PROCEEDINGS
OF THE
24th ANNUAL MEETING

August 15 to 20, 1988

Ocho Rios, Jamaica

Editor:
Walter I. Knausenberger
University of the Virgin Islands

Published by:
Caribbean Food Crops Society

IN VITRO TECHNIQUES FOR MICROPROPAGATION AND GERMPLASM CONSERVATION OF POTATO (*Solanum tuberosum*) IN JAMAICA

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ABSTRACT

In recent years, farmers in Jamaica have experienced difficulties in obtaining seed potatoes in time, which resulted in lose of yields and production. At the same time, there is a growing interest in the introduction of new varieties. Tissue culture techniques for rapid propagation and for the introduction of new varieties as well as germplasm conservation could provide answers for some of these problems. Twenty-one varieties which include the locally grown commercial ones like Desiree, Spunta, Kenebec and some promising new varieties like L.T. 5, 6, 7, 8, 8, 9 and Atlantic, etc. have been subjected to various techniques including single node cuttings, multinode cuttings, liquid media, solid media, liquid cultures without shaking, in vitro tuber production and culture at low temperature. Plantlets obtained through tissue culture have been succesfully introduced into the field with good yields of seed tubers and mother plants for vegetative propagation.

Key Words: Germplasm conservation; Micropropagation: Potato; *Solanum tuberosum;* Tissue culture

INTRODUCTION

Potato (*Solanum tuberosum*) has become one of the important tuber crops in Jamaica. Several small farmers in the hilly areas of St. Catherine and Manchester are involved in the cultivation of this crop. One of the major constraints for the expansion and improvement has been the late arrival of seed potatoes, which are usually imported from Canada and Holland. Due to the difference in the growing seasons of potatoes in these countries and Jamaica, and also because of some local problems in importing these, shipment of the seed potatoes is delayed for the planting season in Jamaica. This results in low yields due to late planting and /or reduces the total production in the country. A local production of planting material through rapid multiplication by tissue culture techniques would greatly enhance the capabilities of Jamaican farmers. Tissue culture techniques will also help in the introduction of new varieties as only a few plantlets under sterile conditions need to be imported. Maintenance of these and other promising varieties under in vitro conditions will ensure the availability for future experimental purposes.

The present work is an attempt towards developing suitable methods or modifying existing ones (Espinoza et al. 1986) for potato tissue culture in Jamaica.

MATERIALS AND METHODS

Twenty-one varieties of potato were obtained from the International Potato Centre in Lima, Peru. These include (Table 1) some locally used varieties like Desiree, Spunta, Kennebec and some promising new varieties specially developed for the tropics.

Micropropagation: Plantlets were removed from the tubes under aseptic conditions. They were gently blotted with a steril absorbent paper (This was obtained by sterilizing filter paper disks in petri dishes. These dishes were opened only under the laminar flow cabinet). Most of the large leaves and roots are trimmed and any sticking agar removed. Single node pieces with an axillary bud were cut and transferred to either tubes or majenta boxes with a solidified medium. The basal medium used was MS (Murashige and Skoog, 1962) with 1mM gibberellic acid (GA) and 1mM calcium pantothenic acid (CaP). These concentrations were determined after an initial experiment to find out the appropriate concentration needed. Sucrose was used at a 3% level and the pH was adjusted to 5.7 ± 0.1. The medium was solidified with Difco bactoagar (use of other brands did not give good results). The cultures were maintained at a day temperature of 26 ± 1°C and a night temperature of 20 ± 1°C and a light-dark cycle of 14h-10h, with a light intensity of 95μE m^{-2} sec. $^{-1}$

A second series of experiments were carried out with liquid media for micropropagation. The medium and the culture conditions were essentially the same as used for solid media except that the agar is omitted. In the initial experiments, it was observed that shaking did not have any considerable increased influence over the growth rates as compared with the cultures which are not shaken. Further experiments were carried out without shaking. Both single nodes and multinode cuttings were used for these experiments. (Fig.1).

In vitro tuber production: A two step procedure was used for the induction of tubers in vitro. In the first stage, plantlets from tubes were removed from the tubes and trimmed. Multinode cuttings were inoculated in conical flasks containing G9 medium (Gamborg et al. 1976) with 1mM GA, 1mM NAA, 1mM BA and 1mM CaP. Sucrose was reduced to 2%. The rest of the culture conditions were same as used for micropropagation. After three weeks when the plantlets have grown, the remaining medium was removed and a fresh medium without GA and with various levels of chlorocholine chloride (CCC) was introduced into the flasks. The sucrose concentration was increased to 8% and the cultures were kept in the dark at 8° C.

Germplasm conservation: Single node cutting cultures were subject to low light intensities of 10μE m^{-2} $sec.^{-1}$. and were kept at 20° C. Secondly, the in vitro tubers obtained were stored in the refrigerator for up to 6 months to test their viability.

