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Cassava Powder and Jaggery as Alternative Low-cost Medium Components for *in vitro* Potato Plantlet Production

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Abstract

In the present experiment, the effectiveness of inexpensive alternative gelling agents (Cassava powder and Isabgol) and sucrose sources (Table sugar, Jaggery, Rock sugar and Sugarcane juice) used for *in vitro* plantlet production of two potato cultivars, Kufri Himalini and Kufri Jyoti was evaluated. These alternatives were compared against standard control as agar 6 g/L and Sucrose 30 g/L, which are known to be expensive. Effective modified media for potato plantlet production was treatment (MS salts + Cassava powder 80 g/L+ Jaggery 20 g/L + Calcium pantothenate 1 mg/L) replacing agar with cassava powder and sucrose with jaggery, resulted in a significantly reduced media cost up to 82.60 per cent compared to control (MS salts + Agar-Agar 6 g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L) for the preparation of one litre MS media.

Keywords : Potato, Cassava powder, Jaggery, Kufri himalini, Kufri jyoti, Tissue culture, Low-cost

Potato (*Solanum tuberosum* L.), originated from Peru is a crucial global food crop, it occupies the largest area among vegetable crops. India produced over 60.14 million tonnes of potatoes from 2.3 million hectares with an average yield of 25.79 tons per hectare (Anonymous, 2022). Potato occupies premier place in the list of vegetable crops in the world including India. It is a member of Solanaceae family (Chromosome number 2n = 48). Potatoes are an important source of carbohydrate, protein, vitamins and minerals. It is used as a staple food in many countries of the world (Srikanth *et al.,* 2023).

The potato is a unique type of crop that can be multiplied sexually using true potato seeds (TPS) and vegetatively using tubers. However, vegetative propagules or tubers are the main method of potato cultivation in the majority of the world's regions, including India. In this case, the seed tubers are one of the main input expenses for potato farming. A hectare of planting requires in estimations, two to three tonnes of potato tubers, which makes up between forty to fifty per cent of the cultivation cost (Simmonds, 1997; Struik & Wiersema, 1999 and Pandey & Sarkar, 2005). One of the biggest obstacles to potato cultivation, besides the high seed rate and high cost of seed potatoes is the lack of virus-free tubers. Potato cultivars can only produce their full potential yield when the planting material is virus-free.

The quick and healthy planting materials produced by tissue culture (TC) technology provide a solution

and serve as vital for increasing crop yields. By tackling problems with food security and agricultural production, TC techniques have opened a new chapter in agricultural science (Ogero *et al.,* 2012). Micropropagation techniques offer a solution to many of the issues associated with traditional seed production systems (Singh *et al.,* 2019). Micropropagation now supports many seed potato production systems by providing nuclear stock material in the form of micro plants or microtubers (*in vitro* produced tubers) for subsequent use in potato seed production channels such as aeroponics, apical rooted technology, mini tuber production in soil and mini tuber production from micro plants (Chindi *et al.,* 2014 and Bharath & Raju, 2023).

However, TC has high operating costs, especially in developing nations, making the technology inaccessible to smallholders (Saraswathi *et al.,* 2016). Production cost limit the scope of economic application of micropropagation. The expense of nutrient medium can account for 30 to 35 per cent of micropropagated plant production (Gitonga *et al.,* 2010). In order to make TC more accessible and advantageous for farmers, alternative low-cost resources are required due to the high cost of production, particularly in medium preparation.

To reduce the unit cost of plant propagules low cost micropropagation was practiced (Savangikar *et al.,* 2002). Reducing the cost of media had a huge effect on horticulture and agriculture sector especially in developing countries. Agar is the most commonly used gelling agent and most expensive media component contributing about 65 to 70 per cent of the cost of culture medium (Prakash, 1993; Tripathi *et al.,* 2021 and Ebile *et al.,* 2022). The selection of a gelling agent is the subject of major concern for cost reduction due to its high impact on cost (Petrovski and Tilette, 2012).

Sucrose is the preferred carbohydrate for most studies due to its ability to be easily translocated and its resistance to enzymatic degradation, owing to its non-reducing nature (Pontis, 1978). While sucrose is commonly used in the vast majority of *in vitro* shoot induction and development studies in woody species, it is not always the most effective carbon source (Thomson and Thorpe, 1987). Additionally, the high cost of sucrose, which accounts for 21.7 to 30 per cent of the media cost (Prakash *et al.,* 2004; Demo *et al.,* 2008; Swamy *et al.,* 2010 and Dantas *et al.,* 2021).

