temperature 24/20°C.

After 3 weeks of cultivation, the growth of shoots was evaluated by the number of nodal segments, shoot length and rooting frequency. Results were statistically analysed by the analysis of variance (ANOVA) and means were then separated by *LSD* test (the least significant difference) at $\alpha = 0.05$ using the statistical software STATGRAFICS Centurion XVI.II.

RESULTS AND DISCUSSION

The experiments were designed to find antibiotics useable for the cultivation of potato shoot cultures without the strong detrimental effect on their growth. In the first experiment the effect of antibiotics Gentamycin, Rifampicin and Chloramphenicol at concentrations 20, 50 and 100 mg/L on shoot growth was evaluated. There were observed statistically significant differences in shoot length (Table 1a) and the number of nodal segments (Table 1b) among genotypes and antibiotic treatments at $\alpha = 0.05$. Chloramphenicol already at the lowest dose had a strong inhibitory effect on regeneration of shoots in comparison with other antibiotics or control variant. Genotypes that best tolerated it were Lyra and Inovator. Rooting of shoots was also affected, only 15% of shoots rooted at the lowest dose of Chloramphenicol, and there were no rooting on the higher dose observed. Addition of Gentamycin inhibited the growth of shoots moderately compared to control variant. There were no statistically significant differences among its dose used neither for shoot length nor for the number of nodal segments. Rooting of shoots reached 80% at the lowest dose, and 5 or 2.5% at 50 and 100 mg/L respectively. In the case of Rifampicin, there was minimal effect on the shoots growth, and rooting reached 100% like for control variant. But in terms of plant morphology, shoots grown on the medium with Rifampicin were characterized by smaller or stunted leaves (Figure 1a) in six of tested genotype, whereas on Gentamycin such effect was observed only for genotype Lady Florina.

Because the shoots were able to grow on media with Gentamycin and Rifampicin in all tested doses, in the second experiment the concentration of these antibiotics were doubled to 200 mg/L and the combination of both antibiotics with 100 mg/L of each was tested. While the average shoots length on medium with 100 mg/L of Rifampicin was not changed significantly, using 200 mg/L, the shoot length was lowered 1.75-fold (Table 2a, Figure 1b). In the case of Gentamycin, both growth parameters decreased gradually. As for the number of nodal segments, all variants with Rifampicin gave in average better re-

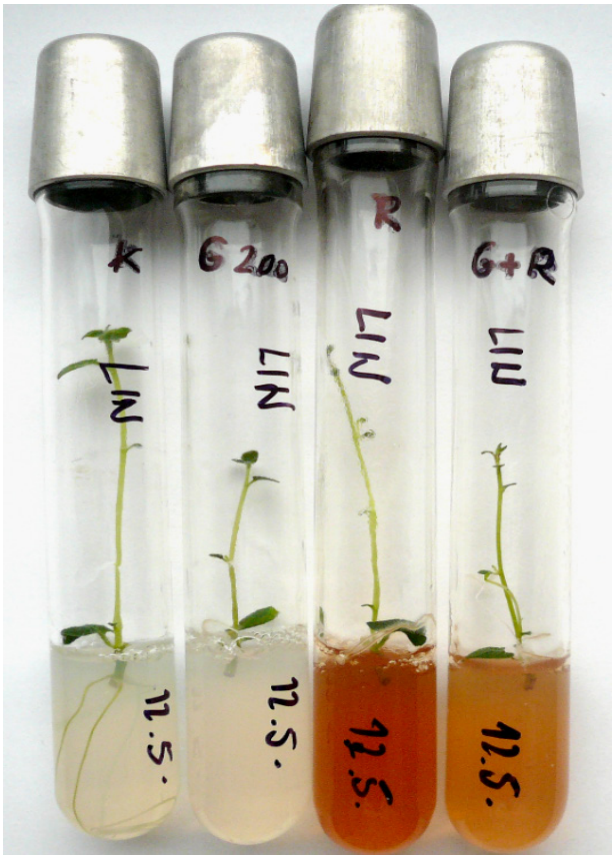

Figure 1b. Shoot culture of *Solanum tuberosum*, cultivar Linzer Delicates, cultivated on media with antibiotics; variants (from left): control, Gentamycin 200 mg/L, Rifampicin 200 mg/L and combination of Gentamycin and Rifampicin in dose 100 mg/L of each

