

Effect of Different Media Additives on *In vitro* Culture of Potato (*Solanum tuberosum*)

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Abstract

Effect of different media additives on *in vitro* culture of three potato varieties viz. Lalpakri, Cardinal and BARI TPS-1 was investigated using single nodes as explant. The single nodes were collected from microplants of tuber sprouts that was preincubated under dark. Survivability of the cultures was statistically insignificant in respect of the media used, but the highest percentage of cultures survived on MS medium supplemented with 60 mg L⁻¹ inositol. The time taken to initiate new shoots was minimum on MS medium supplemented with 1 mg L⁻¹ Benzyl Amino Purine (BAP). The number of shoot was maximum on a medium containing MS + 0.25 mg L⁻¹ Gibberellic Acid (GA₃) and 2 mg L⁻¹ Calcium Pantothenate. The maximum number of roots was obtained on medium containing MS + 0.03 mg L⁻¹ Naphthalene Acetic Acid (NAA). Among the varieties tested, BARI TPS-1 performed better in respect of single node culture. The interaction between different potato varieties and media compositions was significant for different characters of *in vitro* grown plantlets.

Key Words: Growth rate, *In vitro* culture, Media additives, Growth regulator, Potato.

Introduction:

The potato grown in Bangladesh as a vegetable crop is also considered as the substitute of rice. Potato ranks first both in acreage and production among the vegetable crops grown in Bangladesh. However, the average yield of this crop in Bangladesh is very low (10 tons ha⁻¹) compared to the average yield of the world (15.18 tons ha⁻¹) (FAO, 1995). The yield of potato depends on many factors including seed quality, presence of pathogen, variety, management practices etc. Tissue culture techniques are valuable tool for improving potato production through the cultivation of pathogen free plant material (Roca *et al.*, 1979).

In vitro propagation of potato is carried out using axillary buds, stem cuttings or tuber sprouts as explants and Hussey and Stacey (1981) reported very high multiplication rates. The rate of multiplication in "*in vitro*" condition depends on large extent to the composition of medium. The type and concentration of growth regulators, vitamins etc. are some of the important factors for success of such a programme. The node production can also be enhanced by the addition of GA₃ (Firmin, 1984). Cytokinins and auxins can be used to increase the number of shoots and roots growth (Hussey, 1980). *In vitro* growth of potato also depends on the genotype of the material used (Hoque *et al.*, 1996). The present study was undertaken with the view to find out the more effective vitamins and/or growth hormones as media additives for suitable *in vitro* culture of three potato varieties.

Materials and Methods

The present study was conducted to investigate the effect of different media additives, specially some vitamins and hormones on *in vitro* culture of three potato varieties viz., Lalpakri, Cardinal and BARI TPS-1. Potato tubers were collected from healthy and disease free plants grown at the Horticulture Farm, Bangladesh Agricultural University,

Mymensingh. Single node cuttings of three potato varieties, were used as explants. Single nodes were collected from microplants of tuber sprouts that was preincubated under dark.

Murashige and Skoog (MS) (1962) medium was used supplemented with 0.8% agar, 3% sucrose and with varying hormones and vitamins as indicated elsewhere. The pH of the media was adjusted to 5.8 prior to autoclaving at 121°C for 20 minutes at 1.16 kg cm⁻². Potato sprouts of 0.5 ± 0.2 cm in length consisting of a single node were isolated from the microplants with sharp scalpel and the leaf attached to the node was excluded. Isolated single nodes were surface sterilized and inoculated aseptically to baby meal jars, each containing 20 ml of medium. The cultures were incubated in a growth room at a temperature of 25° ± 2°C, illuminated with white cool fluorescent tubes (4.83 fc 84 TDL/Phillips). The room was illuminated 16 hours daily with a light intensity of 1500 lux.

Treatments: Different treatments of the experiment were as follows:

I. Culture media

T₁ = MS + 1 mg L⁻¹ BAP

T₂ = MS + 0.25 mg L⁻¹ GA₃ + 2 mg L⁻¹ Calcium Pantothenate

T₃ = MS + 0.03 mg L⁻¹ NAA

T₄ = MS + 60 mg L⁻¹ inositol

T₅ = MS + 30 mg L⁻¹ Adenine Sulphate

II. Potato varieties:

V₁ = Lalpakri

V₂ = Cardinal

V₃ = BARI TPS-1

There were five replications for each treatment. Data on per cent survivability of cultures, time taken to initiation of new shoots and roots and their number, height of the plantlets (cm), number of leaves and nodes, root length (cm), growth rate (mm/d) and vigour of the plantlets were recorded. The vigour of the plantlets was measured on assumption within a scale of 1 to 10. The data for the characters under study were analysed and evaluated by F-variance test. The significance of differences between pair of means was evaluated by Least Significance Difference (LSD) test (Gomez and Gomez, 1976).

