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Multiplication of seed potatoes in a conventional potato breeding programme: A case of Kenya's national potato programme

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Abstract

In any breeding programme, speedy release of a good cultivar is important in order to reduce the costs and encourage innovativeness. In the Kenyan potato breeding programme, at least 10 tons of seed potato tubers are required before a new potato cultivar can be officially released. The large quantities of seeds required could have contributed to the slow release of new potato cultivars in the country. In addition, production of certified seeds from existing cultivars is low due to slow multiplication rate. Repeated field planting of clones in order to increase the amount of seeds generally puts the health status of such seed lots into danger due to exposure to viruses and soil-borne pathogens. Potato tuber bulking methods that have a high multiplication rate and at the same time reduce the number of field plantings should be adopted in order to produce planting materials of high health standards. Use of aeroponics system appears to be a good method for producing both basic seeds from true potato seeds or bulking of existing cultivars prior to production of certified seeds. Briefly indicate significance and purpose of this review article.

Keywords: Conventional potato breeding, Rapid multiplication, Seed potatoes.

Introduction

Speedy release of a good cultivar is important in order to reduce costs and motivate researchers in potato breeding programmes. In the Kenyan potato breeding programme, at least 10 tons of seed potato tubers are required before a potato cultivar is officially released (Kaguongo et al., 2010). This could have adversely contributed to the slow release of new potato cultivars to the detriment of over 800,000 small scale farmers in the country and the various consumers. For example, four potato cultivars (Kenya Karibu, Kenya Sifa, Kenya Faulu and Kenya Mavuno) preleased in 2002 could only be released in 2006 due to inability to produce the required 10 tons of seed tubers before the official release. Multiplication methods that ensure rapid production of this amount of tubers are always preferred. Rapid multiplication has the added advantage of ensuring high health status of the resultant crop thereby increasing the chances of a potential cultivar being released or a seed crop being certified. This advantage is easily lost when too many field multiplication cycles are applied as the seed potato crop is exposed to virus infection and soil-borne pathogens. This is critical during production of prebasic seed tubers, in clonal multiplication of promising clones and in production of certified seeds from newly released cultivars. In Kenya, there are four institutions producing prebasic seed potatoes (Kenya Agricultural Research Institute, National Potato Research Centre (KARI-NPRC) at Tigoni, and three other private companies) and only one public multiplier, Agricultural Development Corporation (ADC) which is a state parastatal. Combined, they only produce about 1.1% of all the certified seed potatoes required by farmers in the country (The Organic Farmer, June 2013). The main bottleneck in this formal seed supply system is the slow multiplication of basic seed into certified seed. Due to this, the seeds are highly priced and beyond the reach of many farmers. Consequently, farmer seed system currently dominates the potato sub-sector contributing about 96.3% of the total seed used while both 'clean' and 'positively selected' seed contribute 2.6% (Kaguongo et al., 2010). The widespread use of seed tubers from informal sources, whose health status cannot be guaranteed, has led to low yields, poor quality produce, and spread of pests and diseases (Riungu, 2011). This review paper looks at potato seed multiplication in a conventional potato breeding programme in developing countries such as Kenya.

Production of seedling tubers from true potato seeds (TPS)

One way of maintaining good health standards of early generations of TPS-derived materials is the use of nurseries or otherwise well-controlled environments to produce seedlings and seedling tubers. In producing potato tubers from true potato seeds, the most commonly used methods include: (1) direct sowing of TPS in the field for production of seed or ware tubers (Martin, 1983; Almekinders et al., 1996), (2) raising seedlings from TPS in a greenhouse or seedbed and transplanting them later into the field for production of seed or ware tubers in the same season (Rowell et al., 1986), and (3) direct sowing of TPS in the seedbeds at close spacing for production of seedling tubers for producing a commercial crop in the next season (Farook, 2005). Each propagation method has its advantages and disadvantages. Of the three propagation methods, use of seedling tubers is the most common (Almekinders et al., 1996; Simmonds, 1997). Seedling tubers can be produced off-season in a screenhouse thereby allowing another crop to be grown in the field at that time. In addition, use of seedling tubers avoids the problems associated with direct sowing and

