

Original Article

## PERFORMANCE OF AN ACID-CASSAVA STARCH MEDIUM IN THE PROPAGATION OF FUNGI.

O.E. ADELEKE<sup>1</sup>, ODELOLA H.A<sup>2</sup>, Y. ONASANYA<sup>2</sup>.

<sup>1</sup>Department Of Pharmaceutics & Pharmaceutical Technology, Obafemi Awolowo College Of Health Sciences, Sagamu, Ogun State University. <sup>2</sup>Department of Pharmaceutical Microbiology and Clinical Pharmacy, College of Medicine, University of Ibadan, Ibadan.

Propagation of mould and yeast isolates was carried out on a formulated acid-cassava starch medium (A-CSM) and commercial potato dextrose agar (PDA). The two media were also used to enumerate some of the fungal isolates. Cultural and morphological characteristics expected of filamentous fungi and those with transitional forms were observed on both A-CSM and PDA. Growth was more rapid in respect of most of the fungi on A-CSM but it was accompanied with minimal liquefaction, after 48 hrs of incubation. Similarly, the A-CSM produced greater viable counts of most of the fungi. The acid-cassava starch medium reported is considered a potential alternative to its equivalent commercial nutrient media such as PDA.

Afr. J. Biomed. Res. 2(1), 1999

Key words: Cassava starch media, fungal propagation.

Running title: Acid-Cassava Starch medium in fungal propagation

### INTRODUCTION

Fungi are non-photosynthetic microorganisms that derive their nourishment on already elaborated foods, as saprophytes or from host tissues, as parasites (Vines and Rees, 1972). Fungi generally require for their propagation low temperature range of 0° to 35° with 20° to 30°C as optimum, though, some thermophilic species can tolerate up to 50°C (Conney and Emerson, 1964). They prefer an acid medium for growth (Alexopolus and Mims, 1980).

Fungi play important diverse roles in medicine (as sources of antibiotics and agents of diseases), industry (production of wine and alcohol) and agriculture (soil fertility). Hence, the need for suitable growth media that is readily available. Examples of established fungal media are potato dextrose, malt extract and Sabouraud dextrose agar. Using cassava starch powder from liver as broth and sodium chloride, Adeleke and Odelola (in press) have reported the formulation of an acid-medium (A-CSM) as a potential suitable alternative. The nutritive property of liver extract broth and saprophytic nutritional nature of fungi as well as their acidophilic growth preference are essential

factors that could favor fungal growth in the A-CSM.

This study represents a comparative report on the use of an acid-cassava starch medium and potato dextrose agar in the propagation/enumeration of ten selected fungi.

### MATERIALS AND METHODS

**Microorganisms:** *Aspergillus Orchiraceus*, *A. Funigatus*, *A. flavus*, *A. niger*, *Penicillium notatum* and *Trichoderma hamatum* were fungal isolates obtained from the Department of Botany and Microbiology, University of Ibadan (U.I.), Ibadan. Others were *Candida utilis*, *Saccharomyces uvarum* and *Neurospora solani*, from the Department of Food Technology, U.I. *Candida albicans* was obtained from the Department of Pharmaceutical Microbiology and Clinical Pharmacy, U.I.

The fungal isolates were preserved on potato dextrose agar (PDA) slants at 4°C.

**Cassava Starch Media:** The acid-cassava starch medium (A-CSM) of pH 4.8 was compounded with cassava starch powder

(10%); liver extract broth and sodium chloride as previously reported (Adeleke and Odelola, in press).

#### Propagation of fungal isolates

For observing the cultural and morphological characteristics, the filamentous fungi were propagated by fragmentation whereby filaments of growing mycelia were teased on dry, sterile A-CSM and PDA plates. The remaining fungi were cultivated by surface spread using a 4mm-loopful of their respective broth cultures, on the same media. The plates were incubated at 27°C for 24-72 hrs. Viable counts were carried out on the transitional fungal forms by plate count technique. The fungal broth cultures were each diluted up to  $10^{-6}$  from which 0.2ml, was seeded into each of A-CSM and PDA. Following incubation at 32°C for 24-48 hrs, colony-forming units were counted on a colony

counter (Gallenkamp) and number of cells per ml, was estimated.

#### RESULTS

All the fungal isolates grew within 48 hrs of incubation on both A-CSM and PDA, with their usual cultural and morphological characteristics such as shape, colour size, surface appearance and others. No liquefaction accompanied the growth observed within 48 hours on both media but after 48 hours, only three of the isolates exhibited liquefaction on A-CSM alone (Table 1).