Table 1a

Length of *Solanum tuberosum* shoots cultivated on media with antibiotics

Cultivar	Control	G20	G50	G100	R20	R50	R100	Ch20	Ch50	Ch100
L. Delikates	45.50 ± 7.33	29.25 ± 10.44	22.50 ± 7.94	21.50 ± 10.41	68.50 ± 4.65	64.75 ± 10.31	59.00 ± 6.68	11.25 ± 6.13	3.25 ± 4.72	1.25 ± 2.50
Imperia	103.00 ± 26.46	72.50 ± 9.98	80.00 ± 11.34	63.50 ± 10.34	81.00 ± 4.97	77.00 ± 6.83	80.00 ± 9.97	32.50 ± 9.15	4.50 ± 5.45	7.50 ± 7.59
Sázava	82.25 ± 11.53	54.25 ± 9.74	37.00 ± 7.62	34.25 ± 12.20	80.50 ± 9.95	69.50 ± 6.14	66.00 ± 14.76	11.25 ± 5.80	2.75 ± 2.06	0.75 ± 0.96
Breza	83.25 ± 16.96	52.50 ± 7.59	45.75 ± 11.90	50.00 ± 7.35	80.75 ± 6.90	78.50 ± 9.95	92.00 ± 10.42	15.00 ± 10.89	5.75 ± 5.91	0.00 ± 0.00
King Edward	77.50 ± 11.27	42.75 ± 7.80	43.25 ± 18.82	32.75 ± 16.32	79.25 ± 8.85	84.25 ± 13.77	81.00 ± 9.20	19.25 ± 3.40	3.25 ± 3.40	1.25 ± 1.50
Lady Florina	67.00 ± 10.86	40.25 ± 12.50	51.75 ± 12.82	61.50 ± 14.18	76.75 ± 12.97	75.00 ± 8.60	80.75 ± 24.68	5.00 ± 3.74	1.25 ± 1.50	0.75 ± 0.96
Vesna	82.25 ± 19.09	67.00 ± 15.19	74.25 ± 10.08	68.00 ± 16.83	82.00 ± 17.57	86.50 ± 24.37	83.50 ± 7.59	23.50 ± 22.81	0.75 ± 0.96	0.25 ± 0.50
Inovator	94.25 ± 7.23	92.50 ± 7.00	75.50 ± 27.21	55.00 ± 16.71	88.00 ± 24.17	70.75 ± 13.25	85.75 ± 14.20	34.75 ± 11.70	19.25 ± 16.88	2.25 ± 3.86
Fanchette	58.25 ± 7.80	57.50 ± 17.08	51.75 ± 4.03	65.50 ± 16.98	60.00 ± 12.83	59.50 ± 6.19	56.50 ± 13.40	8.50 ± 4.80	1.75 ± 0.96	0.50 ± 1.00
Lyra	74.00 ± 11.75	52.00 ± 8.60	52.50 ± 12.77	55.00 ± 7.02	93.50 ± 7.94	67.25 ± 15.63	65.75 ± 14.10	35.25 ± 10.81	26.75 ± 29.48	8.25 ± 5.19
Average	76.73 ± 15.87^{a}	56.05 ± 17.02^{b}	53.43 ± 17.38^{b}	50.70 ± 15.11^{b}	79.03 ± 8.87^{a}	73.30 ± 8.12^{a}	75.03 ± 11.56^{a}	19.63 ± 10.74^{c}	6.93 ± 8.33^{d}	2.28 ± 2.87^{d}

Legend: means ± SD; G – Gentamycin, R – Rifampicin; Ch – Chloramphenicol – the number after letter indicates the dose of antibiotic in mg/L; different letters in the last row indicate statistically significant differences evaluated by *LSD* test at $\alpha = 0.05$

Table 1b

Number of nodal segments/shoot of *Solanum tuberosum* cultivated on media with antibiotics