transplanting of seedlings. Direct sowing results in slow seedling growth, high vulnerability of the seedlings to pests and diseases and high sensitivity to stress conditions such as high heat, frost and water limitations; all of which result in early tuberization and low yields. Transplanting seedlings results in transplanting shock leading to slow growth, poor stand establishment and hence low yields (Almekinders et al., 2009). The use of seedling tubers is agronomically similar to the use of tubers from conventional cultivars in terms of seed rate, initial crop development, number of tubers per stem etc (Almekinders et al., 1996). Also, the yield potential of seedling tubers competes well with that of clonal cultivars (Wiersema, 1984; CIP, 1987; Love et al., 1994; Benz et al., 1995; CIP, 1995). The common observation is that seedling transplants often have a longer growth duration, higher tuber set and smaller tuber size compared to plants derived from seedling tubers (Thomson, 1980; Chujoy and Cabello, 2007). However, seedling transplants often have a lower yield than seedling tubers (Gisela and Peloquin, 1991; Pael et al., 1998; Chujoy and Cabello, 2007). In situations where field conditions for direct seeding are not favourable or where the growing season is too short, raising seedlings in nursery beds and transplanting them into the field is a good alternative (Almekinders et al., 1996). This shortens the growing period of the crop in the field. Use of seedling transplants is advantageous in that the crop is raised from first generation plants derived from TPS and consequently, the health standard of this crop is optimal due to limited exposure to viruses and soil-borne pathogens (Struik and Wiersema, 1999). In addition, some seedling selection is possible through elimination of plants with low vigour or off-type plants during transplanting; this selection may enhance uniformity of the TPS family and improve crop performance after transplanting (Golmirzaie and Mendoza, 1986). Direct seeding or transplanting of seedlings for ware tuber production only seems to have potential in areas where the market accepts small tubers for consumption (Almekinders et al., 1996; Alekinders et al., 2009). Recent studies have found that seedling transplants produce more tubers per plant than the seedling tuber crop while the opposite was true when it came to total tuber yield (ton/ha) and proportion of ware-sized tubers (45mm< in diameter)(Jane et al., 2013). Kenya, just as in other developing countries, has no tradition for conventional potato breeding; local researchers have traditionally been evaluating advanced clones from the International Potato Centre (CIP) for adaptability to local conditions. Only recently has actual potato cross-breeding activities started at the National Potato Research Centre, Tigoni. From the foregoing, it might be prudent for these Kenyan potato breeders to consider using seedling transplants or seedling tubers to produce the initial prebasic seed tubers from their crosses.

Seed multiplication from promising clones and production of certified seed from existing potato cultivars

