RESEARCH ARTICLE
COMPARATIVE GROWTH RESPONSE OF POTATO PLANTLETS DEVELOPED ON LIQUID VS SOLIDIFIED (MS) MEDIUM USING TISSUE CULTURE TECHNOLOGY
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ABSTRACT
The efficacy of liquid MS medium for potato multiplication was evaluated in this study with the objective to find a cost effective multiplication media for potato. The data was recorded for growth parameters i.e. no of days to shoot/root initiation, no of leaves, no of leaves and nodes, intermodal distance, root and shoot length at transplantable stage. Phenotypic differences in growth were observed between the plantlets of both types of media. Plantlets cultured on liquid media showed better growth of shoot and roots as compared to solid media. Liquid media plantlets emerged earlier and having greater number of leaves and nodes per plantlet. Shoot and root length was significantly greater in plantlets of liquid media with mean values 11.34cm and 1.72cm respectively, while in solid media, it was 6.04cm and 1.59cm respectively. The tuber yield and weight was also higher for plantlets developed on liquid media (2.91 and 2.04g) as compared to solid media plantlets (1.76 and 1.12 g).

INTRODUCTION
Potato (Solanum tuberosum L.) is a leading vegetable and the main cash crop of the world. It is a wholesome food and a good source of carbohydrates, vitamins, minerals and protein. It is also an important crop of Pakistan covering an area of 159.4 thousand hectares with a production of 3491.7 thousand tons, giving an average yield of 18.5 tons/ha during 2010-11 (MINFAL, 2011).

Potato production in Pakistan is considerably low because of a number of factors including viral diseases. Potato leaf roll virus (PLRV), potato virus X (PVX), potato virus Y (PVY), potato virus S (PVS) and potato virus A (PVA) are reported throughout Pakistan which causes severe losses in potato production (Khalid et al., 2000).

Tissue culture has become a powerful and successful tool for virus elimination from infected plants (Paet and Zamora, 1990) and has been successfully applied in vegetatively propagated species like potato for production of virus free plants (Prakash and Karihaloo, 2007). In vitro propagation by nodal cuttings has become an established method for rapid multiplication in potatoes (Dodds et al., 1991; Ranalli et al., 1994).

During the past decades, many types of media have been developed for in vitro plant culture (Pierik, 1989; Torres, 1989) with different formulations for the specific plants and tissues (Conger, 1981). Some tissues respond much better on solid media while others on liquid media. In general, the choice of medium is dictated by the purpose and the plant species or variety to be cultured. The most commonly used media for potato plant tissue culture is MS medium (Murashige and skoog, 1962). This basal medium is the combination of different kinds and concentrations of organic and inorganic salts, vitamins, phytohormones and carbon source.

Agar is the most commonly used gelling agent and an important ingredient of tissue culture media. Agar quality could affect the plant developmental processes, especially the regeneration of adventitious shoots and roots (Scholten and Pierik, 1998).

Agar can be the most expensive component of plant tissue culture media. The cost of commercial micropropagation can be drastically reduced if cheap alternatives to the highly expensive purified agar could be used. The use of ispaghol gelled medium in tissue culture is described as a means of reducing the cost of micropropagation by Shah et al., 2003.

It has been observed that low agar concentration media increase the growth of certain potato species cultivated in-vitro (Debergh, 1983; Rossel et al., 1987), due to increased availability of water and dissolved substances to the explants which on the other hand also lower the cost of production. Keeping in view these facts, the present study has been designed to observe the growth response of potato plantlets developed on solid (with agar) and liquid (without agar) media and its effect on tuber yield and weight in pre basic virus free potato seed production by using tissue culture technology in order to select a cost effective potato micropropagation technology for developing countries like Pakistan.

MATERIALS AND METHODS
The experiment was conducted in potato tissue culture laboratory at Hazara Agriculture Research Station, Abbottabad during the year 2011. The study was conducted on potato
cultivar ‘Desiree’. Basal Murashige and skoog (1962) (MS) medium containing 1.0 mg/l Ca-pentosethenate, 0.25 mg/l Gibberellic acid (GA3), 100mg/l Myoinsitol and 30gl/l sucrose at pH 5.7 was used in this study.

Single nodal cuttings from in vitro propagated virus free plantlets were cultured into the test tubes containing 10 ml MS medium under aseptic conditions. 50 replicates each for both types of treatments i.e solid and liquid medium were cultured. After inoculation the cultures were maintained in the growth chamber at 20 °C under 16/8 hours photoperiod with 30 µ mol/m2/s photosynthetic photon flux density provided by white fluorescent illumination for 30 days.

Data was recorded on various parameters including number of days to shoot initiation, number of days to root initiation, number of leaves, number of roots and number of nodes per plantlet, internodal distance, number of days taken to be transferred to the green house, shoot and root length at transferable plant stage.

After 30 days the plantlets were transplanted on sterilized peat moss media in pots in the green house, where they grow for 90 days to get mini tubers. The data was analyzed by using paired sample T-test at 5% significance level.