Cultivar	Control	G20	G50	G100	R20	R50	R100	Ch20	Ch50	Ch100
L. Delikates	5.75 ± 0.96	4.25 ± 0.50	3.75 ± 0.50	3.50 ± 0.58	5.75 ± 0.50	5.75 ± 0.50	5.75 ± 0.50	3.00 ± 1.83	1.25 ± 1.89	0.50 ± 1.00
Imperia	6.25 ± 1.89	5.25 ± 0.50	5.25 ± 0.50	4.50 ± 0.58	6.00 ± 0.82	5.50 ± 0.58	6.00 ± 0.82	5.50 ± 1.00	1.75 ± 1.71	1.75 ± 1.71
Sázava	6.75 ± 0.50	6.25 ± 0.96	5.50 ± 0.58	5.00 ± 0.82	8.00 ± 0.82	8.00 ± 0.82	8.25 ± 0.96	3.25 ± 2.06	0.75 ± 0.50	0.50 ± 0.58
Breza	7.25 ± 0.50	5.50 ± 1.29	5.50 ± 1.91	6.25 ± 0.96	8.25 ± 0.50	8.00 ± 0.82	8.00 ± 1.41	4.00 ± 2.58	1.75 ± 2.22	0.00 ± 0.00
King Edward	6.75 ± 0.50	5.50 ± 0.58	5.25 ± 0.50	4.50 ± 0.58	6.00 ± 0.82	6.50 ± 0.58	6.75 ± 0.96	4.75 ± 0.96	1.50 ± 1.73	0.50 ± 0.58
Lady Florina	6.75 ± 0.50	5.00 ± 0.82	5.25 ± 0.96	6.25 ± 0.96	7.25 ± 0.96	6.75 ± 0.50	7.25 ± 0.96	1.75 ± 1.71	0.50 ± 0.58	0.50 ± 0.58
Vesna	6.00 ± 0.82	5.50 ± 1.29	5.75 ± 1.50	5.25 ± 1.26	6.75 ± 0.96	6.00 ± 1.15	6.50 ± 1.29	3.75 ± 2.63	0.50 ± 0.58	0.25 ± 0.50
Inovator	5.75 ± 0.96	4.75 ± 0.50	4.75 ± 0.96	5.00 ± 0.82	6.25 ± 0.50	7.25 ± 0.50	7.25 ± 1.26	6.00 ± 0.82	4.25 ± 1.50	1.00 ± 1.41
Fanchette	6.25 ± 0.50	5.00 ± 0.00	5.00 ± 0.82	4.75 ± 0.50	6.50 ± 0.58	6.50 ± 1.29	7.25 ± 1.26	4.50 ± 1.00	3.25 ± 1.26	3.00 ± 1.83
Lyra	6.25 ± 0.50	5.00 ± 0.00	5.00 ± 0.82	4.75 ± 0.50	6.50 ± 0.58	6.50 ± 1.29	7.25 ± 1.26	4.50 ± 1.00	3.25 ± 1.26	3.00 ± 1.83
Average	6.38 ± 0.4^{b}	5.20 ± 0.51^{c}	5.10 ± 0.53^{c}	4.98 ± 0.78^{c}	6.73 ± 0.81^{ab}	6.68 ± 0.81^{ab}	7.03 ± 0.75^{a}	4.10 ± 1.18^{d}	1.88 ± 1.23^{e}	1.10 ± 1.05^{f}

Legend: means ± SD; G – Gentamycin; R – Rifampicin, Ch – Chloramphenicol – the number after letter indicates the dose of antibiotic in mg/L; different letters in the last row indicate statistically significant differences evaluated by *LSD* test at $\alpha = 0.05$

T a b l e 2a

Length of *Solanum tuberosum* shoots cultivated on media with antibiotics

Cultivar	Control	G200	R200	G100+R100
L. Delikates	59.50 ± 8.43	32.25 ± 6.95	48.00 ± 8.76	37.25 ± 3.86
Imperia	95.00 ± 8.52	58.25 ± 10.01	54.00 ± 7.53	58.25 ± 12.28
Sázava	80.50 ± 23.46	27.75 ± 14.89	36.50 ± 6.19	30.75 ± 20.02
Breza	81.80 ± 26.29	48.67 ± 18.15	38.67 ± 5.13	50.75 ± 8.54
King Edward	109.00 ± 10.71	48.50 ± 15.42	60.00 ± 7.62	66.50 ± 12.29
Lady Florina	104.25 ± 6.95	35.00 ± 20.51	46.00 ± 5.48	56.75 ± 9.54
Vesna	81.00 ± 50.19	29.75 ± 15.88	48.50 ± 4.20	53.75 ± 19.19
Inovator	96.25 ± 4.19	29.25 ± 18.17	60.75 ± 9.43	55.25 ± 12.89
Fanchette	78.50 ± 13.08	56.75 ± 4.99	53.50 ± 5.00	54.25 ± 17.37
Lyra	99.75 ± 9.74	56.00 ± 21.80	60.25 ± 4.79	46.25 ± 7.23
Average	88.56 ± 14.15[a]	42.22 ± 11.93[c]	50.62 ± 8.23[b]	50.98 ± 9.92[b]