The conventional method of bulking seed potatoes is by repeatedly multiplying a set of tubers that have been proven to be disease-free in a process known as clonal multiplication (Bryan, 1981). This method has a low multiplication rate, about 6-8 daughter tubers per plant (Otazú, 2008). This means that one has to replant the seed tubers through many cycles before obtaining enough seeds for a cultivar to be formally registered. It therefore takes a long time before a cultivar is officially released; this is a major disincentive to conventional potato breeding. In addition, continuous field multiplication of the clones exposes the tubers to viruses and soil-borne pathogens especially bacterial wilt. This calls for regular disease management through thermotherapy thereby increasing the breeding period and raising the costs even further. To mitigate the bottlenecks caused by this conventional multiplication method, rapid seed potato multiplication techniques have been adopted which include micropropagation (tissue culture), hydroponics and aeroponics. Plant tissue culture is the science of growing plant cells, tissues or organs isolated from the mother plant, on artificial media. This is facilitated through the use of a liquid, semi-solid or solid growth media in sterilized tubes or containers. Tissue culture is characterized by a flexible and a high multiplication rate (Beukema and Zaag, 1990). Micropropagation can be divided into three stages: establishing the culture *in vitro*; propagating the materials and rooting; and transplanting and establishment in the soil (Murashige, 1974). The production of plants from axillary shoots has proved to be the most generally applicable and reliable method of *in vitro* propagation (Ng, et al., 1992). The two approaches that are usually used are shoot-tip culture and single node culture. The shoot tip is usually taken from the tender tip of the growing shoot(about 2 cm long), and the node cuttings are from either terminal or axillary buds with the stem segment attached. These two types of explants are preferred over meristem-tip culture in micropropagation when virus elimination is not part of the objective. Meristem-tip culture is the most commonly used for virus elimination in crop plants (Ng et al., 1992; Naik and Karihaloo, 2007; Badoni and Chauhan, 2010). Usually, meristem tips, about 0.5-1mm long and consisting of the meristematic dome and two leaf primordial, are excised from surface-disinfected apical or axillary buds and allowed to grow into plantlets on artificial nutrient media under controlled conditions. Generally, the percentage of virus-free plants obtained is inversely proportional to the size of the tips cultured. This technique is used for elimination of viruses in the planting materials as many viruses are unable to infect the apical/axillary meristems of a growing plant and a virus-free plant can be produced if a small piece of meristematic tissue is propagated (Wang and Hu, 1980; Kassanis, 2008). Elimination of viruses through tissue culture is possible because the vascular system through which viruses are spread is not well developed in the meristematic region. The high chromosome multiplication (due to high cell division) and high auxin content in the meristematic tissue possibly inhibit virus multiplication through interference with viral nucleic acid metabolism. Also, there exists virus inactivating system with greater activity in the apical region than elsewhere (Naik and Karihaloo. 2007). Heat treatment (thermotherapy) at temperatures ranging from 33 to 40^0C (constantly or alternatively) of the parent plant materials for a certain length of time, followed by meristem-tip culture has been reported to increase the number of meristem tips that may be regenerated to plantlets and the percentage of virus-free plants obtained from such meristem tips (Ng et al., 1992). Heat treatment prior to the excision of explants should be considered where it has proved difficult to eliminate viruses by meristem-tip culture alone. In addition to apical or axillary meristem culture, stem cuttings are made which consist of short stem pieces containing at least one node (Buck and Akeley, 1966). Stem cutting propagation is highly productive, efficient and cheap; a single plantlet can yield up to 100,000 plantlets in six months (Buck and Akeley, 1966). The lower surface of a stem cutting is dipped in uxin to enhance rooting and then planted out in trays containing sterilized sand. The plantlets root easily within a week and grow into perfectly normal potato plants to produce

