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EFFECT OF LOW COST COMPONENT CULTURE MEDIA ON THE QUALITY AND YIELD OF *IN VITRO* PRODUCED MICROTUBERS OF POTATO

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ABSTRACT
The present investigation was carried out with an aim to evaluate the effect of culture media having low cost components on production of micro tubers of potato cultivar Kufri Himalini. For the commercialization and acceptance of *in vitro* propagation technique, the cost of production has to be competitive with conventionally propagated plant material. Therefore the reduction of cost of media becomes inevitable. Keeping this vision in mind, in the present study, the MS media drawn cultures were exposed to 10 mg/l BAP and the LC media drawn cultures were exposed to the varying concentration of BAP during tuberization. The number of microtuber harvested/ flask, yield number of eyes/microtuber, size of each microtuber, fresh and dry weight and percent biomass of each microtuber were observed and found significantly better in LC media than MS media.

KEY WORDS: Low cost, *in vitro*, potato and microtuber.

INTRODUCTION

For the commercialization and acceptance of *in vitro* propagation technique, the cost of production has to be competitive with conventionally propagated plant material. Therefore the reduction of cost of media becomes inevitable. In the recent past some research work has been initiated on this aspect and the same is being reviewed here. A low cost media was used for the propagation of potato made by using V_8 juice, table sugar, and NPK fertilizer, $CaCo_3$ and agar (V_8 juice media). The plant grown on V_8 media were slower to develop roots but after 3 week cultivation in Magenta media vessels at 22^0C, there was no difference in plant height and number of nodes per plant between plants repeatedly grown on MS media and V_8 media (Bains, 1991). Chandra *et al.*, (1991) found that the inorganic constitutions of MS media and sucrose could be replaced with commonly available fertilizers like CAN, SSP and MOP in the proportion of 6.5, 1.0 g/l respectively with 3% ordinary sugar and tap water (potable) for *in vitro* propagation of potato using single node in liquid media over filter paper bridges. As different concentration of ordinary sugar *i.e.* 8, 10, 12 and 14% obtained from open market did not have any statistical differences over 231 mM sucrose AR for the micro tubers formed in cultivar K. Jyoti, K. Badshah and Kufri Sinduri. Chandra (1992) recommended that lowest concentration of sugar *i.e.* 8% could replace 23 mM sucrose AR in the micro tuber induction media. Purnima (1997) replaced expensive ingredients like analytical grade chemicals and sucrose with commercial grade chemicals and table sugar, distilled water was substituted with tap water without any adverse impact on the efficiency for economy of *in vitro* propagation process. Nene and Sheila (1997) have used a low cost mediain which tapioca was used as a substitute for agar for propagation chick pea. Gebre and Sathyanarayana, (2001) used tapioca and sago as cheaper alternative to agar for direct shoot regeneration and microtuber production and concluded that tapioca at 11 – 15% gave comparable and/ or significantly higher results of *in vitro* shoot proliferation than the agar and sago. The best concentration (14%) was found to be higher than the report of Nene and Shelia (1997) for Tobacco and Chickpea culture. They also found that tapioca based media was better than agar based media for microtuber development. In view of the above background, the present investigation is undertaken to study the effect of low cost component culture media on the quality and yield of *In vitro* produced micro tubers of potato.

MATERIALS & METHODS

The present investigation was carried out with an aim to evaluate the effect of culture media having low cost components on production of micro tubers of potato cultivar Kufri Himalini. After successful shoot proliferation and multiplication the plantlets were shifted to pre-tuberization media consisted of MS (Murashige and Skoog, 1962) and LC (Low Cost) liquid propagation media (without agar and tapioca) with the hormonal combination which was found best in shoot proliferation and multiplication stage. Tuberization media consisted of MS liquid media (without agar) with 80 gm/l sucrose supplemented with 10 mg/l BAP and LC liquid media (without tapioca) with various concentration of BAP *i.e.* 5 ($LCBAP_5$), 8 ($LCBAP_8$), 10 ($LCBAP_{10}$), 12 ($LCBAP_{12}$) and 15 ($LCBAP_{15}$) mg/l.

After this the plantlets from pre-tuberization media were shifted to tuberization media and kept at 18 $\pm1^0$C temperature under complete darkness for the duration of 70 – 90 days, depending on the growth of microtubers (Naik and Karihaloo, 2007). The microtubers were pulled out from the flask and roots, stem and leaves were removed. These microtubers were thoroughly washed with double distilled water to remove the media and air-dried fro 24 hr. After drying, microtubers were packed in perforated polythene bags and stored at 4 $\pm1^0$C in the refrigerator. The observations for number of microtuber produced/ flask and yield/ flask were recorded from 10 randomly selected flask of each treatment. The fresh and dry weight of each microtuber and number of eyes and size of each microtuber were recorded from 10 randomly selected microtubers from different flask of various treatments.