Legend: means ± SD; G – Gentamycin, R – Rifampicin – the number after letter indicates the dose of antibiotic in mg/L; different letters in the last row indicate statistically significant differences evaluated by *LSD* test at $\alpha = 0.05$

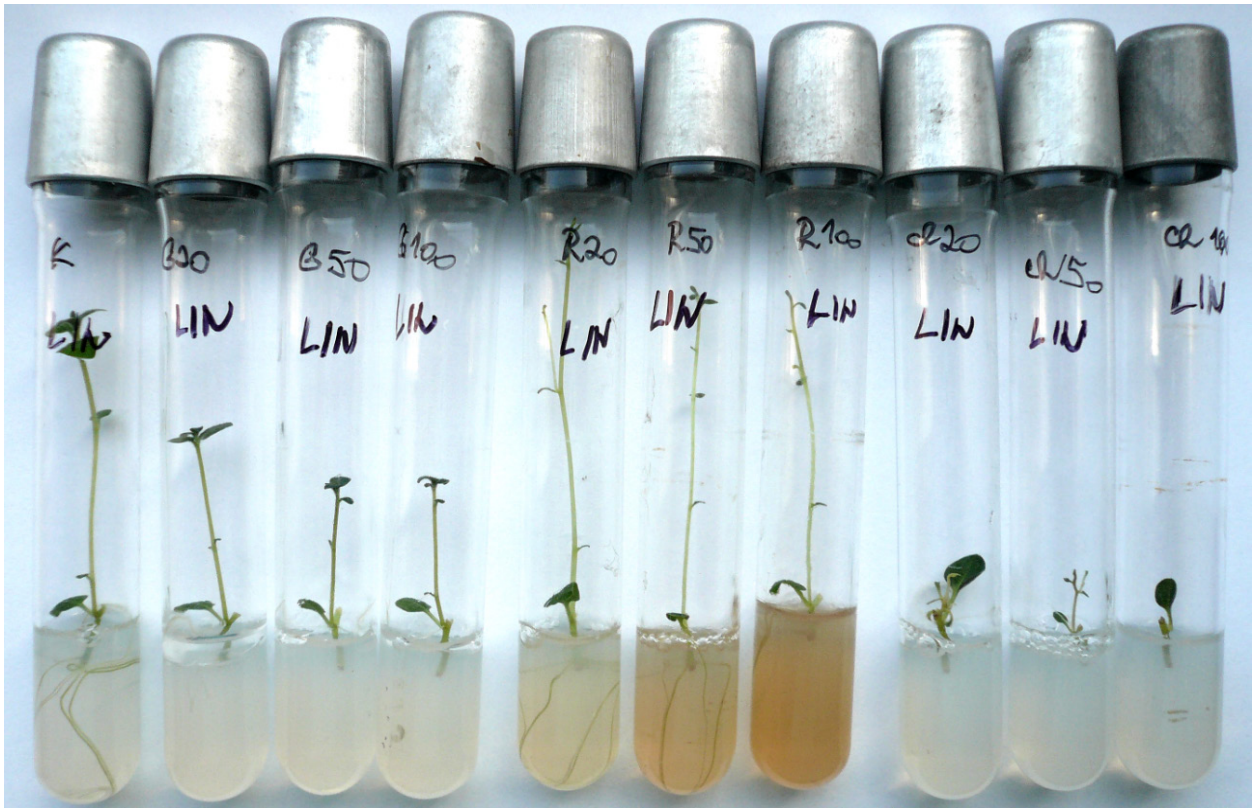

Figure 1a. Shoot culture of *Solanum tuberosum*, cultivar Linzer Delicates, cultivated on media with antibiotics; variants (from left): control, Gentamycin 20, 50, 100 mg/L, Rifampicin 20, 50, 100 mg/L and Chloramphenicol 20, 50 and 100 mg/L

sults than the control variant (Table 1b, 2b). Combination of 100 mg/L Rifampicin and Gentamycin gave a similar result like 200 mg/L of Rifampicin alone for shoot length, although the number of nodal segments slightly decreased. The rooting of shoots was 95% for 200 mg/L of Rifampicin, but Gentamycin had a negative impact on rooting also in the combination with Rifampicin.

