Enhancing Potato Seed Production Using Rapid Multiplication Techniques

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Introduction

Potato is regarded as a high-potential food security crop due to its ability to provide high yield and quality product per unit input with a short crop cycle (mostly <120 days). The national average yield at present is approximately 10.5 t/ha, which is lower than the world's average yield of 17 t/ha (Muthoni et al., 2011). The potential of potato crop has not been adequately exploited. The crop is mainly grown at high altitudes of 1,500–3,000 masl by small-scale farmers, who account for over 90% of the production. Most of the production is under rainfed conditions and carried out in scattered patches of intensive small-scale agriculture (McArthur, 1989).

There are a number of production problems. The major ones are unavailability and high cost of seed tubers; lack of well-adapted cultivars to the major agro-ecological zones; suboptimal agronomic practices; the prevalence of diseases and insect pests; and inadequate storage, transportation, and marketing facilities. To address these problems, the Ethiopian Institute Agricultural Research (EIAR)—the then Institute of Agricultural Research—in collaboration with the International Potato Center (CIP), initiated potato research. The research had as its main objectives to

- develop adaptable and high-yielding potato cultivars with good resistance to biotic and abiotic stresses;
- identify the best agronomic practices and storage systems;
- adopt the use of botanical seed as an alternative propagation method;
- develop seed production system in the country; and
- train farmers and other stakeholders.

Among potato production problems, a major bottleneck that contributes to low yield in Ethiopia is the lack of healthy and quality seed tubers in the required quantity and quality (Berga and Gebremedhin, 1994). There is no formal seed system operating for clean/healthy potato seed multiplication and distribution in Ethiopia. Hirpa et al. (2010) reported that potato seed production in Ethiopia is basically informal, which in most cases operates by recycling planting materials

from previous crop harvest. At present, 98.7% of the requirements for potato seed are met by informal seed supply. Sources of informal seed vary and include seed from neighboring farmers, friends, relatives, merchants, and markets where potato is sold for consumption. Moreover, it is a common practice to save the smaller size and inferior tubers that are not sold for consumption for seed purposes; consequently, this practice allows diseases to build up and yield may gradually decline, as seed gets degenerated.

Shortage of seed potato has been recognized as one of the most important factors limiting potato production in developing countries (Naik and Karihaloo, 2007). The production of clean seed is very crucial to sustain high production and productivity of potato in the country. Currently, the common method for propagation of important potato cultivars is through tubers. However, this propagation method has encouraged accumulation of tissue-borne viruses, fungi, and bacteria in subsequent seasons. This has led to significant losses in yield and tuber quality over the seasons (Tsoka et al., 2012). Therefore, the multiplication of clean tuber seed is an essential part of a strategy for organizing a potato seed program and involves different methodologies and approaches. A prerequisite to a successful and sustainable seed scheme is a continuous supply and maintenance of pathogen-free seed. Quality seed of an improved potato variety is key to increasing the productivity of a potato crop. The genetic potential and other traits of a variety are determined by the use of healthy and improved seed. This is true because the usual method of potato propagation throughout the world is using the vegetative seed tuber.

This paper seeks to review and assess the various options available for clean seed tuber production/multiplication and suggests the way forward, with particular emphasis on the applicability of rapid multiplication techniques (RMTs) such as tissue culture (TC) and aeroponics technologies for minituber production in Ethiopia.

The conventional method of propagation is one of the slowest of seed multiplication. Compared with other seed propagation techniques, like TC and aeroponics, this traditional method would create only about (1:10) daughter tubers in the course of a year (Otazu, 2008). This method has also shown to be time specific, particularly in tropical and subtropical regions where potato is a winter crop (Burton, 1989).

The main disadvantages of a conventional seed potato program are the low multiplication rate of field-grown potato plants, resulting in a slow and inflexible system, and the rising risk of catching viral, fungal, or bacterial diseases with an increasing number of field multiplication. A reduction in the number of multiplication years requires a propagule that can be produced in large numbers in protected environments in a short period (Lommen, 1995). However, pathogen-free planting material of selected potato varieties and clones are multiplied at Holetta and Jeldu research fields through a conventional method and distributed to growers to be used as a source of planting materials. To avoid diseases like bacterial wilt and viruses, a new system has been established for producing healthy seeds based on virus testing and in-vitro rapid multiplication of virus-free planting materials. Thus, different RMTs have been used for bulking up of prebasic and basic released potato varieties and promising clones for distribution to growers.