minitubers. Tissue culture is not limited by the time of the year or weather; healthy plants can be grown in a laboratory at any time of the year. However, most developing countries fail to maximize on tissue culture technology due to high capital and operational costs involved; expensive specialized equipment is needed to set up the laboratory. In addition, different nutrients, energy sources, vitamins and growth regulators used for media formulation are also expensive (Badoni and Chauhan, 2010). The techniques of tissue culture require special skills and knowledge which can only be acquired after going through formal training. A lot of care and high level of hygiene are required in tissue culture; inadequate sterilisation of equipment can result in 100% contamination and complete loss of planting materials. Plantlets produced from tissue culture can be transferred into hydroponic system for rapid production of high quality disease-free minitubers for commercial use. In its basic principle, hydroponic system entails culturing plants in a nutrient solution containing balanced amount of essential nutrients that are necessary for plant growth and development. The main hydroponics systems available for the cultivation of leafy vegetables and potatoes are nutrient film techniques (NFT), the deep flow techniques (DFT) and aeroponics (Ricardo et al., 2009). The NFT system consists of series of PVC or asbestos-cement growing troughs with a 1- 4% slope, through which a thin film of nutrient solution (1 cm deep) flows over the roots of the plants. The solution is collected in a tank at the end of the slope and pumped back to the top of the channels by a submersible pump, thus allowing the constant circulation of the nutrient solution. The DFT hydroponics system involves a tank containing the nutrient solution (5-20cm deep), and the plants are placed on a platform with roots completely submerged in the solution. In this system, nutrient solution recirculation occurs through a typical entry-exit mechanism with the aid of a pump. The hydroponic method facilitates adequate supply of nutrients to the plant as well as permitting multiple harvesting of mini-tubers over a period of time thus increasing the yield of tubers compared to conventional method of using potted media. Aeroponic is seen as a more costly and complex hydroponic system that involves growing plants in an air or mist environment without the use of soil or an aggregate media. It refers to the method of growing crop with their roots suspended in a misted nutrient medium. Reports show that the system is ten times more successful than conventional pot techniques, tissue culture and hydroponics, which take longer and are also more labour intensive (CIP, 2008). The aeroponic system has the ability to conserve water and energy. The system uses nutrient solution recirculation hence, a limited amount of water is used. It offers comparatively lower water and energy inputs per unit growing area (Ritter et al., 2001; Farran and Mingo-Castel, 2006). Furthermore, the method is one of the most rapid methods of seed multiplication; an individual potato plant can produce over 100 minitubers in a single season (Otazú, 2008), as opposed to conventional potted media method that produces approximately 6-8 daughter tubers only in the same period (Hussey and Stacey, 1981; CIP, 2008; Otazú, 2008). Another advantage of this system is that of easy monitoring of nutrients and pH. The system provides precise plant nutrient requirements for the crop, thereby reducing fertilizer requirements and minimizes risk of excessive fertilizer residues moving into the subterranean water table (Nichols, 2005). In addition, the system allows the measurement of nutrient uptake over time under varying conditions. It is also space efficient, with plants taking up minimal room. In contrast with other techniques such as hydroponics and conventional system, aeroponics exploits better vertical space for root and tuber development (Stoner, 1983). As a result, many plants can grow at higher density (plants per unit area) than in the traditional forms of cultivation such as hydroponic and soil (Stoner, 1983). However, the system also requires constant power supply throughout the growing season and any prolonged interruption of power to water-pumps may lead to irreversible damage to the plants. The system has high installation and operational costs making it unsuitable for most developing countries. It also requires skilled labour. Both the hydroponics and aeroponics systems produce more tubers than the conventional pot method. In addition, they can be used to produce disease-free (virus- and bacterial wilt-free) potato seed tubers. Although use of seed tubers maintains genotype integrity of the potato (Grout, 1990), there are dangers of somaclonal variations that are induced during tissue culture (Lizarraga et al., 1989; Kaeppler et al., 2000). At the Kenyan potato breeding programme at KARI-NPRC, Tigoni, multiplication of disease-free seed potato tubers starts with tissue culture using meristem tip culture (KARI, 2007). The *in-vitro* plantlets produced are then multiplied 3 to 4 times in the laboratory using nodal cuttings. Six to seven weeks after the last multiplication step, the plantlets are transferred into the sand trays to harden off. After 15 - 20 days in the sand trays, plants should have formed enough root system and should be ready for transplanting into aeroponics boxes or pots for production of pre-basic seeds (generation 0) (Jane et al., 2011). The generation 0 seed tubers obtained are then multiplied in the field for three generations to produce basic seeds. The three generations (1, 2 and 3) are only meant to increase the amount of seeds. Aeroponics system has been promoted in Kenya because it produces a large number of disease-free seed tubers thereby eliminating the need for many field generations (beyond 3). This reduces seed production costs, saves time and ensures good healthy status of the seed tubers for further multiplication by private companies and selected institutions in the public sector. The prebasic (generation 0) minitubers that have been produced from aeroponics or from pots can be planted in sand trays in a similar way as explained for *in vitro* plantlets provided they have vigorous sprouts. These plants only require clean water for watering since emerging plants will depend on the mother tuber for nourishment. Depending on the cultivar and weather conditions, after two to three weeks in sand trays, plants should have formed small stems and enough root system for transplanting into aeroponics boxes. Depending on the apical dominance of seed, usually more than one stem per tuber is obtained. Mother tubers should be discarded(Otazú, 2010). When dealing with stem cuttings, cuttings from young branches should be obtained for rooting in sand boxes prepared in a similar way, as previously explained. Dipping the cuttings into a rooting hormone solution or powder just before placing each cutting into the sand will facilitate the rooting process. When they have developed enough root system, they are placed into aeroponics boxes just like in-vitro plantlets (Otazú, 2010).However, running an aeroponics unit is expensive because electricity is the main source of power used to operate the fertigation system(Mbiyu et al., 2012). In KARI-Tigoni, wind has effectively replaced electricity thus lowering the operational costs.

