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Short Communication

Selection of Marine Yeasts for the Generation of Single Cell Protein from Prawn-shell Waste

Abstract

Marine yeasts (33 strains) were isolated from the coastal and offshore waters off Cochin. The isolates were identified and then characterized for the utilization of starch, gelatin, lipid, cellulose, urea, pectin, lignin, chitin and prawn-shell waste. Most of the isolates were Candida *species. Based on the biochemical characterization, four potential strains were selected and their optimum pH and NaCl concentration for growth were determined. These strains were then inoculated into prawn-shell waste and SCP (single cell protein) generation was noted in terms of the increase in protein content of the final product. © 1998 Published by Elsevier Science Ltd. All rights reserved*

Key words: Marine yeast, characterization, prawn-shell waste, single cell protein.

INTRODUCTION

Marine yeasts are reported to be highly versatile agents of biodegradation (De souza & D'souza, 1979; Kobatake *et al.*, 1992). Utilization of this potential in the biotransformation of cheap raw material or waste matter into value-added products could be a highly rewarding endeavour. Partial conversion of the raw material into yeast biomass (SCP) is useful because of the high nutritional quality of the yeasts, and this would give possible uses in aquaculture or animal feed formulations. The advantages of microbial protein are high productivity, a high proportion of cell mass as protein, a good profile of desirable amino acids, good performance in feeding livestock and no toxic or carcinogenic compounds. Besides these, the yeasts have the advantages of large size, low nucleic acid content, long history of use as food and ability to grow at low pH (Mitchel, 1974).

The present work was aimed at the isolation and characterization of marine yeasts from various environments and testing their potential in the generation of SCP from prawn-shell waste.

METHODS

Water samples were collected from coastal as well as offshore waters off Cochin. The water was collected in presterilized bottles and transported to the laboratory in an ice box. The samples were enriched by growth in malt extract broth (malt extract, 30 g; mycological peptone, 0.5 g; sea water, 1000 ml; pH 5.5), incubated at room temperature (28 ± 2°C) for 48–72 h. The culture broth was then streaked onto malt extract agar and the yeast colonies were isolated. The isolated yeast strains were classified according to Barnett *et al.* (1990). Characterization of the strains was carried out by checking their utilization of starch, gelatin, cellulose, chitin, pectin, lignin and urea, and measuring their growth on prawn-shell agar plates (dried prawn-shell powder, 10 g; agar, 2 g; sea water, 1000 ml). Optimum pH and NaCl concentrations for the growth of the selected strains were determined using malt extract broth.

Prawn-shell waste was collected from different peeling sheds at Cochin, dried in an oven at 80°C overnight, powdered well and the protein content was analysed on a dry weight basis as per Lowry *et al.* (1951). This shell waste was a mixture of shells from different prawns such as *Penaeus indicus*, *P. monodon* and *Metapenaeus dobsoni*. Selected yeast strains were grown in malt extract agar slants for 18 h and then harvested using sterile sea water. The optical densities of the cell suspensions were adjusted to 1.00 at 640 nm and these cell suspensions were then used as inoculum.

Upgrading of the prawn-shell waste through SCP production was performed by inoculating 1 ml of the selected cultures into prawn-shell broth (dried prawn shell powder, 10 g; sea water, 100 ml; pH 5). Immediately after inoculation (0 h) and again after 7 days of incubation at room temperature (28 ± 2°C), the protein content was estimated using the method of Lowry *et al.* (1951). After incubation, the contents of the flasks were in a semi-solid condition and the whole culture along with the flask was dried in an

Table 1. Protein enrichment of raw material (prawn-shell waste) by SCP production

Culture no. and genera	Concentration of protein (% on dry weight basis)	
	Initial (a)	Final (b)*
M10 *Candida*	38.5	65.2±3.1
M15 *Candida*	38.5	70.4±2.8
M23 *Rhodotorula*	38.5	62.9±3.6
M28 *Rhodotorula*	38.5	60.6±2.1
Mixed culture (M10+M15+M23+M28)	38.5	69.3±3.6

*Mean±SD. $N = 4$

oven at 80°C for 16 h. The protein estimation was then performed.

RESULTS AND DISCUSSION

Of the 33 cultures isolated, 27 were *Candida* species, 3 were *Rhodotorula* and 3 were *Leucosporidium*.

All the yeasts were found to be amylolytic and most of them were lipolytic. Only a few showed proteolytic, chitinolytic, cellulolytic and ureolytic activity and none were capable of pectin utilization. All the strains had nitrate-reduction capacity and one strain (M23, *Rhodotorula*) was capable of producing starch-like substances. All strains except M4 (*Candida*) and M5(*Candida*) showed growth on prawn-shell agar plates where the shell served as the sole nutrient source, and certain strains exhibited notably good growth and proliferation.

Based on the various physiological and biochemical characteristics studied, four strains were selected for further studies. They were M10 (*Candida*), M23 (*Rhodotorula*), M15 (*Candida*) and M28 (*Rhodotorula*).

Extent of growth on prawn-shell waste was one of the most important criteria used for the selection of the strains. All the four strains selected exhibited very good growth on prawn-shell agar plates. The hydrolytic properties of the isolates were considered as criteria for the selection of the strains. M10 was chitinoclastic and proteolytic. M15 was chitinoclastic and non-proteolytic. M23 produced starch-like substances and was lipolytic and amylolytic, but lacked proteolytic and chitinoclastic potential. M28 was proteolytic and lipolytic but non-chitinoclastic. Two per cent NaCl was found to be optimum for M23 and M15, whereas 2.5% NaCl was optimum for M10 and 1% for M28. The optimum pH of M15, M23 and M28 was pH 5. M10 (*Candida*) exhibited a wide pH tolerance from 4 to 10 with very little difference in growth. Low growth was noticed in the alkaline range for the other strains.

Examination of prawn-shell waste from different peeling sheds showed that the protein concentration ranged from 29 to 41% in the various samples. The protein content of the final products (i.e., after biotransformation using the yeast strains) varied from 60.6 to 70.4% (Table 1). The protein enrichment was found to be maximum with M15 and minimum with M28. In this study the non-proteolytic and chitinoclastic combination was best. Conservation of the already available protein along with a further enrichment in protein through SCP generation using the chitinoclastic potential is the reason for this potential yield by M15. The biotransformation of prawn-shell waste utilizing selected strains showed that they were highly effective for this purpose. This product could find wide application in aquaculture particularly as a feed supplement which would be economically viable and cost effective. Moreover the prawn-shell, a waste from the shrimp processing industry becomes a valuable raw material for a novel industry aimed at microbial transformation of prawn-shell waste into aquaculture feed which would contribute to pollution abatement and recycling of waste back into the food chain.

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R. Rhishipal & Rosamma Philip*

School of Marine Sciences, Cochin University of Science and Technology, Lake Side Campus, Kochi-682013, Kerala, India

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*Author to whom correspondence should be addressed.