Considering the importance of finding alternative gelling agent and sucrose source for the mass multiplication of potato plantlets, the experiment was designed to find an inexpensive gelling agent and sucrose source for *in vitro* potato plantlet production.

Material and Methods

This experiment was conducted at the Plant Tissue Culture Laboratory, Department of Horticulture, University of Agricultural Sciences, Bangalore with the objective to examine the effectiveness of alternative gelling agent and sucrose source for *in vitro* potato plantlet production. The three main gelling agents used for experiment were agar-agar, cassava powder and isabgol, agar-agar being a standard gelling agent. The five carbon sources, used for experiment were sucrose table sugar, jaggery, sugarcane juice, rock sugar, sucrose being used as stranded carbon source for this experiment.

Cassava powder is prepared from cassava tubers (*Manihot esculenta*) as small tapioca pearls. Originally granular kind of product obtained from sago palms (*Metroxylon sagu*). Tapioca pearls were procured from D'Mart, Yelahanka, Bengaluru. These tapioca pearls were ground and sieved and added to media at various concentrations (60 g/L, 80 g/L and 100 g/L) it was added when the medium was warm by thorough mixing to ensure proper gelling of the medium.

Isabgol powder is derived from the seeds of *Plantago ovata*, an herb belonging to the family Plantaginaceae. Dried seeds of the plant contain over 30 per cent mucilage. The husk, which contains all the mucilaginous matter, is separated from seeds by crushing and winnowing. Isubgol is a polysaccharide in nature (Jain *et al.,* 1997) and composed of xylose, arabinose, galactouronic acid, rhamnose and galactose (Babbar and Jain, 1998). Unlike other gelling

materials, isabgol was added to the MS medium at the rate of 20 g/L, 30 g/L and 40 g/L of the medium when it was just lukewarm. Overheating rendered the medium rubbery consequently leading to difficulty in dispensing the medium. Isabgol was brought from Government Ayurvedic Medical College, Bengaluru, India.

Table sugar is a crude sucrose derived by crushing and extraction of sugarcane (*Saccharum officinarum*). Sugar is hard, white, dry crystals, lumps or powder, sweet taste, odourless soluble in water and very slightly soluble in alcohol (Arthur and Rose, 1996). Raw cane sugar contains 96 to 97 per cent sucrose, 0.75 to 1.0 per cent reducing sugar, 0.75 per cent moisture, 0.5 per cent ash and remainder, organic non-sugars (Anonymous, 1972). In the present study, the medium was supplemented with common grade sugar (D'Mart, Yelahanka, Bengaluru, India) at 20, 30 and 40 g/L of the medium for plantlet production.

Jaggery is the product obtained on concentrating sugarcane juice with or without prior purification into a solid or semi solid state. It is also called gur, contains all the constituents of cane juice, some of them having undergone slight changes during boiling. Per cent composition of gur is: sucrose, 65 to 85; invert sugar, 10-15; ash, 2.5 and moisture, 3 to 6. It also contains carotene, 280 I.U./100 g.; nicotinic acid, 1.0 mg./100 g.; vitamin B1, 20 ug/100 g. and traces of iron and copper (Anonymous, 1957). In the present study, the medium was supplied with jaggery (D'Mart, Yelahanka, Bengaluru, India) at 20, 30 and 40 g/L of the medium for plantlet production.

Rock sugar, a confectionery mineral with large sugar crystals, forms from a supersaturated sugar-water solution crystallizing onto a surface. Heating the solution enhances sugar dissolution, yielding larger crystals after several days. Originating in India and Iran, rock candy is used to sweeten tea and as a mouth freshener, often with aniseed. It also relieves cough. In this study, the medium was supplemented with rock sugar at concentrations of 20, 30 and 40 g/L for potato plantlet production (D'Mart, Yelahanka, Bengaluru, India).

Sugarcane juice is an opaque liquid, grey to dark green in color, containing in solution all the soluble constituents of cane. Sugarcane juice (specific gravity; 1.07; pH, 5.8 to 6.4) contains; water, 80 to 85 per cent; sucrose, 13 to 18 per cent; glucose, 0.3 to 1.0 per cent and non-reducing sugar, 1.0 per cent. The analysis of the non-sugars is as follows: nitrogen, 0.019; protein, nil; ash, 0.37; calcium, 0.021 and phosphorus, 0.032 g/100 cc. juice (Anonymous, 1957). In the present study, medium was added with sugarcane juice (Cane-O-La, Jayanagar, Bengaluru, India) at 100, 150 and 200 ml/L of the medium.