Rapid Multiplication Techniques

RMTs are extensive methods used to increase the amounts of nuclear seed stocks for further seed multiplication. RMTs provide better multiplication rates than the conventional method in vegetative increase of potato (Endale et al., 2008). The conventional method gives a lower multiplication ratio—ranging from 1:3 to 1:15—and more likely rapid virus infection. RMTs provide higher multiplication ratios (1:40–1: several thousand per year) and lower rate of contamination, particularly from soil- and seed-borne pathogens. Approximately 15% of the total area under potato cultivation around the world is used for the production of seed tubers. With the conventional propagation methods, potatoes are often prone to pathogens such as fungi, bacteria, and viruses, thereby resulting in poor quality and yield (FAO, 2008), whereas using healthy, quality seed is essential for growing an optimal potato crop (Parrot, 2010).

Different RMTs—TC-produced plantlets, stem cuttings, and aeroponics—have been used to bulk up selected potato varieties for multiplication and distribution to growers. Selected improved potato varieties of Tolcha and Menagesha have been multiplied using stem cuttings before the establishment of the system, and varieties Jalene, Gudene, Belete, and Awash have been multiplied using aeroponics (Table 1). For the stem-cutting activities, healthy and clean in-vitro plantlets received from TC and minitubers imported from CIP were planted in pots in screen houses where they get intensive care and management. Although these plants reach 20-30 cm high, the growth point of each stem was removed to stimulate growth of lateral shoots from the auxiliary buds. Cuttings, developed from auxiliary buds at each leaf, were then taken to root in moist, coarse sand at a distance of 5 x 5cm between individual cuttings for minituber production. Several techniques have been evaluated and proved to be suitable for minituber production, including aeroponics culture (Kang et al., 1996; Kim et al., 1999; Nugaliyadde et al., 2005).

In Ethiopia, techniques for minituber production that have been used include stem cuttings and TC-produced in-vitro plantlets that are then planted either in pots in screen houses or under aeroponics facility.

Variety	Year of p	Total				
	2011	2012	TOLAI			
Belete	38,390	67,078	105,468			
Gudene	864	1,278	2,142			
Awash	4,659	-	4,659			
Jalene	233	2,362	2,595			
Total	44,146	70,718	114,864			

Table 1: Number of minitubers produced under aeroponics at HARC in 2011/12

Source: CFC project annual report

Tissue Culture Techniques

Plant TC is the science of growing plant cells, tissues, or organ isolated from mother plant, on artificial media. This is facilitated using liquid, semi-solid, or solid growth media in sterilized tubes or containers. TC is one of the important new methods of plant propagation available to growers, and its use in seed production has allowed mass production of potato plants in a very short time. The system is characterized by very flexible rapid multiplication giving a high rate of multiplication (Beukema and Van de Zaag, 1990).

The TC technique employed in the micropropagation of potatoes consists of the aseptic cultivation of cells or fragments of plant tissues and organs in an artificial medium under controlled temperature and light conditions. Vigorous and disease-free potato plantlets can be obtained in the laboratory using this method, and then transferred to screen house in pots and aeroponics conditions for the production of minitubers. Moreover, the seed materials should be free of disease-causing pathogens. Clean stocks are first obtained by meristem culture, then these plantlets are transferred to seed beds, screen house in pots, and aeroponics to produce minitubers. Minitubers are commonly used in seed potato production to increase seed tubers (Öztürk, 2010). One of the advantages of this method is the maintenance of genotype identity, as meristem cells preserve their genetic stability more uniformly (Grout, 1990).

Minituber Production Using Aeroponics

Aeroponics is the process of growing plants in an air or mist environment without the use of soil or an aggregate media. Aeroponics refers to the method of growing crops with their roots suspended in a misted nutrient medium. This is an alternative method of soil-less culture in growth-controlled environments. Minitubers are those progeny tubers produced on in-vitro-derived plantlets. The term refers to their size, as they are smaller than conventional seed tubers but larger than in-vitro tubers (or micro tubers) produced under aseptic conditions on artificial media. The size of minitubers may range from 5 to 25 mm, although in current systems larger minitubers have also become common (Hassanpanah et al., 2009). Minitubers can be produced throughout the year and are principally used for the production of clean seed by direct field planting (Ritter et al., 2001). The use of minitubers in a seed program reduces the number of field multiplications. This may increase the flexibility of seed production, improve the health status of the ultimate seed, and reduce the time for adequate volumes of seed from new cultivars to become available for growers (Lommen and Struik, 1992). Though the technique is in its early stage, attempts have been made to improve production of healthy/high-quality planting materials.

