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EFFECT OF METHOD OF PLANTING OF *IN VITRO* PLANTLETS ON POTATO MINI-TUBER PRODUCTION UNDER PROTECTED CONDITIONS

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In potato cultivation, availability of healthy planting material is of utmost importance to ensure high productivities. Maintaining the seed stocks free from viruses under conventional (clonal multiplication) system of seed potato production is quite cumbersome. But, the recent advances in micro-propagation (tissue culture) techniques have facilitated the production, multiplication and maintenance of healthy potato clones. In this system, mini-tubers are produced by different ways either by direct transplanting of the whole *in vitro* plantlets under protected *in vivo* conditions (Lommen and Struik, 1992) or through aeroponics (Buckseth *et a*l., 2016). Direct planting of *in vitro* generated plantlets (micro-plants) in soil medium under aphid proof net/ polyhouses has also been reported to be a well established method for the production of potato mini-tubers in the hills as well as plains of India (Sharma *et al*., 2013; Kumar *et al*., 2011). Producing mini-tubers from *in vitro* plantlets allows a faster rate of multiplication and reduces the number of field generations needed in seed production (Ranalli, 1997).

Mini-tuber production is a classical way to multiply or acclimatize *in vitro* material before its use in the open field. This system creates a bridge between the *in vitro* rapid multiplication and the field multiplication of seed tubers (Sharma *et al*., 2013). However, transplanting of tender micro-propagated plantlets from *in vitro* condition to external environment has a high failure rate in some varieties (Chandra *et al*., 1992) and usually only one to two mini-tubers are formed on each plantlet (Garner and Blake, 1989). The objective of this phase of seed production system is to produce as many mini-tubers above a certain minimum size per *in vitro* plantlet as possible (Struik, 2007); and as early as possible (Hosseini *et al*., 2011). High production cost has always been an impediment to the adoption of tissue culture techniques, and it has further limited the technology to a few institutions and rich farmers while locking out the resource-challenged subsistence farmers. In order to increase application of tissue culture technology in potato farming, innovative approaches are needed to lower the cost of seed potato production (Venkatasalam *et al*., 2013). For achieving faster rate of multiplication and to explore the possibility of reducing the costs of plantlets an attempt has been made by using different planting methods (mainly cutting the plantlets into two halves; upper and lower) of healthy *in vitro* plantlets of variety Kufri Himalini.

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The study was conducted at Central Potato Research Station, Kufri (Fagu unit), 2700 feet above mean sea level during Kharif 2014 under the polyhouse. 21 days old *in vitro* Plantlets of potato cv. Kufri Himalini were used for study. Before planting, all the plantlets were hardened for one week under polyhouse conditions. Soil media in polyhouse consisted of soil: sand: farmyard manure in 2:1:1 ratio. Fertilizer mixture of nitrogen (N), phosphorus (P_2O_5) and potash (K_2O) in the form of calcium ammonium nitrate, single super phosphate and muriate of potash, respectively was applied at the rate of 50 kg each/ha in the soil one week before planting. At the time of planting, the rooting media was washed off and long adventitious roots were slightly trimmed. Thereafter, plantlets were prepared for the different treatments like i) full plantlet with slightly trimmed roots ii) full plantlet after cutting at the base (without root primordia), iii) and iv) plantlets after cutting in two halves *viz.* the upper and lower half (containing the root primordia) respectively **(Figure 1)**.

Planting was done during the first week of April and two plantlets were planted per hill at 30 × 10 cm spacing in 2.0 m long beds. A light irrigation was given before and after the planting of micro-plants. Thereafter, crop was irrigated according to its requirement. After about 21 days of planting, nitrogen in the form of calcium ammonium nitrate (10 kg/ha) was top dressed followed by earthing up. Each treatment was replicated four times. Haulms were cut after 120 days of planting. Data were collected on the percent plantlet establishment at 30 days after planting (DAP), plant height and number of shoots at 90 DAP and haulms weight at 120 DAP. At harvest, data on number and yield of mini-tubers/m^2 as well as on the proportions of large (>20g) and small (<3g) mini-tubers was recorded. The data was analyzed using the ANOVA, as per the standard procedure described by Gomez and Gomez (1984) and means were separated using the F test at 5%.

Fig. 1. Different planting methods viz., i) full plantlet with slightly trimmed roots, ii) full plantlet after cutting at the base (without root primordia) , iii) and iv) plantlets after cutting in two halves viz. the upper and lower half (containing the root primordia).