Explants used in this experiment were collected from CIP (International Potato Centre), Bengaluru. Single nodal cuttings were used as explant for this experiment. Cultivars selected were Kufri Himalini and Kufri Jyoti. Observations were recorded three weeks after the culturing on growth parameters like plantlet height, number of leaves, number of nodes, root length and number of roots per plantlet for statistical analysis. During the growth stage temperature was maintained at 22±2^{0}C with16 hours of light period and 8 hours of dark period.

Table 1

Alternative gelling agent used *in vitro* potato plantlet production

Treatments	Treatment details
Control	MS salts + Agar 6 g/L+ Sucrose 30 g/L+ Calcium pantothenate 1 mg/L
A_1	MS salts + Isabgol 20 g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L
A_2	MS salts + Isabgol 30 g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L
A_3	MS salts + Isabgol 40 g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L
A_4	MS salts + Cassava powder 60g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L
A_5	MS salts + Cassava powder 80 g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L
A_6	MS salts + Cassava powder 100 g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L

TABLE 2

Alternative sucrose sources along with cassava powder as alternative gelling agent for *in vitro* potato plantlet production

Treatments	Treatment details
Control	MS salts + Agar 6 g/L+ Sucrose 30 g/L+ Calcium pantothenate 1 mg/L
B_1	MS salts + Agar 6 g/L+ Table sugar 20 g/L+ Calcium pantothenate 1 mg/L
B_2	MS salts + Agar 6 g/L+ Table sugar 25 g/L+ Calcium pantothenate 1 mg/L
B_3	MS salts + Agar 6 g/L+ Table sugar 30g/L+ Calcium pantothenate 1 mg/L
B_4	MS salts + Agar 6 g/L+ Jaggery 20 g/L+ Calcium pantothenate 1 mg/L
B_5	MS salts + Agar 6 g/L+ Jaggery 30 g/L+ Calcium pantothenate 1 mg/L
B_6	MS salts + Agar 6 g/L+ Jaggery 40 g/L+ Calcium pantothenate 1 mg/L
B_7	MS salts + Agar 6 g/L+ Rock Sugar 20 g/L+ Calcium pantothenate 1 mg/L
B_8	MS salts + Agar 6 g/L+ Rock Sugar 30 g/L+ Calcium pantothenate 1 mg/L
B_9	MS salts + Agar 6 g/L+ Rock Sugar 40 g/L+ Calcium pantothenate 1 mg/L
B_{10}	MS salts + Agar 6 g/L+ Sugarcane juice 100 ml/L+ Calcium pantothenate 1 mg/L
B_{11}	MS salts + Agar 6 g/L+ Sugarcane juice 150 ml/L+ Calcium pantothenate 1 mg/L
B_{12}	MS salts + Agar 6 g/L+ Sugarcane juice 200 ml/L + Calcium pantothenate 1 mg/L
AB_1	MS salts + Cassava powder 80 g/L+ Table sugar 20 g/L+ Calcium pantothenate 1 mg/L
AB_2	MS salts + Cassava powder 80 g/L + Table sugar 25 g/L+ Calcium pantothenate 1 mg/L
AB_3	MS salts + Cassava powder 80 g/L + Table sugar 30g/L+ Calcium pantothenate 1 mg/L
AB_4	MS salts + Cassava powder 80 g/L + Jaggery 20 g/L+ Calcium pantothenate 1 mg/L
AB_5	MS salts + Cassava powder 80 g/L + Jaggery 30 g/L+ Calcium pantothenate 1 mg/L
AB_6	MS salts + Cassava powder 80 g/L + Jaggery 40 g/L+ Calcium pantothenate 1 mg/L
AB_7	MS salts + Cassava powder 80 g/L + Rock Sugar 20 g/L+ Calcium pantothenate 1 mg/L
AB_8	MS salts + Cassava powder 80 g/L + Rock Sugar 30 g/L+ Calcium pantothenate 1 mg/L
AB_9	MS salts + Cassava powder 80 g/L + Rock Sugar 40 g/L+ Calcium pantothenate 1 mg/L
AB_{10}	MS salts + Cassava powder 80 g/L + Sugarcane juice 100 ml/L+ Calcium pantothenate 1 mg/L
AB_{11}	MS salts + Cassava powder 80 g/L + Sugarcane juice 150 ml/L+ Calcium pantothenate 1 mg/L
AB_{12}	MS salts + Cassava powder 80 g/L + Sugarcane juice 200 ml/L + Calcium pantothenate 1 mg/L

The complete data was analysed using CRD. The critical difference of the experimental data was tested by using F test at 1 per cent level of confidence. The analysis was done using OPSTAT online analysis tool.