Conclusion

Seed potato tuber multiplication methods that have a high multiplication rate and at the same time reduce the number of field plantings should be adopted in order to produce planting

materials of high health standards. Use of aeroponics system appears to be a good choice for producing both basic seeds from true potato seeds or bulking of existing cultivars to produce certified seeds. Kenyan potato breeders should consider using aeroponics to produce basic seeds from seedling transplants or seedling tubers or to bulk the promising clones from their crosses.

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References

Almekinders CJM, Chilver AS, Renia HM (1996) Current status of the TPS technology in the world. Pot Res. 39*:* 289-303.

Almekinders CJM, Chujoy EThiele G (2009) The use of True Potato Seed as a pro-poor technology: The efforts of an International Agricultural Research Institute to innovating potato production. Pot Res. 52*:* 275-293.

Badoni A, Chauhan JS (2010) Conventional vis -a- vis biotechnological methods of propagation in potato: A review. Stem Cell. 1*:* 1-6.

Barak P, Smith JD, Krueger AR, Peterson LA (1996) Measurement of short- term nutrient uptake rates in cranberry by aeroponics. J Plant Cell Environ. 19*:* 237-242.

Benz JS, Keller ER, Midmore DJ (1995) Planting materials for warm tropic potato production: Growth and yield of transplanted seedlings or rooted cuttings and tuber materials in the field. Field Crops Res. 40*:* 179-192.

Beukema HP, Van der Zaag (1990) Introduction to potato production. Wageningen. The Netherlands.

Bryan E (1981) Rapid multiplication techniques for potatoes. CIP, Lima, Peru

Buck RW, Akeley RV (1966) How to obtain the most plants from one potato tuber. Ame Pot J. 43*:* 128-130.

Chujoy E, Cabello R (2007) The canon of potato science: The true potato seed (TPS). Pot Res. 50*:* 323-325.

CIP (1987) Annual Report 1986-1987. Centro Internacional de la Papa, Lima, Peru.

CIP (1995) Program Report 1993-1994. Centro Internacional de la Papa, Lima, Peru.

CIP(2008) Quality Seed potato production using aeroponics. Centro Internacional de la Papa. Lima, Peru.

Farook K (2005) Use of true potato seeds for better yields. Ph. D Thesis. University of Arid Agriculture, Pakistan.

Farran I, Mingo-Castel AM (2006) Potato minituber production using aeroponics: Effect of plant density and harvesting intervals. Ame J Pot Res. 83*:* 47-53.

Gisela CS, Peloquin JS (1991) Performance of true potato seed families. II. Comparison of transplants versus seedling tubers. Pot Res. 34*:* 409-418.

Golmirzaie AM, Mendoza HA (1986) Effect of early selection for seedling vigour on open-pollinated true potato seed. Ame Pot J. 63*:* 426 (Abstr).

Grout BWW (1990) Meristem tip culture .p. 597. *In* J.W. Pollard and J.M. Walker (ed.) Methods in molecular biology: Plant cell and tissue culture. Humana Press, New Jersey.

Hussey G, Stacey NJ (1981) *In vitro* propagation of potato (*Solanum tuberosum* L.). Ann Bot. 48*:* 787-796.

Jane M, Mbiyu M, Kabira JN (2011) Up-scaling production of certified potato seed tubers in Kenya: Potential of aeroponics technology. J Horticulture Forestry 3*:* 238-243.

Jane M, Kabira J, Shimelis H Melis R (2013) Producing potato crop from true potato seed (TPS): A comparative study. Paper presented at the 13th HAK Workshop on Sustainable Horticultural Production in the Tropics. Meru University of Science and Technology, Kenya. 3-6 December 2013.

Kaeppler SM, Kaeppler HF, Rhee Y (2000) Epigenetic aspects of somaclonal variation in plants. Plant Mol Biol. 43*:* 179-188.

Kaguongo W P, Ng'ang'a NM, Muthoka N, Muthami F, Maingi G (2010). Seed potato subsector master plan for Kenya (2009-2014). Seed potato study sponsored by GTZ-PSDA, USAID, CIP and Government of Kenya. Ministry of Agriculture, Kenya.

KARI (2007) Annual Report 2006, Kenya Agricultural Research Institute, Nairobi, Kenya.