Results and discussions

The effects of different media additives, different vitamins and growth hormones, indicate that they had different effects in respect of the characteristics of *in vitro* plantlets. The media supplemented with 60 mg L⁻¹ inositol (T₄) performed better than others. Survivability of the cultures was statistically insignificant in respect of the media used, but highest percentage (87.2%) of cultures survived on a MS medium supplemented with 60 mg L⁻¹ inositol (Table 1). Development of shoot was earlier (2.5 days) in the medium supplemented with 1 mg⁻¹ BAP.

Table 1. Effect of media additives on *in vitro* performance of single node culture of potato

Treatments	Surviv-ability (%)	Days to new shooting	Days to new rooting	No. of roots/ plantlet	Root length (cm)	Growth rate (mm/d)	Vigour (scale 1-10)
T ₁	80.46	2.54	8.54	3.90	2.04	2.07	6.17
T ₂	78.10	3.16	6.12	3.93	4.90	2.55	6.87
T ₃	76.98	3.01	5.91	5.04	5.32	2.39	7.77
T ₄	87.17	3.30	3.76	4.54	7.26	3.09	8.20
T ₅	84.69	3.50	5.98	4.65	4.47	2.45	7.07
LSD at 5%	Insignificant	Insignificant	0.99	0.05	0.02	0.39	0.27

But minimum days (3.76 days) required for new rooting in T₄, which was due to the improvement of growth of the cultures by inositol. Number of root was highest (5.04) in T₃ (MS+0.03 mg⁻¹ NAA) and higher root length (7.26cm) was in T₄. Resende and Paiva (1986) and Letal *et al.* (1985) also observed the enhancing effect of NAA on root formation of potato in “*in vitro*” condition. Hezsky *et al.* (1983) and Chavdarov *et al.* (1983) found higher root length in potato plantlets when the media contained GA₃ and IBA at lower concentration. The later results are not consistent with the present findings, which might be due to the promotion of growth by higher concentration of inositol than lower concentration of GA₃ and IBA. Maximum growth rate (3.09 mm/d) and more vigours plantlets were found in the media containing inositol. Inositol influenced the rapid growth of the plantlets. These results are consistent with the results of Robert *et al.* (1980). The rate of increment in height of the plantlets, number of shoots, number of nodes and number of leaves was also influenced by media (Fig. 1). Increment in length of shoot was more rapid in T₄. But number of shoot and node increased rapidly in T₂ (MS + 0.25 mg⁻¹ GA₃ + 2 mg⁻¹ calcium pantothenate). Obordo and Zamora (1988) also found rapid increment in number of shoot and node on medium supplemented with GA₃ and calcium pantothenate. Rate of increment in number of leaves was more or less similar in T₂, T₃ and T₄. The results on effect of varieties on *in vitro* growth of potato have been presented in Table 2. Significant difference was found among the varieties in respect of *in vitro* characteristics under study. Among the varieties BARI TPS-1 (V₃) performed better followed by Lalpakri (V₁).

Table 2. Main effect of varieties on *in vitro* performance of single node culture of potato

Variety	Survivability (%)	Days to new shooting	Days to new rooting	No. of roots/ plantlet	Root length (cm)	Growth rate (mm/d)	Vigour (scale 1-10)
Lalpakri	79.76	3.13	7.23	5.40	3.75	2.63	6.17
Cardinal	83.66	3.47	4.73	2.05	3.79	1.74	7.83
HPSII-67	81.01	2.71	6.23	5.80	6.86	3.16	7.77
LSD at 5%	Insignificant	0.52	0.77	0.04	0.02	0.31	0.21

Table 3. Interaction between varieties and media additives on *in vitro* culture of potato