RESULTS AND DISCUSSION

Influence of Alternative Gelling Agents on Potato Plantlet Growth: Among different treatments, treatment A_5 (MS salts + Cassava powder 80 g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L) produced significantly longer plantlets, more number of leaves, maximum root length, more number of roots and nodes per plantlet for Kufri Himalini (10.31 cm, 9.20, 7.92 cm, 6.70 and 9.30, respectively) which was statistically on par with the control (MS salts + Agar 6 g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L) for plantlet height, number of leaves, root length, number of roots and number of nodes for Kufri Himalini (10.17 cm, 8.30, 7.78 cm, 6.50 and 9.30, respectively). Conversely, shorter plantlets, minimum leaves, shorter roots, least number of roots and nodes were recorded in treatment A_3 (MS salts + Isabgol 40 g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L) for Kufri Himalini (7.02 cm, 5.90, 5.01 cm, 4.20

TABLE 3

Influence of alternative gelling agents on growth parameters of *in vitro* potato plantlet production cv. Kufri Himalini

Treatments	Plantlet height (cm)	Number of leaves	Root length (cm)	Number of roots	Number of nodes
Control	10.17	8.90	7.78	6.50	9.30
A_1	9.96	8.70	7.71	6.20	9.10
A_2	8.97	7.40	6.84	5.40	7.70
A_3	7.02	5.90	5.01	4.20	6.50
A_4	8.95	7.60	6.81	4.90	7.90
A_5	10.31	9.20	7.92	6.70	9.50
A_6	8.76	7.40	6.57	5.40	7.80
F-test 1%	**	**	**	**	**
S. Em ±	0.07	0.19	0.07	0.18	0.17
CD at 1%	0.27	0.74	0.26	0.69	0.64

** -Significant at 1% level; S. Em- Standard error mean; CD-Critical difference

and 6.50, respectively) as presented in Table 3 and depicted in Plate 1.

Among different treatments, treatment A_5 (MS salts + Cassava powder 80 g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L) produced significantly longer plantlets, more number of leaves, maximum root length, more number of roots and nodes per plantlet for Kufri Jyoti (8.69 cm, 7.20, 6.16 cm, 6.60 and 7.50, respectively) which was statistically on par with the control (MS salts + Agar 6 g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L) for all the growth parameters for Kufri Jyoti (8.53 cm, 7.30, 5.98 cm, 6.40 and 7.50, respectively). Conversely, shorter plantlets, minimum leaves, shorter roots, least number of roots and nodes were recorded in treatment A_3 (MS salts + Isabgol 40 g/L + Sucrose 30 g/L + Calcium pantothenate 1 mg/L) for Kufri Jyoti (5.19 cm, 4.30, 4.31 cm, 3.90 and 4.70, respectively) as presented in Table 4 and depicted in Plate 2 and 3.

Plate 1: Influence of alternative gelling agents on growth of *in vitro* potato plantlet production cv. Kufri Himalini

TABLE 4

Influence of alternative gelling agents on growth parameters of *in vitro* potato plantlet production in cv. Kufri Jyoti

Treatments	Plantlet height (cm)	Number of leaves	Root length (cm)	Number of roots	Number of nodes
Control	8.53	7.30	5.98	6.40	7.50
A_1	8.45	7.20	5.92	6.10	7.40
A_2	7.39	5.90	5.07	5.40	6.20
A_3	5.19	4.30	4.31	3.90	4.70
A_4	7.14	5.60	5.17	5.30	5.70
A_5	8.69	7.50	6.16	6.60	7.80
A_6	6.96	5.80	4.83	4.90	6.10
F-test 1%	*	*	*	*	*
S. Em ±	0.08	0.21	0.07	0.15	0.23
CD at 1%	0.33	0.79	0.26	0.59	0.88

** -Significant at 1% level; S. Em- Standard error mean; CD-Critical difference

Plate 2: Influence of potato plantlet growth on media gelled with different concentration of isabgol and stranded gelling agent cv. Kufri Jyoti; a) 20 g/L b) 30 g/L c) 40 g/L Cassava powder d) Agar-Agar 6 g/L

Explants cultured on cassava powder produced healthy plantlets reason might be due to the richness of nutrients like carbohydrates, amino acids and minerals present in cassava powder. Similar results were reported by Prabhakara (1999) in anthurium, Srivastava (1998) in jasmine, Ssamula (2012) in banana and Umesh & Urguru (2013) in ginger. Another advantage of using cassava powder compared to agar was that cassava powder gelled media was easily separable from roots. A similar observation was also reported in chrysanthemum by Bhattacharya *et al.* (1994) and Sharma *et al.* (2015) in *Aloe vera*.