Figure 2 summarizes both experiments where the differences among all used doses of antibiotics in both experiments relative to control variant, that is considered to be 100%, are illustrated. These results

T a b l e 2b

Number of nodal segments/shoot of *Solanum tuberosum* cultivated on media with antibiotics

Cultivar	Control	G200	R200	G100+R100
L. Delikates	6.00 ± 0.00	4.00 ± 0.82	6.00 ± 0.82	6.50 ± 1.00
Imperia	4.75 ± 0.50	3.50 ± 0.58	5.00 ± 0.00	5.00 ± 0.82
Sázava	6.00 ± 0.82	4.25 ± 1.71	8.00 ± 1.41	5.25 ± 2.87
Breza	6.60 ± 0.89	4.67 ± 1.15	8.00 ± 1.00	7.00 ± 0.82
King Edward	5.50 ± 0.58	4.75 ± 0.96	6.25 ± 1.26	6.50 ± 1.00
Lady Florina	5.25 ± 0.50	3.25 ± 0.50	5.50 ± 0.58	6.00 ± 0.82
Vesna	4.50 ± 1.00	3.00 ± 0.82	6.50 ± 0.58	5.75 ± 1.26
Inovator	5.25 ± 0.96	3.50 ± 1.29	6.50 ± 0.58	5.50 ± 1.29
Fanchette	5.75 ± 0.96	3.75 ± 0.50	6.25 ± 0.96	5.75 ± 0.96
Lyra	6.00 ± 0.82	4.50 ± 1.29	7.50 ± 1.00	6.50 ± 1.00
Average	5.56 ± 0.61^{b}	3.92 ± 0.58^{c}	6.55 ± 0.95^{a}	5.98 ± 0.61^{ab}

Legend: means ± SD; G – Gentamycin, R – Rifampicin – the number after letter indicates the dose of antibiotic in mg/L; different letters in the last row indicate statistically significant differences evaluated by *LSD* test at $\alpha = 0.05$

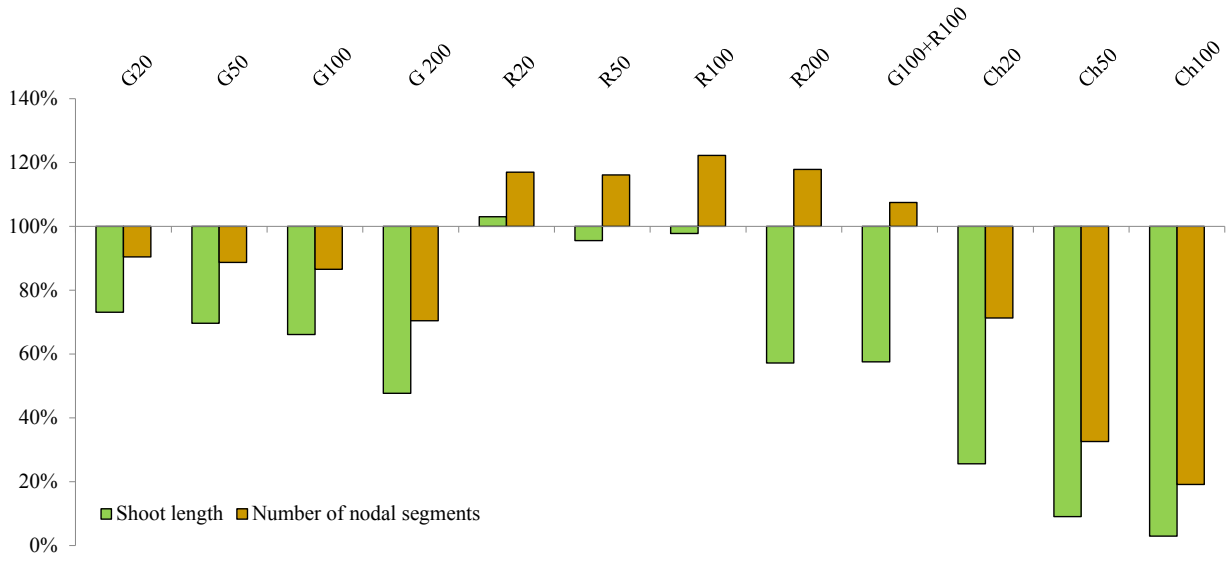

Figure 2. Growth parameters of *Solanum tuberosum* shoot cultures cultivated on media with antibiotics relative to control variant that is considered to be 100%; G – Gentamycin, R – Rifampicin, Ch – Chloramphenicol – the number after letter indicates the dose of antibiotic in mg/L