RESULTS

The present investigation was carried out to develop a low cost component culture media for production of micro tuber of potato. Various combinations of hormones with culture media were studied and the results obtained during the study are described here.

Effect on number and yield of micro tubers

The data present in Table -1 indicates that the average number of microtuber in MS media reached 10.8 ± 1.3 microtubers /flask with the yield of 0.775 ±0.08 mg/ flask. LC media with 5 mg/l BAP ($LCBAP_5$) concentration showed 4.0 ±0.8 number of microtuber /flask with the yield of 0.202 ±0.04 mg/flask. In $LCBAP_8$ media microtuber was reported 12.8 microtuber/ flask with the yield of 0.872 ±0.01 mg/flask. $LCBAP_{10}$ media showed reduced number (8.1 ± 1.1 per flask) and yield (0.678 ±0.02 mg/flask) of microtruber. With the increasing concentration of BAP in LC media, number of microtuber and yield of microtuber/ flask was reduced and even no response was found in higher concentration of BAP *i.e.* 12 and 15 mg/l. The above data have shown that the number and yield of microtuber/ flask were reported higher in LC media with 8 mg/l BAP concentration than MS media. $LCBAP_5$ media showed least mean of number and yield of microtuber/flask than all other treatments. The concentration of 12 and 15 mg/l BAP with LC media showed no response till to the completion of the experiment.

TABLE-1: Effect of MS and LC media on no. of microtuber harvested/ flask and yield of microtuber/ flask (mg.)

Media Used	No. of microtuber harvested/ flask	Yield (mg.) of microtuber/ flask
MSBAP	10.8 ±1.3	0.775 ±0.08
$LCBAP_5$	4.0 ±0.8	0.202 ±0.04
$LCBAP_8$	12.8 ±1.3	0.872 ±0.01
$LCBAP_{10}$	8.1 ±1.1	0.678 ±0.02
$LCBAP_{12}$	No response	No response
$LCBAP_{15}$	No response	No response
F Value	108.44**	366.88**
LSD (P< 0.05)	0.86	0.03

***Significant*

Effect on number of eyes and size of micro tubers

To find out the quality of microtuber produced in different media, the number of eyes in each microtuber and diameter (size) of each microtuber was also observed (Table-2). In MS media the average number of eyes in each microtuber reached 9.3 ±1.7 with the size of 6.92 ±0.4 mm. In $LCBAP_5$ media 6.4 ±1.2 eyes were reported in each microtuber with 4.91 ±0.4 mm diameter. 8 mg/l BAP concentration of LC media showed 10.6 ±1.3 numbers of eyes and 7.32 ±0.1 mm sizes in each microtuber. In $LCBAP_{10}$ media number of eyes reported 9.0 ±1.0 with 6.51 ±0.4 mm sized microtuber. The average number of eyes (10.6 eyes in each microtuber) and maximum diameter of each microtuber (7.32 mm) was found better in LC media with 8 gm/l BAP concentration ($LCBAP_8$ media) than MS media with 10 mg/l BAP.

TABLE 2: Effect of MS and LC media on no. of eyes and size of each microtuber

Media Used	No. of eyes in each microtuber	Size of each microtuber (mm)
MSBAP	9.3 ±1.7	6.92 ±0.4
$LCBAP_5$	6.4 ±1.2	4.91 ±0.4
$LCBAP_8$	10.6 ±1.3	7.32 ±0.1
$LCBAP_{10}$	9.0 ±1.0	6.51 ±0.4
$LCBAP_{12}$	No response	No response
$LCBAP_{15}$	No response	No response
F Value	16.17**	69.43**
LSD (P< 0.05)	1.03	0.30

***Significant*

Effect on fresh and dry weight and percent biomass of micro tubers

To evaluate the strength of microtuber, fresh and dry weight and percent biomass was also observed. In MS media, fresh and dry weight of each microtuber, reported 0.075 ±0.009 and 0.025 ±0.001 mg respectively and biomass was reached 34.53% LC media with 5 mg/l BAP showed least mean of fresh (0.038 ±0.006) and dry (0.015 ±0.002) weight of each microtuber, but the biomass reached higher (39.33%) than all other treatments (Table-3). $LCBAP_8$ media showed maximum fresh (0.081 ±0.004) and dry (0.022 ±0.002) weight, but the biomass (27.66%) was least than all other treatments. In $LCBAP_{10}$ media fresh and dry weight was reported 0.075 ±0.004 and 0.023 ±0.002 mg respectively with the biomass of 30.78%. The above data indicates that maximum percent biomass was observed in $LCBAP_5$ media (39.33%) followed by MS (34.53%), $LCBAP_{10}$ (34.78%) and $LCBAP_8$ (27.66%).