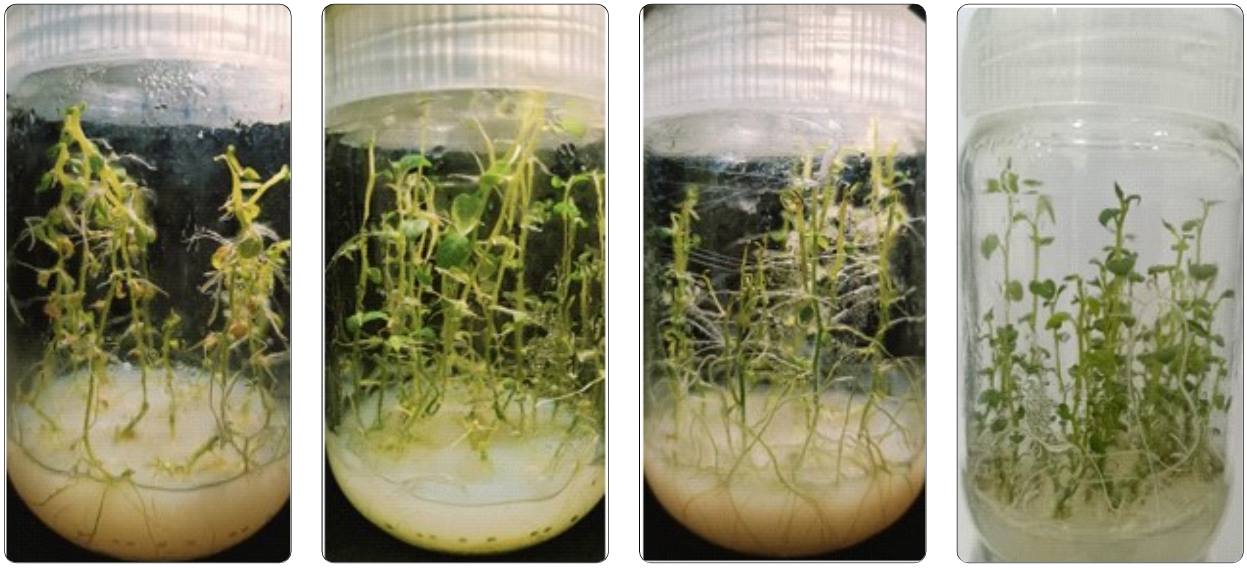

Plate 3: Influence of potato plantlet growth on media gelled with different concentration of casava powder and stranded gelling agent cv. Kufri Jyoti; a) 60 g/L b) 80 g/L c) 100 g/L Cassava powder and d) Agar-Agar 6 g/L

However, cassava powder is less transparent and contamination is difficult to detect. The results are in conformity with the findings of Buah (2014), Kuria *et al.* (2008) and Saraswathi *et al.* (2016).

An increase in the concentration of Isabgol was negatively correlated with growth of the plantlets. This might be due to the hardness of the media; higher solidification leads difficult to root growth and absorption of nutrients from the media. Similar findings were reported by Naik and Sarka (2001) in potato. Moreover, media solidified with a higher concentration of Isabgol was difficult to dissolve, pour and clear. Similar observations were also reported by Kodym and Zapata-Arias (2001) and Prakash *et al.* (2004).

Compared to isabgol, cassava powder performed slightly better concerning all the growth parameters reason may be due to unlike in isubgol, cassava powder contains calcium, fibre, sugars and mineral nutrients present in cassava powder might supplemented the growth and development of potato plantlets. Similar results were reported by Prakash (1993) *in vitro* culture of ginger and turmeric, Prabhakara and Reddy (2004) in anthurium, Shalija and Patil (2004) in gerbera Umeh and Uguru (2013) in ginger and Eble *et al.* (2022) for *in vitro* culture of Banana.

Prabhakara (1999) opined that, on heating, cassava powder converted complex polysaccharide dextrin witch along with nutrient media supports the growth of the cell. The better response on cassava powder gelled medium could be also due absence of inhibitors which have been present in agar as reported by Debergh (1983), Singha (1984), Pierik (1997) and Puchooa & Purseramen (1999).

Tissue culture technique is highly suited for rapid multiplication of genotypes. An often-cited disadvantage of modern plant tissue culture methods is the relatively higher costs involved as compared to other methods (Sahu and Sahu, 2013). The need for low-cost plant tissue culture systems, applicable for micropropagation has been emphasized to allow the large-scale application of such technology in developing countries.

These findings suggest that both cassava powder and isabgol are effective substitutes agar for *in vitro* culture media. However, given its lower cost, cassava powder

can be a more suitable and cost-effective alternative for potato micropropagation, making large-scale production more economically viable.