RESULTS AND DISCUSSION

Micropropagation requires rapid growth and multiplication to facilitate the production of large quantities of plantlets. In the present experiments, each single node cutting produced a plantlet with an average of six nodes within six weeks. When majenta boxes with solid media were used, 15 - 20 nodes were inoculated, resulting in the production of 90 - 120 nodes in 6 weeks. Subculturing of plantlets from each tox resulted in the availability of up to 120 plantlets for hardening or 720 nodes for subculturing within 11 to 12 weeks. Use of liquid media instead of solid media resulted in the production of plantlets within four weeks instead of six weeks, thereby reducing the time for production. Also liquid media are economical, as agar is very expensive. Espinoza et al. (1986), obtained plantlets within three

weeks on liquid media with constant shaking on shakers. It took only a week more in the present experiments to achieve the results without shaking. Since the use of shakers require energy, cultured plantlets produced without shaking will be more cost effective in countries like Jamaica.

All the varieties tested for tuber production responded favourably to the two step procedure for in vitro tuber production. In four to six weeks, 2 to 10 mm size tubers were formed at many nodes on each plantlet. In Spunta, the tubers were exceptionally large, reaching up to 15 mm in size. The number of tubers ranged from 2 to 5 for each plantlet. This resulted in obtaining an average of 40 tubers per flask. While the best reeults were obtained with 3mM CCC, reducing the level up to 2.25 mM did produce a good number of tubers. Increasing the concentration had no further positive effect.

The cultures kept under low light conditions remained stable up to 8 months without the necessity for transfer or subculture. Similarly, the in vitro tubers could be stored up to six months in the refrigerator without losing the capacity to sprout.

For field propagation, majenta boxes or flasks with plantlets were taken out of the culture room and were kept under the screen house conditions without opening them, for one week. Later the plantlets were transferred to small pots with a mixture of sterilized peat and soil. These plantlets gave good results in yields of seed tubers and also served as good mother plants for vegetative propagation. Details of these will be published elsewhere.

The present work has clearly established a successful tissue culture programme for the rapid multiplication of potato in Jamaica.

ACKNOWLEDGEMENTS

Financial support for this work, provided by IICA in Jamaica and the Department of Botany, University of the West Indies, is gratefully acknowledged.

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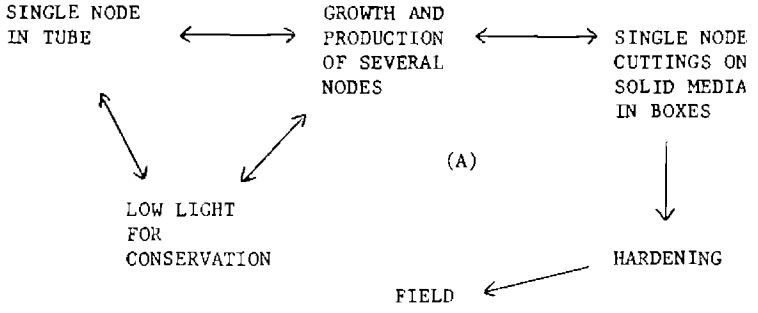
FIG. 1 : Schemes For Micropropagation (A) & In Vitro Tuber Induction (B)



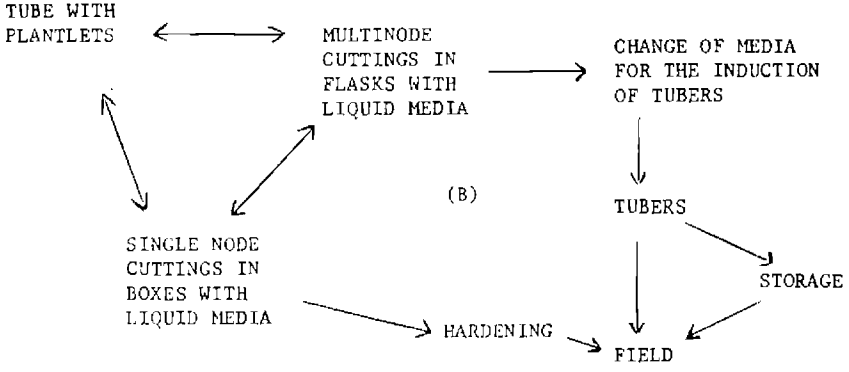


TABLE 1: Varieties Used In The Present Study

ATLANTIC	K. JYOTHI	PIROLA
BZURA	L.T. 2	P. CROWN
COSIMA	5	RADOSA
DESIREE	6	SATURNA
I 822	7	SPUNTA
I 1085	8	SUPERIOR
KENNEBEC	9	ULTIMUS