RESULTS

Phenotypic differences in growth rate were observed and the plantlets cultured on liquid media showed much better growth of shoot and root than solid media plantlets.

| Table 1 Relative no of days to shoot and root initiation in solid and liquid media |
|-------------------------------------------------|-----------------|-----------------|-----------------|
| Table 2 Mean values for different growth parameters of potato variety Desiree towards solid and liquid media |
| Type of media | Number of leaves | Number of nodes | Internodal distance | Shoot length | root length | No of roots | Number of days taken to transfer |
| Solid media   | 7.05±*            | 6.22±*          | 1.26±cm            | 7.12±cm      | 1.40±cm     | 3.83±*      | 31 days                         |
| Liquid media  | 9.08±*            | 7.97±*          | 1.51±cm            | 11.37±cm     | 1.92±cm     | 4.95±*      | 18 days                         |
| * = Mean values significant at probability level 0.05 in a column. |
| N0 = Mean values non significant at probability level 0.05 in a column. |

Data showed no significant difference in mean values for number of roots for solid medium (3.83) and liquid medium plantlets (4.95) however, the average number nodes and internodal distance differed significantly for both types of media (Table 2).

Figure 1 Mean values for number of leaves in solid and liquid media on 7th, 14th day and at transplantable stage

Shoot/root length at transplantable stage

The data pertaining to shoot/root growth for both media types as measured by shoot/root length is presented in Table 2. Plantlets of liquid media showed greater shoot/root length as compared to solid media plantlets (Fig. 2). Average shoot length of plantlets of solid media was 7.12cm which differed significantly from the shoot length of liquid media plants (11.37cm).

Number of days taken to transplantation

It was observed that plantlets of solid media took more number of days (31) to attain transferable plant height as compared to plantlets of liquid media which were ready for transplantation after 18 days (Table 2) (Fig. 2).

| Table 3 Mean values for tuber yield and weight of potato variety Desiree towards solid and liquid media |
|-------------------------------------------------|-----------------|-----------------|-----------------|
| Tuber yield and weight |

Data was recorded for tubers number and weight per plantlet. Mean values (Table 3) showed significant difference of tuber yield and weight for solid media (1.66 and 1.11g) from that of liquid media (2.91 and 2.17g), respectively. The tubers of liquid media plantlets were comparatively larger in size than solid media tubers (Fig. 3).
resistance to diffusion and closer contact between the explant and culture medium (Pierik, 1990; Smith and Spomer, 1994; Sandal et al., 2001). A study by Douglas (1984) demonstrated that Rhododendron shoots were ten-fold longer in liquid medium than on agar-solidified medium.

Upon harvesting higher tuber yield with increased tuber weight and size was recorded in liquid media plantlets as compared to solid media. The possible reason of this increase is that comparatively it may be due to vigorous and healthy growth of plantlets of liquid media, which ultimately results in better yield and larger tuber size. Good rooting systems observed in plantlets of liquid media increases the survival rate after transplantation in the soil and also give more yields as compared to plantlets of solid media.

In solid media, agar is most expensive constituent of culture media. The most important aspect of this study was to reduce the cost of virus free tuber production through micropropagation for a low income country like Pakistan and it can be achieved by using liquid media, because liquid media not only gives better growth of plantlets but also reduce the expenses in terms of cost of inputs by eliminating an expansive ingredient (agar) from the MS medium.

**DISCUSSION**

Comparative studies on growth of potato plantlets developed in liquid and solid medium revealed that the nodal sections cultured on liquid media showed better growth and are more vigorous as compared to solid media. The use of growth regulators in liquid cultures proved to be more effective and it is due to the direct contact of plant with the medium.

Comparatively early shoot and root initiation was observed in liquid media plantlets with faster growth rate and these were ready for transplantation only after 18 days. Mehrotra et al., (2007) described that the use of semi solid and liquid medium enhances early growth and reduced the time taken to multiply.

Also greater number of leaves and nodes with larger internodal distance in liquid media plantlets may be due to the fact that in liquid media plants develop roots earlier which facilitate absorption of nutrients resulting in rapid and vigorous growth. Pati et al., (2010) observed better response of a commercially important medicinal plant Catharanthus roseus for number of leaves in liquid medium as compared to solid medium. Similar Kuria et al., (2008) also reported more number of nodes per plantlet in liquid medium as compared to solid medium in potato. The shoot/root length was found to be greater in plantlets of liquid medium (Table 2). The direct contact of plantlets with the liquid medium contents causes ease in availability of nutrients resulting in better plantlet growth as compared to solid media where nutrients uptake is comparatively difficult due to solidification of medium. This increased nutrient availability may be induced by decrease in

**Fig. 2** Shoot length difference of potato plantlets for liquid (a) and solid (b) MS medium at transplantable stage

**Fig. 3** Potato tubers harvested from (a) solid media and (b) liquid media plants

**References**


Pati, P.K, J. Kaur and P Singh. 2010, A liquid culture system for shoot proliferation and analysis of pharmaceutically active

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