Importance of aeroponics

Aeroponics method of propagation is one of the most rapid methods of seed multiplication. An individual potato plant can produce over 100 minitubers in a single row (Otazu, 2008). This contrasts with conventional methods that create only about 8–10 daughter tubers in a year and only 5–6 tubers per plant are produced using soil in the greenhouse in 90days(Hussey and Stacey, 1981; CIP, 2008). Another advantage of aeroponics is that nutrients and pH are easy to monitor. The system provides precise plant nutrient requirements for the crop, thereby reducing fertilizer requirement and minimizing risk of excessive fertilizer residues moving into the subterranean water table (Nichols, 2005) (Fig.1).

Farran and Mingo-Castel (2006) reported that soil-less production techniques, such as aeroponics, have successfully been employed in tuber production, with good prospects for certification in seed production systems. However, the worst drawbacks are the low volumes available to the root system and any loss of power to pumps that can produce irreversible damages. Traditionally, minituber production in sub-Saharan Africa countries is done on soil-base substrates that are steam sterilized to avoid soil-borne diseases and pests. However, rising prices of fossil fuels and scarcity of firewood used for the steam boilers render this practice almost prohibitive. The best alternative is to use nutrient solutions instead of soil substrates, which is a common practice of aeroponics already established for minituber production in industrialized Asian countries. CIP has developed an economic module that is being promoted for developing countries.

In aeroponics, plantlets are grown in specially designed boxes where shoots grow on top and roots grow suspended in the air within the box and in darkness. Roots are fed with pressurized nutrient solution mist at short intervals; as plants develop tubers are formed from stolons near the roots (Fig. 1). There are several advantages of this technology compared to the soil-based substrates for minituber production—namely healthier tubers due to the absence of soil-borne diseases, higher number of tuber set per plant, and reduced costs per minituber. In this system, minituber production is 10 times that of soil substrate(Fig. 2).



Figure 1: Foliage and root development under aeroponics conditions (Source: HARC, 2011)



Figure 2: Tuber development under aeroponic conditions (Source: HARC, 2011)

EIAR, with support from the Common Fund for Commodities (CFC) and USAID, established two aeroponics units at Holetta Agricultural Research Center (HARC) in 2010 for the production of minitubers of both popular and newly released varieties. The technique is suitable for early stages of seed multiplication in which the production operations are handled by the best technical support system. The technique is effective in giving high number of minitubers—up to 50 per plant—but adoption rates will be determined by availability of stable and low-cost power supply and expansion in the seed market. Minitubers produced in the aeroponics

unit are multiplied in the aphid-proof screen house/net house in HARC to produce seed (Fig. 3). The minitubers that are produced in the screen house and aeroponics are usually either planted in open field or in pots in the screen house based on the size of the tubers. The aim is to improve the health status of existing seed stock by reducing the number of field multiplications (Kleingeld, 1997). Harvesting in aeroponics is convenient, clean, and allows a greater size control by sequential harvesting (Ritter et al., 2001). The number and timing of nondestructive harvests are key factors in the optimization of minituber production. To optimize the system, appropriate nutrient solutions, plant densities, number of harvests and harvesting intervals, as well as possible interaction between them should be considered (Farran and Mingo-castel, 2006).

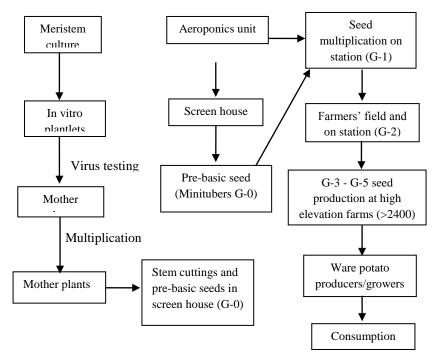


Figure3: Schematic diagram of prebasic seed potato production using RMTs at HARC

The multiplication and distribution scheme

In this program, high-quality planting materials are either imported from CIP's regional office in Nairobi or multiplied at Holetta and Jeldu research fields as well as TC laboratory. The materials are distributed to growers and different research centers for seed production or research purposes. A total of 1,669.5 tons of seed potatoes of 14 released potato varieties have been produced on station at Holetta and Jeldu from 2008 to 2011. The distribution of the materials to farmers was

practiced using farmer groups organized into Farmer Field Schools or Farmer Research Groups. This had laid the foundation for reaching more farmers.