Kassanis B (2008) The use of tissue cultures to produce virus-free clones from infected potato varieties. Int J Ann Appl Biol. 45*:* 422-427.

Lizarraga R, Huaman Z, Dodds JH (1989) *In vitro* conservation of potato germplasm at the International Potato Center. Ame Pot J. 66*:* 253-276.

Love SL, Thomson-John A, Werner B (1994) Evaluation of TPS lines for 1992 and 1993. Research Report. University of Idaho, College of Agriculture, Aberdeen, Idaho.

Martin MW (1983) Field production of potato from true seed and its use in breeding programme. Pot Res. 26*:* 219-227.

Mbiyu MW, Muthoni J, Kabira J, Elmar G, Muchira C, Pwaipwai P, Ngaruiya J, Otieno S, Onditi J (2012) Use of aeroponics technique for potato (*Solanum tuberosum*) minitubers production in Kenya. J Horticulture Forestry. 4*:* 172-177.

Murashige T (1974) Plant propagation through tissue culture. Annual Review Plant Physiol. 25*:* 135-166.

Naik PS, Karihaloo JL (2007) Micropropagation for the production of quality potato seed in Asia-Pacific. Asia-Pacific Consortium on Agricultural Biotechnology. New Delhi, India.

Ng, SYC, Thottapily G, Rossel HW (1992) Tisue culture in disease elimination and micropropagation. p. 171-182. *In* G. Thottapilly, L.M. Monti, D.R. Mohan Raj and A.W. Moore(ed) Biotechnology: Enhancing Rsearch on Tropical Crops in Africa. CTA/IITA co-publication. International Institute of Tropical Agriculture, Ibadan, Nigeria.

Nichols MA (2005) Aeroponics and potatoes. p. 201-206. *In* Proceedings of the First International Symposium on Root and Tuber Crops 'Food Down Under'. Leiden. Netherlands. Int Soc Hort Sci. (ISHS).

Otazú V (2010) Manual on quality seed potato production using aeroponics.Centro Internacional de la Papa, Lima, Peru.

Otazú V (2008) Quality seed potato production using aeoponics. A potato production manual. Centro Internacional de la Papa, Lima, Peru.

Patel PK, Pande PC, Sarkar BB, Kadian M (1998) Multilocation evaluation of seedling and seedling tubers derived from true potato seed (TPS). J Indian Potato Assoc. 25*:* 30-32.

Ricardo MC, Jose EDPP, Valdemar F, Ce'sar ABPP, Erika SR (2009) The production of seed potato by hydroponic methods in Brazil: Fruits, vegetables and biotechnology. Global Science Books 1*:* 133-139.

Ritter E, Angulo B, Riga P, Herrán C, Relloso J (2001) Comparison of hydroponic and aeroponics cultivation Systems for the production of potato minituber. Ame J Pot Res. 44*:* 127-135.

Riungu C (2011) No easy walk for potatoes. Horticultural News. The East African Fresh Produce Journal . 19*:* 16-17.

Rowell AB, Ewing EE, Plaisted RL (1986) General combining ability of neo-tuberosum for potato production from true potato seed. Ame Pot J. 63*:* 143-153.

Simmonds NW (1997) A review of potato propagation by means of seed, as distinct from clonal propagation by tubers. Pot Res. 40*:* 191-214.

Stoner RJ (1983) Aeroponics versus bed and hydroponic propagation. Florists' Review. 173*:* 44-77.

Struik PC, Wiersema SG (1999) Seed potato technology. Wageningen University Press. The Netherlands.

The Organic Farmer (June 2013) Vested interests cripple the potato industry. The Organic Farmer. The Magazine for Sustainable Agriculture in East Africa. 97: 2-5.

Thomson PG (1980) Breeding for adaptation to TPS propagation.p.149-157. *In* utilization of the genetic reosurces of the potato III. Centro Internacional de la Papa, Lima, Peru.

Wang PJ, Hu CY(1980) Regeneration of virus-free plants through in vitro culture.p. 61-99. *In* A. Flechter (ed.) Advances in biochemical engineering. Springer-Verlag, Berlin.

Wiersema SG (1984) The production and utilization of seed tubers derived from true potato seed. Ph.D. Thesis. University of Reading. UK.