confirm the hypothesis that antibiotics Gentamycin and Rifampicin may be used for the cultivation of potato *in vitro* cultures in the case they are contaminated by bacteria. The use of Chloramphenicol had a detrimental effect on explant regeneration and growth and is not suitable for most of the potato genotypes.

There are only several reports concerning this problem in potato explant cultures. Rákosy-Tican *et al*. (2011) cultivated shoots of *Solanum chacoense* with Cefotaxime and confirmed the stimulating effect of this antibiotic mainly on root length and leaf fresh weight. Mahadev *et al*. (2014) confirmed a positive effect of Cefotaxime on the culture of *Solanum viarum*. Venkatasalam *et al*. (2013) compared the growth of three potato genotypes on Cefotaxime, Carbenicillin and Streptocycline (combination of 90% streptomycin sulphate and 10% tetracycline hydroxide) and found out positive effect of Carbenicillin up to 100 mg/L and Cefotaxime up to 200 mg/L on growth parameters, and negative effect of all used concentration of Streptocycline (100 – 250 mg/L). On the other hand, Buckseth *et al.* (2017) used Streptomycin and Gentamycin and observed improvement of plantlets vigour in potato culture using 100 and 200 mg/L of Streptomycin. Gentamycin at 10 or 25 mg/L had a positive or neutral effect on growth parameters, however higher concentration had a negative impact on all four genotypes studied. Such effect of Gentamycin is in concordance with our results. To our knowledge, the effect of Rifampicin on potato shoot culture has not yet been published. Its effect on shoot growth rate was positive in our experiment, but the morphology of shoots was changed compared to Gentamycin that only slowed plantlets growth. To our observation, these changes in morphology seem to be reversible, however, this has to be checked seriously for more genotypes. Chloramphenicol had the detrimental effect in all concentration used, but for other species it may be different. For example, Amissah *et al*. (2016) described that 500 mg/L of Chloramphenicol had eradicated the contamination in the culture but minimally inhibited growth of plantlets of sweet potato.

Nevertheless, the tolerance of explants to antibiotics or other additives in culture media is species and genotype-dependent and needs optimization in each individual case. To summarize the results, both, Gentamycin and Rifampicin up to 100 mg/L or their combination had not toxic effect on potato plantlets. Together with Cefotaxime, the positive effect of which has been confirmed in many plant species, as well as in potatoes, these antibiotics may be used to treat bacterial contamination in potato shoot cultures. Ultimately, for each genotype of potato and mainly for each contaminating bacteria the proper antibiotic and its dose has to be determined due to the bacterial resistance or sensitivity to different antibiotics.

CONCLUSIONS

Addition of antibiotics affected shoot regeneration from nodal segments of potato. Using of Chloramphenicol had a strong inhibition effect on all tested parameters – the number of nodal segments, shoot length, and rooting of shoots. Addition of Gentamycin decreased the number of nodal segments and shoot length gradually with the increasing dose of it, rooting of shoot was negatively affected using the dose 50 mg/L or higher. Rifampicin had a minimal effect on shoot growth and rooting, the number of nodal segments was even higher compared to control. Morphology of shoots was not changed in 9 of 10 genotypes using Gentamycin, in one genotype smaller leaves were observed. But such negative effect on morphology was observed in 6 genotypes using Rifampicin. Summarizing the results, the antibiotics Rifampicin and Gentamycin in dose up to 100 mg/L or their combination can be used to treat potato shoot cultures contaminated with bacteria, but the reaction of explants may be affected by genotype. Since all 10 genotypes used in the experiments were able to grow at the highest dose of these antibiotics, it is highly probable that they would be suitable for most genotypes in the collection. The bactericidal or bacteriostatic effect of these antibiotics is, of course, dependent on the sensitivity or resistance of the bacterial species occurred in the culture to selected antibiotics.

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