In vitro multiplication of sweetpotato - OP59

TITLE	**In vitro multiplication of sweetpotato - OP59**
OWNER	Head_IVU, B.Zea
APPROVER	Head Genebank
APPROVAL DATE	
ISSUE DATE	Feb 16, 2010 17:26
CONTRIBUTORS	
CITATION	
KEYWORDS	accredited , procedure , head_ivu , head_genebank , b_zea , op59
NEXT REVISION	
DOCUMENT ID	OP59
VERSION NUMBER	26

INTERNATIONAL POTATO CENTER - CIP

Version	Date	Author	Comment
26	Feb 16, 2010 17:26	Brenda Zea	
25	Feb 15, 2010 15:32	Luis Rojas	Changes given by Ana Panta
24	Feb 04, 2010 16:14	Alison Light	
23	Feb 03, 2010 09:31	Luis Rojas	
22	Feb 01, 2010 16:50	Alison Light	Header formatting
21	Nov 13, 2009 08:41	Maria Elena Vargas	Header formatting
20	Nov 12, 2009 17:45	Maria Elena Vargas	Header formatting
19	Oct 13, 2009 21:26	Ana Panta	
18	Sep 17, 2009 09:09	Maria Elena Vargas	
17	Mar 18, 2009 12:41	Brenda Zea	

INTRODUCTION

This technique is based on the principle that appropriate culture conditions induce the growth of a pre-existent bud, resulting in a new *in vitro* plantlet. The nutritional and hormonal conditions of the medium break the bud dormancy and promote its rapid growth. Callus formation, followed by plant regeneration, must be avoided because they can affect the genetic integrity of the clone. Following the protocol indicated below, after 4-5 weeks of sweetpotato *in vitro* culture, plantlets may be sub-cultured or transferred to soil in greenhouse.

SCOPE

This procedure covers the multiplication of *in vitro* sweetpotato germplasm.

SAFETY

No specific requirements above the normal laboratory safety procedures

See the following link for details of media preparation : Preparation of cultivation media

MATERIALS

Equipment	
Autoclave	Medium dispenser
PHmeter	Analytical balance
Laminar flow chamber	Shakers
Refrigerator	Cultivation growth chamber
Oven	Microwave oven
Other materials	
Glass test tube	Cotton
Petri dishes	Alcohol
Forceps	Burner
Blades	Sterilizer
Saran wrap	Used for multiplication
Parafilm	Used for distribution

See +Preparation of cultivation media procedure+for media recipes.

PROCEDURE

1. Under aseptic conditions, plantlets are taken out of *in vitro* vessels, and placed on a petri dish using sterilized forceps. It is recommended to use 4-5 week-old plantlets grown in propagation medium MPB (Table 1).

2. Using a sterilized knife handle with a No. 10 blade, remove the leaves and roots, and cut the stem in several segments of approximately 1.5 cm containing 1-2 buds.

3. Place one explant per 13x100 mm tube, two explants per 18x150 mm and 25x150 mm tubes, or five per magenta vessel on the surface of medium, maintaining the bud facing upwards.

4. Seal the tubes with a gas-permeable plastic tape and label correctly. Culture tubes for multiplication are sealed with saran wrap. Parafilm is used in the last culture for distribution.

5. Cultures are incubated in a controlled climate chamber at 23-25°C with a photoperiod of 16 h of light and 8 h of dark.

6. After 4-5 weeks of *in vitro* culture, plantlets are visually evaluated for growth development and bacteria/fungi contamination. Plantlets with abnormal growth or with contamination are incinerated.

7. Plantlets with normal development are ready for a new *in vitro* multiplication, or for distribution.

Table 1. Propagation media composition for sweetpotato *in vitro* culture (MPB)

Components	**Quantity**
MS salts (g/L)	4.3
Ascorbic acid (g/L)	0.2
Calcium nitrate (g/L)	0.1

Calcium panthotenate (mg/L)	2
L-Arginine (g/L)	0.1
Putrescine-HCl (mg/L)	20
Gibberellic acid (mg/L)	10
Sucrose (g/L)	30
Phytagel (g/L)	3
pH	5.7