Treatments	Survivability (%)	Days to new shooting	Days to new rooting	Height of the plantlets (cm)	No. of shoots/ plantlet	No. of roots/ Plantlet	Root length (cm)	No. of nodes/ plantlet	No. of leaves/ plantlet	Growth rate (mm/d)	Vigour (scale 1-10)
T ₁											
Lalpakri	89.19	2.06	8.43	5.37	1.26	3.89	1.68	8.39	10.12	2.69	5.50
Cardinal	67.80	3.29	8.23	2.25	1.15	1.22	1.82	2.81	3.65	2.66	6.17
BARI TPS-1	71.51	2.93	8.20	4.39	2.89	6.59	2.62	9.50	12.09	1.71	6.73
T ₂											
Lalpakri	87.02	3.46	4.40	5.33	2.13	4.09	3.87	11.46	13.27	3.48	7.27
Cardinal	83.27	3.90	6.90	3.02	1.34	1.40	2.18	3.78	4.91	2.59	5.17
BARI TPS-1	73.64	3.13	5.07	6.96	3.84	6.29	8.64	17.38	20.83	1.13	6.77
T ₃											
Lalpakri	80.01	3.67	4.57	3.43	1.23	6.87	1.85	7.46	9.60	1.51	7.23
Cardinal	86.84	3.77	4.51	3.56	1.17	2.97	6.60	4.71	6.59	1.78	7.87
BARI TPS-1	84.34	3.53	3.93	7.37	2.73	5.33	7.80	14.40	21.10	2.56	8.63
T ₄											
Lalpakri	93.49	3.23	8.57	6.96	1.54	5.03	9.07	11.17	13.20	1.72	8.10
Cardinal	78.53	2.43	12.12	5.12	1.16	2.56	4.99	6.39	7.71	2.39	6.27
BARI TPS-1	86.48	2.53	5.56	6.43	2.60	6.06	7.73	11.59	15.59	3.48	7.20
T ₅											
Lalpakri	72.60	2.32	5.03	5.13	1.63	7.10	2.57	9.99	10.86	3.68	8.70
Cardinal	90.14	2.90	2.95	3.27	1.25	2.13	3.34	3.85	5.21	3.22	8.67
BARI TPS-1	77.29	3.36	5.46	6.09	2.43	4.73	7.49	11.96	15.29	3.02	7.93
LSD at 5%	16.21	NS	1.72	0.04	0.04	0.09	0.04	0.04	0.04	0.69	0.47

T₁ = MS+1mg⁻¹ BAP, T₂= MS+0.25mg⁻¹ calcium pantothenate, T₃= MS+0.03 mg⁻¹ NAA, T₄= MS+ 60mg⁻¹ inositol, T₅= MS+ 30mg⁻¹ adenine sulphate

The varieties did not differ significantly in case of survivability of the cultures. Hossain (1988) also found that different varieties have no effects on survivability of the cultures. Minimum days

(2.71 days) were required for shoot initiation in BARI TPS-1 and for new rooting (4.73 days) in Cardinal (V₂). Maximum number of roots (5.80) with higher root length (6.80cm) was found in the plantlets of BARI TPS-1. Maximum growth rate (3.16 mm/d) and more vigours plantlets were also obtained in BARI TPS-1. According to Kollist and Tikke (1994), regenerative ability of potato explants depends on variety. Similar results were also found by Bajaj (1981) while he was working with potato varieties. Rapid increment in height of the plantlets, number of shoots, nodes and leaves was also found in the plantlets of BARI TPS-1 (Fig.2). The plantlets of Cardinal had slow growth in respect of those parameters mentioned above.

Significant interaction was observed between media additives and potato varieties. Maximum number of plantlets (93.49%) was obtained in the treatment combination of Lalpakri x T₄ (MS+60 mg⁻¹ inositol). Minimum days (2.95 days) were required for new rooting in Cardinal grown on T₅ (MS+30 mg⁻¹ adenine sulphate) and maximum number of root (7.10) was found in Lalpakri grown on the same media (Table 3). This was due to the effect of adenine sulphate on root initiation, which agrees with the report of Robert *et al.* (1980). Maximum root length (9.07 cm) was found in Lalpakri grown on T₄ and maximum growth was also found in the same treatment combination. More vigours plantlets were in the treatment combination of Lalpakri x T₅. Height of the plantlets (7.37 cm), number of shoot (3.84) and number of leaves (21.10) were maximum in BARI TPS-1 grown on T₃ (MS+0.03 mg⁻¹ NAA). But minimum height and less number of shoots were found in Cardinal grown on T₁ (MS+1 mg⁻¹ BAP). This was due to the effect of NAA and BAP, respectively on shoot formation in potato. Smolenskaya (1989) also reported similar results. Maximum number of nodes (17.37) was found in BARI TPS-1 x T₂ (MS+0.25 mg⁻¹ GA₃+2 mg⁻¹ calcium pantothenate) which might be due to the effect of GA₃ and calcium pantothenate. The results of present experiment suggest that BARI TPS-1 is a promising hybrid with suitable multiplication qualities through *in vitro* culture and inositol-supplemented medium can be used as a suitable medium for *in vitro* culture of potato.

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