TABLE 3: Effect of MS and LC media on fresh and dry weight of microtuber and percent biomass

Media Used	Fresh weight of each microtuber (mg)	Dry weight of each microtuber (mg)	% Biomass of each microtuber
MSBAP	0.075 ±0.009	0.025 ±0.001	34.53
$LCBAP_5$	0.038 ±0.006	0.015 ±0.002	39.33
$LCBAP_8$	0.081 ±0.004	0.022 ±0.002	27.66
$LCBAP_{10}$	0.075 ±0.004	0.023 ±0.002	30.78
$LCBAP_{12}$	No response	No response	No response
$LCBAP_{15}$	No response	No response	No response
F Value	92.73**	46.03**	15.94**
LSD (P< 0.05)	1.53	1.62	2.99

***Significant*

DISCUSSION

To evaluate the comprehensible difference and effect of low cost culture media the culture proliferating in MS and LC media were maintained separately for tuber induction. In the present study, the MS media drawn cultures were exposed to 10 mg/l BAP and the LC media drawn cultures were exposed to the varying concentration of BAP during tuberization. The number of microtuber harvested/ flask, yield number of eyes/microtuber, size of each microtuber, fresh and dry weight and percent biomass of each microtuber from LC media were significantly better than that recorded from MS media (Table -1, 2 and 3). With the increasing concentration of BAP in LC media, number of microtuber and yield of microtuber/ flask was reduced and even no response was found in higher concentration of BAP *i.e*. 12 and 15 mg/l. The data showed that the number and yield of microtuber/ flask was reported higher in LC media with 8 mg/l BAP concentration than MS media. $LCBAP_5$ media showed least mean of number and yield of microtuber/ flask than all other treatments and the concentration of 12 and 15 mg/l BAP with LC media showed no response till to the completion of the experiment. To find out the quality of microtuber produced in different media, the number of eyes in each microtuber and diameter (size) of each microtuber was observed (Table-2). The average number of eyes (10.6 eyes in each microtuber) and maximum diameter of each microtuber (7.32 mm) was found better in LC media with 8 gm/l BAP concentration ($LCBAP_8$ media) than MS media with 10 mg/l BAP. To evaluate the strength of microtuber, freshj and dry weight and percent biomass was also observed. The data indicates that maximum percent biomass was observed in $LCBAP_5$ media followed by MS (34.53%), $LCBAP_{10}$ (30.78%) and $LCBAP_8$ (27.66%).

The cultures were exposed to a major change from vegetative growth to reproductive phase leading to tuber development, when shifting from pretuberization to tuberization stage. Kn and GA was an integral component of pretuberization media but during tuber induction stage, Kn and GA was withdrawn as it channelizes all the carbohydrates towards shoot development during pretuberization as procedure suggested by Krauss, 1978. Decrease in Kn and GA promotes have been reported to be partitioning of biomass to the tubers by Krauss, 1978 and 1982. Hence tuber induction could be achieved by withdrawal of above maintained hormones and addition of the inhibitor. Various concentrations of BAP with LC media were studied to see their effect on tuberization. BAP as an inhibitor has been used in varying concentration from 5 mg/l to 15 mg/l and due to its inhibitory role and the presence of BAP channelizes all the resources of plants towards tuberization, *i.e.* anabolic activity rather than elongation of stem, *i.e.* a catabolic activity. With this idea of inhibitory metabolites, triazoles have come up as suitable tuber inducing substances which has been used in several studies by Harvey, 1990 and Simko, 1994. Several other inhibitors have also been used like CCC, triademefon and paclobutrazol for tuberization by researchers (Harvey, 1990; Simko, 1991, 1994). The result of the present study showed that tapioca and agar are the best alternative of agar and sucrose respectively, to reduce the cost of media. For tuberization LC nutrients with 8 mg/l BAP concentration was found suitable in comparison to MS nutrients with 10 mg/l BAP. By using LC nutrients in the place of MS nutrients, in tuberization stage, the number of microtuber produced, yield of microtubers and quality of microtubers was found superior.

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