The healthy planting materials/stocks multiplied using RMTs are of the selected varieties, which are maintained under strict hygienic conditions in an insect-proof screen house. Subsequent propagation is carried out using rooted cuttings in the screen house to obtain enough planting material before they are planted in open field. A total of 227,333 MT of several clones and released varieties were multiplied in screen houses which include Awash (1710 MT), Gudene (39,518 MT), Jalene (13,179 MT) and Belete (145.870MT). Source materials for this MT production were either in-vitro plantlets or smaller MT produced from the previous season in the aeroponics units. These materials have been introduced into the center's seed production program and have played a paramount role in regenerating the stock materials (Table 2).

Variety/Clones	2008	2009	2010	2011	2012	Total
Different clones	15,041	-	1,692	6,043	-	22,776
Jalene	3,760	-	7,440	1,979	-	13,179
Guassa	1,200	-	1,080	-		2,280
Gudene	1,800	-	1,352	9,231	27,135	39,518
Belete	-	-	-	42,871	102,999	145,870
Awash	228	228	-	1,254	-	1,710
Gorebela	1,264	-	-	-	-	1,264
Zengena	516	-	-	-	-	516
Tolcha	220	-	-	-	-	220
Total	24,029	228	11,564	61,378	130,134	227,333

 Table 2: Minitubers produced under screenhouses planted from in-vitro

 plantlets and small sized minitubers of aeroponics

Source: CFC project annual report

References

- Beukema, H.P. and D.E. Van der Zaag, 1990. Introduction to potato production. Pudoc, Wageningen, The Netherlands, 208pp.
- Bryan, E. 1981. Rapid multiplication techniques for potatoes. International Potato Centre. Lima, Peru, 20 pp.
- Burton, W.G. 1989. The Potato. Longman Scientific and Technical, Essex, UK. 470-504pp.
- Chiipanthenga, M., Maliro, M., Demo, P.,and Njoloma, J. 2012. Potential of aeroponics system in the production of quality potato (*Solanum tuberosumL.*) seedin developing countries. *Afr. J. of Biotechnol.* Vol. 11(17) 3993- 3999.
- CIP. 2008. Quality Seed Potato Production Using Aeroponics. Lima, Peru.
- Endale, G., W. Gebremedhin, and B. Lemaga. 2008a. Potato seed management. *In: Root and tuber crops: The untapped resources*,ed. W. Gebremedhin, G. Endale, and B. Lemaga, 53–78. Addis Abeba: Ethiopian Institute of Agricultural Research.
- Factor, T.L, Araujo, J.A.C de, Kawakami, F.P.C, and Lunck, V. 2007. Potato basic mini-tuber production in three hydroponic systems. 25(1): 82-87.

- Farran, I. and Mingo-Castel, A.M. 2006. Potato minituber production using aeroponics: effects of plant density and harvesting intervals. Am. J. Potato Res.83(1):47-53.
- Food and Agriculture Organization of the United Nations. 2008. International year of the potato, 2008. Retrieved Sept. 3, 2009, from http://www.potato2008.org
- Gebremedhin, W., G. Endale, and B. Lemaga. 2008. Potato variety development. *In: Root and tuber crops: The untapped resources*,ed. W. Gebremedhin, G. Endale, and B. Lemaga, 15–32. Addis Abeba: Ethiopian Institute of Agricultural Research.
- Grout, B.W.W. 1990. Meristem tip culture. I:pollard JW, Walker JM (eds)Methods in Molecular Biology: plant cell and tissue Cult. Humana Press, New Jersey. p. 597.
- Hassanpanah, D., Hosienzadeh, A.A., and Allahyari, N. 2009. Evaluation of planting date effects on yield and yield components of Savalan and Agria cultivars in Ardabil region. *Journal of Food, Agriculture and Environment* Vol. 7 (3and4): 525-528.
- Hirpa, A., Meuwissen, M.P.M., Tesfaye, A., Lommen, W.J.M., Lansink, A.O., Tsegaye, A., andStruik, P.C. 2010. Analysis of Seed Potato Systems in Ethiopia. *Am. J.Potato Res.* 87:537–552.
- Hussey, G. and Stacey, N.J. 1981. In VitroPropagation of Potato (*Solanum Tuberosum* L.). J. Ann. Bot. 48: 787-796.
- Jones, E.D. 1988. A current assessment of in vitro culture and other rapid multiplication methods in North America and Europe. *Am.Potato J.*65: 209-220.
- Kaguongo, W., P. Gildemacher, P. Demo, W. Wagoire, P. Kinyae, J. Andrade, G. Forbes, K. Fugilie, and G.