Influence of Alternative Sucrose Sources, Combination of Alternative Sucrose and Cassava Powder on Potato Plantlet Growth : Among alternative sucrose sources and cassava powder as a gelling agent potato plantlet production significantly maximum plantlet height, more number of leaves, maximum root length, more number of roots and number of nodes per plantlet was recorded in treatment B_4 (MS salts + Agar 6 g/L+ Jaggery 20 g/L+ Calcium pantothenate 1 mg/L) for both the cultivars Kufri Himalini (10.20 cm, 9.30, 8.16 cm, 7.10 and 9.80, respectively) and Kufri Jyoti (8.68 cm, 8.10, 6.34 cm, 6.50 and 8.40, respectively). This was statistically at par with treatment control for all growth parameters (MS salts + Agar 6 g/L+ sucrose 30 g/L + Calcium pantothenate 1 mg/L) for both cultivars Kufri Himalini (10.18 cm, 8.80, 7.91 cm, 6.80 and 9.60, respectively), treatment B_3 (MS salts + Agar 6 g/L+ Table sugar 30 g/L + Calcium pantothenate 1 mg/L) averaging (10.12 cm, 8.60, 7.85 cm, 6.70 and 7.50, respectively) and treatment AB_4 (MS salts + Cassava powder 80 g/L + Jaggery 20 g/L + Calcium pantothenate 1 mg/L) with averaging (10.01 cm, 8.40, 7.78 cm, 6.30 and 8.90, respectively) for Kufri Himalini. In contrast to this shortest plantlets, minimum leaves, shorter roots, least number of roots and nodes were recorded in treatment AB_{10} (MS salts + Cassava powder 80 g/L + Sugarcane juice 100 ml/L+ Calcium pantothenate 1 mg/L) for Kufri Himalini (5.73 cm, 4.90, 4.57 cm, 3.20 and 5.90, respectively) as presented in Table 5.

Among different treatments, treatment significantly maximum plantlet height, more number of leaves, maximum root length, more number of roots and number of nodes per plantlet was recorded in treatment B_4 (MS salts + Agar 6 g/L+ Jaggery 20 g/L+ Calcium pantothenate 1 mg/L) for Kufri Jyoti (8.68 cm, 8.10, 6.34 cm, 6.50 and 8.40, respectively). This was statistically at par with treatment control for all growth parameters (MS salts + Agar 6 g/L+ sucrose 30 g/L + Calcium pantothenate 1 mg/L) for Kufri Jyoti (8.47 cm, 7.80, 6.26 cm, 6.40 and 8.10, respectively), treatment B_3 (MS salts + Agar 6 g/L+ Table sugar 30 g/L + Calcium pantothenate 1 mg/L) averaging (8.41 cm, 7.60, 6.11 cm, 6.10 and 7.90, respectively) and treatment AB_4 (MS salts + Cassava powder 80 g/L + Jaggery 20 g/L + Calcium pantothenate 1 mg/L) with averaging (8.36 cm, 7.50, 5.97 cm, 5.90 and 7.80, respectively) for Kufri Jyoti. Conversely, the shortest plantlets, minimum leaves, shorter roots, least number of roots and nodes were recorded in treatment AB_{10} (MS salts + Cassava powder 80 g/L + Sugarcane juice 100 ml/L+ Calcium pantothenate 1 mg/L) for Kufri Jyoti (4.49 cm, 4.70, 4.42 cm, 3.10 and 4.90, respectively) as presented in Table 6.

Jaggery as a sucrose source increased plantlet height compared to HiMedia sucrose this might be because of its complex composition, which includes important minerals, vitamins and other organic substances in addition to sucrose. Plantlet growth is supported by a richer and more balanced environment, the presence of micronutrients potassium, calcium and iron, which can strengthen a plant's ability to absorb nutrients, increase its metabolic activity and speed up its growth. Results were conflicting with the findings of Prakash *et al.* (2004), Joshi *et al*. (2009) and Rajavel & Stephan (2014) in ginger, turmeric and *Wrightia tomentosa,* respectively.

A notable observation regarding the use of jaggery as a carbon source is that jaggery at higher concentrations reduced all growth parameters. This effect may be due to osmotic interference caused by elevated sugar levels in the medium, which aligns with the findings of Perata *et al.* (1997), who reported that high sugar concentrations can disrupt the signal transduction pathway of GA_3. Increased sugar concentration in the medium also leads to hyperhydricity, resulting in reduced cellulose and chlorophyll content, lower ethylene production and abnormal nitrogen and sugar metabolism.