Viable counts of the transitional forms of fungi tested virtually indicated a relatively higher rate of fungal growth on A-CSM than on PDA. For instance, *Neurospora solani* gave a count of  $1.250 \times 10^9$  cells per ml on A-CSM against  $2.15 \times 10^7$  cells per ml on PDA (Table 2).

**Table 1**  
**Growth of fungal isolates on acid-cassava starch medium (A-CSM) and potato dextrose agar**

Organism	A-CSM		PDA	
	24-48hrs	72hrs	24-48hrs	72hrs
<i>Aspergillus orchiraceus</i>	+	++ (No liquefaction)	+	++ (No liquefaction)
<i>A. niger</i>	+	++ (No liquefaction)	+	++ (No liquefaction)
<i>A. flavus</i>	+	++ (liquefaction present)	"	"
<i>Penicilliumnotatum</i>	"	"	"	"
<i>Trichoderma hamatum</i>	"	"	"	"
<i>Aspergillus Fumigatus</i>	"	++ (No liquefaction)	"	"
<i>Neurospora solani</i>	+	"	"	"
<i>Candida albicans</i>	"	"	"	"
<i>Candidida utilis</i>	"	"	"	"
<i>Saccharomyces uvarum</i>	"	++ (liquefaction present)	"	"

KEY: + = growth . ++ = Increased growth.

Table 2.

## VIALE COUNTS OF SOME FUNGI ON A-CSM AND PDA

Organism	A-CSM	PDA
<i>Neurospora solani</i>	1.250 x 10 <sup>9</sup> cell/ml	2.15 x 10 <sup>8</sup> cells/ml
<i>Candida utilis</i>	3.50 x 10 <sup>8</sup> cells/ml	2.80 x 10 <sup>8</sup> cells/ml
<i>Saccharomyces uvarum</i>	7.50 x 10 <sup>8</sup> cells/ml	1.75 x 10 <sup>8</sup> cells/ml
<i>Candida albicans</i>	1.000 x 10 <sup>9</sup> cells/ml	2.500 x 10 <sup>9</sup> cells/ml

## DISCUSSION

The ability of the A-CSM with pH 4.8 to support fungal growth in the same manner as PDA emphasises the preference of fungal growth on elaborated foods (vines and Rees, 1972) in an acid medium, (Alexopolus and Mims, 1980). The minimal hydrolysis observed on A-CSM in respect of three of the fungal isolates, suggests the potential suitability of cassava starch in detecting microbial starch hydrolysis, a property that is usually observed with commercial soluble starch (Iverson and Millis, 1974; Harrigan, 1976).

The observation that the hydrolysis was a function of the length of incubation period (Twomey and Machie, 1985) could be utilised in the study of other properties of microorganisms (besides hydrolysis), by restricting the incubation period to 48

hours. This suggestion is favoured by the higher rate of fungal growth, which was apparent in greater viable counts obtained on A-CSM, reflecting a more intense fermentative activity by the fungi (Alexopolus and Mims, 1986) on A-CSM relative to PDA.

The results reported in this study on growth and viable counts of fungi on A-CSM should facilitate further work to perfect the use of starch powder from cassava tubers in the formulation of culture media. The medium would be found a suitable alternative to its equivalent commercial media if its major ingredients could be compounded into dehydrated powder. The slimy characteristics of A-CSM during inoculation and its inability to be stored sterile and re-melted in appropriate media bottles, have to be addressed before such formulation could be desirable.

## REFERENCES

- Adeleke, O.E. & Odelola, H.A. (in press).** Formulation of Novel Microbiological Media with Cassava Starch. Nig. J. Sci.
- Alexopolus, C.J. & Mims, O.W. (1986):** Introductory Mycology. 3rd Ed. Willy Eastern Ltd. Dehli pp. 235-277.
- Conney, D.G. & Emperson, R. (1986).** Thermophilic fungi. 9th Ed. Pacific Book, Pala Alto, Falif.
- Harrigan, W.F. (1976).** Laboratory methods in Food and Dairy Microbiology. Revised Ed. Academic Press, London, New York San Francisco. Pp. 72 & 366.
- Iverson, W.G. & Millis, N.F. (1974).** A method for the detection of starch hydrolysis by bacteria. J. Appl. Bacteriol. 16, 126-130.
- Twomey, D. G. and Mackie, D. L. (1985).** A medium for the rapid growth and detection of moulds. Lts. Appl. Microbiol. 1: 105 - 107.
- Vines, A.E. & Rees, N. (1972).** Plant and Animal Biology Vol. 1 4th Ed. Pitman Publishing Kingsway London. Kenya pp. 1199-1202.