Thiele.2008.FarmerpracticesandadoptionofimprovedpotatovarietiesinKenyaandUganda. Working Paper 2008-5. International Potato Center (CIP), Lima, Peru.

- Kang, J.G., Kim, S.Y., Om, Y.H., and Kim, J.K. 1996. Growth and tuberization of potato (Solanum tuberosum L.) cultivars in aeroponic, deep flow technique and nutrient film technique culture films. *J. Korean Soc Hort Sci* 37:24-27.
- Kim, H.S., Lee, E.M., Lee, M.A., Woo, I.S., Moon, C.S., Lee, Y.B., and Kim, S.Y.1999. Production of high quality potato plantlets by autotrophic culture for aeroponic systems. J. Korean Soc. Hort Sci. 123:330-333.
- Kleingeld, C. 1997. The potential use of mini-tubers and the production volumes in South Africa. In: L.Urquhart(ed.), Potato short course: Potato production in SA with emphasis onKwaZulu-Natal. Agricultural Research council, Roodeplaat.Pp.193-198.
- Lemaga, B., and W. Gebremedhin. 1994. Prospects of seed potato production in Ethiopia. In: Proceedings of the second national horticultural workshop of Ethiopia, ed. E.Hareth and D. Lemma, 254–275. Addis Abeba: Institute of Agricultural Research and FAO.
- Lommen, W.J.M. 1994. Effect of weight of potato minitubers on sprout growth, emergence and plant characteristic at emergence. *Potato Res.* 37: 315-322.
- Lommen, W.J.M. 1995. Basic studies on the production and performance of potato minitubers. PhD thesis, Wageningen University, Wageningen, The Netherlands, 181p.
- Lommen, W.J.M. and Struik, P.C. 1992. Potato Production of potato minitubers by repeated harvesting: Plant productivity and initiation, growth and resorption of tubers. *Neth. J. Agr Sci.* 40:342-359.
- McArthur, Crissman, L. 1989. Evaluation, choice and use of potato varieties in Kenya. Social Science Department, Working paper 1989-1. International Potato Center. Lima, Peru, 44 pp.
- Naik, P.S. and Karihaloo, J.L. 2007. Micropropagation for Production of Quality Potato Seed in Asia-Pacific. Asia-Pacific Consortium on Agricultural Biotechnology, NewDelhi, India. 54 pp.
- Nichols, M.A. 2005. Aeroponics seed potatoes in North Korea, practical hydroponics and greenhouses. 81: 45-48

- Nugaliyadde, M.M., De Silva, H.D.M., Perera, R., Ariyaratna, D., and Sangakkara, U.R. 2005. An aeroponic system for the production of pre-basic seed potato. Annals of the Sri Lanka Department of Agriculture. 7: 199-288.
- Otazu, V. 2008. International Potato Center. Quality Seed Potato Production using Aeoponics. A potato Production Manual. Lima Peru.
- Otazu, V. 2010. Manual on quality seed potato production using aeroponics. International potato Center. Lima, Peru. 44 pp.
- Öztürk,G. and Yildirim, Z. 2010. A comparison of field performances of minitubers and microtubers used in seed potato production. *Turkish Journal of Field Crops*, 15(2): 141-147.
- Parrot, S.F. 2010. Five Stages of a Potato Plant eHow.com http://www.ehow. Com/list_6382 688_five stages-potato-plant.html#ixzz0qwbDsQ3G. University of Idaho: Potato Growth and Development.
- Ritter, E., Angulo, B., Riga, P., Herran, C., Relloso, J., and San J.M. 2001. Comparison of hydroponics and aeroponics cultivation systems for the production of potato minitubers. *Potato Res* 44:127-135.
- Stoner, R.J. 1983. Aeroponics versus Bed and Hydroponic Propagation. Florists' Review (1)173: 4477.
- Struik, P.C.and Wiersema, S.G. 1999. Seed Potato Technology. Wageningen Pers, Wageningen.
- Tsoka, O., Demo, P., Nyende, A.B., and Ngamau, K. 2008. Seed production of selected potato clones under aeroponic conditions. MSc. Desertation. Department of horticulture, Faculty Agriculture, Jomo Kenyetta University of Agriculture and Technology, Nairobi, Kenya.
- ——. 2012. Potato seed tuber production from *in vitro* and apical stem cutting under aeroponic system. Afr. J. Biotechnolo. Vol. 11(63): 12612-12618.
- Yildirim, Z. 1995. Microtuber production in potato (*Solanum tuberosum* L.). The Journal of Agricultural Faculty of Ege University (in Turkish). 32: 73-77.