Table sugar as a sucrose source performed comparable results to HiMedia sucrose reason might be that to efficient translocation and

Plate 4: Poor response was noticed in media prepared by using alternative sucrose sources cv. Kufri Himalini (A) and Kufri Jyoti (B), a) Sugarcane juice b) Rock sugar

TABLE 5

Influence of alternative sucrose source, combination of alternative sucrose and cassava powder on growth parameters of *in vitro* potato plantlet production cv. Kufri Himalini

Treatments	Plantlet height (cm)	Number of leaves	Root length (cm)	Number of roots	Number of nodes
Control	10.18	8.80	7.91	6.80	9.60
B_1	7.67	6.50	5.71	4.30	7.40
B_2	8.52	7.80	6.64	5.40	8.20
B_3	10.12	8.60	7.85	6.70	9.50
B_4	10.32	9.30	8.16	7.10	9.80
B_5	8.75	7.60	6.67	5.90	8.60
B_6	6.76	5.80	5.84	4.10	6.90
B_7	7.84	6.40	6.07	4.40	6.70
B_8	8.05	6.60	6.28	4.70	6.90
B_9	8.27	7.10	6.46	4.90	7.30
B_{10}	6.06	5.40	4.64	3.60	6.30
B_{11}	6.39	5.70	4.96	3.70	6.60
B_{12}	6.55	6.10	5.22	4.10	6.80
AB_1	7.49	6.30	5.54	4.10	7.10
AB_2	8.31	7.10	6.27	5.20	7.80
AB_3	9.62	7.90	7.62	5.90	8.40
AB_4	10.01	8.40	7.78	6.30	8.90

Continued....

Table 5 Continued....

Treatments	Plantlet height (cm)	Number of leaves	Root length (cm)	Number of roots	Number of nodes
AB_5	8.31	6.90	6.43	5.60	8.10
AB_6	6.54	5.20	5.61	3.90	6.50
AB_7	7.53	5.90	5.82	4.10	6.30
AB_8	7.81	6.30	6.05	4.30	6.60
AB_9	7.97	6.60	6.22	4.70	6.80
AB_{10}	5.73	4.90	4.57	3.20	5.90
AB_{11}	5.98	5.30	4.69	3.50	6.20
AB_{12}	6.22	5.60	4.85	3.90	6.50
F-test 1%	**	**	**	**	**
S. Em ±	0.09	0.20	0.10	0.20	0.21
CD @ 1%	0.34	0.75	0.38	0.75	0.76

** -Significant at 1% level; S. Em- Standard error mean; CD-Critical difference

Table 6

Influence of alternative sucrose source, combination of alternative sucrose and cassava powder on growth parameters of *in vitro* potato plantlet production cv. Kufri Jyoti

Treatments	Plantlet height (cm)	Number of leaves	Root length (cm)	Number of roots	Number of nodes
Control	8.47	7.80	6.26	6.40	8.10
B_1	6.62	6.30	4.96	4.50	6.60
B_2	7.81	6.80	5.27	5.30	7.20
B_3	8.41	7.60	6.11	6.10	7.90
B_4	8.68	8.10	6.34	6.50	8.40
B_5	7.47	7.20	5.48	5.40	7.50
B_6	5.83	6.40	4.87	4.90	6.70
B_7	6.02	5.60	4.97	4.80	5.90
B_8	6.49	5.90	5.18	5.10	6.20
B_9	6.68	6.30	5.39	5.30	6.60
B_{10}	4.61	5.10	4.78	4.30	5.40
B_{11}	4.78	5.30	4.93	4.60	5.70
B_{12}	4.85	5.70	5.24	4.90	5.90
AB_1	6.36	5.90	4.72	4.20	6.30
AB_2	7.48	6.50	5.09	4.90	6.70
AB_3	8.01	7.10	5.72	5.50	7.60
AB_4	8.36	7.50	5.97	5.90	7.90
AB_5	7.19	6.90	5.34	4.90	7.20
AB_6	5.64	6.10	4.61	4.50	6.30
AB_7	5.83	5.40	4.74	4.40	5.70
AB_8	6.32	5.70	5.02	4.70	5.90

Continued....

Table 6 Continued....

Treatments	Plantlet height (cm)	Number of leaves	Root length (cm)	Number of roots	Number of nodes
AB_9	6.45	5.90	5.25	4.90	6.30
AB_{10}	4.49	4.70	4.42	3.10	4.90
AB_{11}	4.61	4.80	4.74	3.40	5.20
AB_{12}	4.74	5.20	4.72	3.70	5.50
F-test 1%	**	**	**	**	**
S. Em ±	0.12	0.16	0.07	0.15	0.14
CD at 1%	0.45	0.57	0.28	0.57	0.53

** -Significant at 1% level; S. Em- Standard error mean; CD-Critical difference

assimilation of table sugar by explants leads to eventual growth which is similar to sucrose. The results of the present study was in accordance with previous studies reported by (Ganapati *et al.,* 1995; Saeed, 2006 and Das & Gupta, 2009).