Establishment of plantlets was maximum (99.2%) in upper half segment and was at par to full plantlet with root (Control). The plantlet establishment was minimum with lower half segments. Initially, the rate of growth was apparently slow with lower half segments but it picked up at later stages. Finally, plant height after 90 days of planting was maximum in lower half segments and minimum with upper half counterparts (**Table 1**). Early picking up of growth with upper segments can be attributed to the inherent potential of different types of ex-plant tissues to reproduce into new plants. Sharma and Venkatasalam (2013) have also reported that the ex-plants from top portion of mother plantlets result in a faster rate of growth during culturing under *in vitro* condition. Number of shoots per plant did not vary significantly among the different

Table 1. Effect of method of planting of in vitro plantlets on the plant vigour.

Plantlet/segment	% plantlet establishment	Plant height (cm)	No. of shoots/plant	Haulms weight/plant (g)
Full plantlet (with roots)	98.5	94.4	1.80	80.8
Full plantlet (without roots)	98.0	94.0	1.81	78.2
Upper half	99.2	89.0	1.68	71.1
Lower half (with roots)	96.9	104.9	1.72	149.0
CD (p=0.05)	1.1	2.4	N.S.	7.8

treatments. Fresh haulms weight per plant (120 DAP) was significantly more with lower half segments and minimum with upper half segments (**Table 1**). It can be attributed to the late onset of plant growth with lower half segments than the full plantlets and upper half segments. Moreover, higher plant height with the lower segments may also be responsible for the gain in haulms weight. Minimum haulms weight with upper plantlets is also indicative of early plant maturity as compared to the lower half segments (**Table 1**).

Number and yield of mini-tubers/ m^2 was found to be maximum (144.9 and 2.03 kg/m^2 respectively) with upper half segment followed by full plantlet without roots and minimum (96.7 and 1.29 kg/m^2) with lower half segments (**Table 2**). Getting higher productivity with upper half segments may be on account of full exploitation of production potential as evidenced by early onset of growth, tuberization and ultimately early crop maturity as indicated by the low haulms weight in comparison to the crop raised from lower segments. In addition to higher productivity with upper half segments, proportion of <3g mini-tubers was also reduced by about 4.0% than full plantlets, whereas, proportion of such small (<3g) mini-tubers was same with rest of treatments. The proportion of large (>20g mini-tubers) was significantly more with lower half segments than other treatments. The average weight / mini-tuber did not vary significantly among the different types of plantlets and was found to be in the range of 12.6 to 14.3g (**Table 2**).

Using the plantlets in two halves, number and yield of mini-tubers/m^2 collectively with both segments (upper + lower) is found to be 75.8% and 79.5% higher respectively in comparison to the one obtained with the average of full plantlets used either way *i.e.* with or without roots (**Table 3**).

Thus, it can be concluded from the study that for the effective utilization of precious planting material, use of *in vitro* plantlets after cutting in two halves is a viable option for the production of mini-tubers. It will nearly double the number and yield of mini-tubers/ m^2, over the one got from full plantlets.

Table 2. Effect of method of planting the *in vitro* plantlets on the production of mini-tubers.

Plantlet/segment	No. of mini-tuber/m^2	Yield of mini-tuber/m^2 (kg)	>20g mini-tuber (%)	<3g mini-tuber (%)	Average wt./ mini-tuber (g)
Full plantlet (with roots)	134.6	1.70	19.9	28.6	12.6
Full plantlet (without roots)	140.2	2.00	20.2	25.1	14.3
Upper half	144.9	2.03	21.2	24.4	14.0
Lower half (with roots)	96.7	1.29	24.4	27.9	13.3
CD (p=0.05)	16.3	0.29	2.5	2.7	N.S.

Table 3. Percent gain in number and yield of mini-tubers with cutting of plantlets over the full plantlets.

	Plantlet/segment	No. of mini-tuber/ m^2	Yield of mini-tuber/ m^2 (kg)
1.	Full plantlet (with roots)	134.6	1.70
2.	Full plantlet (without roots)	140.2	2.00
a)	Average (1 and 2)	137.4	1.85
3.	Upper half	144.9	2.03
4.	Lower half (with roots)	96.7	1.29
b)	Total (3 and 4)	241.6	3.32
c)	% Gain in 'b' over 'a'	75.8 %	79.5%

Fig 2. Mini-tuber production after cutting of micro-plants in two halves. Sixty days old crop from upper (a) and lower half (b) segments of micro-plants

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