In the present study, poor response was observed when sugarcane juice and rock sugar were used as alternative sources for sucrose for both the cultivars Kufri Himalini [Plate 4 (A)] and Kufri Jyoti [Plate 4 (B)] examined during *in vitro* study. Sugarcane juice was found to be unsatisfactory because to medium turned to a dark brown color that adversely affected the culture growth and concerned rock sugar inhibitors already present or formed during the autoclaving as reported by Maheswari *et al.* (1980) and Hsiao and Bormann (1989). These findings are also in conformity with the reports of Prakash (1993), Prakash *et al.* (2004) and Joshi *et al.* (2009) in ginger, turmeric and *Wrightia tomentosa,* respectively.

Among the two different cultivars used for *in vitro* studies, Kufri Himalini gave the better response compared to Kufri Jyoti. This could be attributed to the fact that the genetic characteristics and interaction with the prevailing *in vitro* conditions which play a vital role in the performance of a variety. Such variations in growth among the varieties were reported by Hafsan *et al.* (2018), Srikant & Mohan (2022) and Bharath & Raju (2023).

Cost Analysis

Modified media (MS salts + Casava powder 80 g/L + Jaggery 20 g/L+ B-Complex 15 mg/L) resulted in a reduction in media cost up to 82.60 per cent compared to control (MS salts + Agar 6 g/L+ Sucrose 30 g/L+ Calcium pantothenate 1 mg/L) for potato plantlet production, as presented in Table 7. The results were in accordance with the findings of Gayatri *et al.* (2004) who found a reduction in media cost by adopting low-cost alternatives for *in vitro* culture of ginger. Similar,

Table 7

Cost analysis of MS-media and modified media for *in vitro* potato plantlet production

Constituents	Stock-A (Rs.)	Stock-B (Rs.)	Stock-C (Rs.)	Stock-D (Rs.)	Agar (Rs.)	Sucrose (Rs.)	Calcium pantothenate (Rs.)	Total cost (Rs.)
Control	7.30	0.34	0.29	2.61	68.40	34.80	0.038	113.77
Modified media	7.30	0.34	0.29	2.61	5.61	3.60	0.038	19.78
Per cent reduction	-	-	-	-	91.79	89.65	-	82.6

Control: MS salts + Agar 6 g/L+ Sucrose 30 g/L+ Calcium pantothenate 1 mg/L

Modified media: MS-Salts + Cassava powder 80 g/L + Jaggery 20 g/L+ B-Complex 15 mg/L

TABLE 8

Deferential cost of one litre media for standard and alternative sources

Components	Quantity used / liter (g/L)	Price/Kg/L/ (Rs.)	Price/ litre of medium (Rs.)	Manufacture/ Procured from
Agar	6	11400	68.4	HiMedia
Isabgol	20	1300	26	Government Ayurvedic Medical College, Bengaluru
	40		52	
	60		78	
Cassava power	60	70	4.2	D Mart Yelahanka, Bengaluru, India
	80		5.6	
	100		7.0	
Sucrose	30	1160	34.8	HiMedia
Table sugar	20	48	0.96	D Mart Yelahanka, Bengaluru, India
	30		1.44	
	40		1.92	
Jaggery	20	120	2.4	D Mart Yelahanka, Bengaluru, India
	30		3.6	
	40		4.8	
Rock sugar	20	220	4.4	D Mart Yelahanka, Bengaluru, India
	30		6.6	
	40		8.8	
Sugarcane juice	100	80 (1L)	8	Cane-O-La, Jayanagar, Bengaluru, India
	150		12	
	200		16	

results were obtained by Goel *et al.* (2007), Demo *et al.* (2008) and Dhanalakshmi & Stephan (2014) in *Rauwolfia serpentina*, potato and banana, respectively.

The results of the present study emphasize the potential of using cost-effective alternative media components for the *in vitro* mass multiplication of potato plantlets. The replacement of traditional, expensive agar and sucrose with more economical options such as cassava powder and jaggery, resulted in a significant reduction in production costs without compromising the effectiveness of plantlet production. These findings were in accordance with Ullah *et al.* (2013). They reported that, cost of media can be reduced by up to 70 per cent using different alternatives agents for *in vitro* production of orchid.

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