

ISOLATION, IDENTIFICATION AND SCREENING OF CELLULOLYTIC MICROORGANISMS FROM SOIL AND ORGANIC WASTE

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INTRODUCTION

Cellulose degradation and its subsequent utilization is important for global carbon sources. The value of cellulose as a renewable energy source has made hydrolysis of this compound the subject of intense research and industrial interest.

OBJECTIVE

The present research was conducted to find out the utilization effect of microorganism that may be likely adapted for the hydrolysis in first methane fermentation stage.

MATERIAL AND METHODS

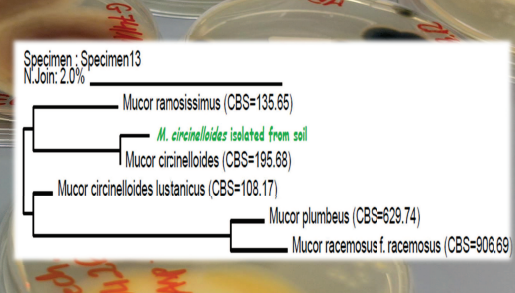
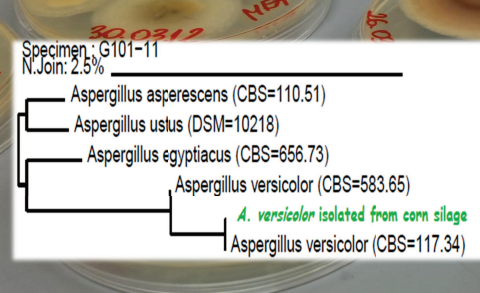
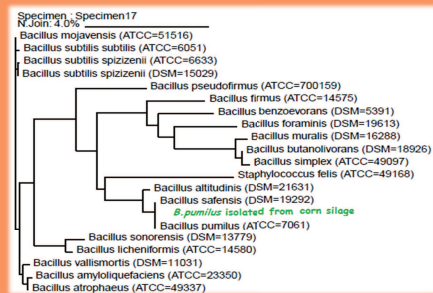
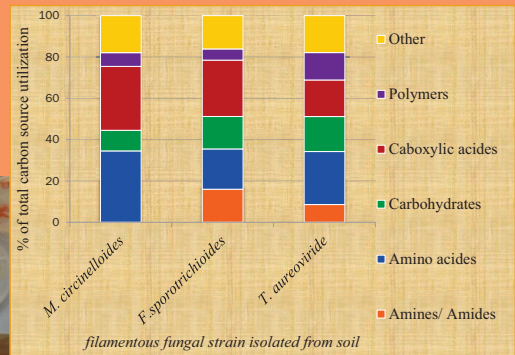
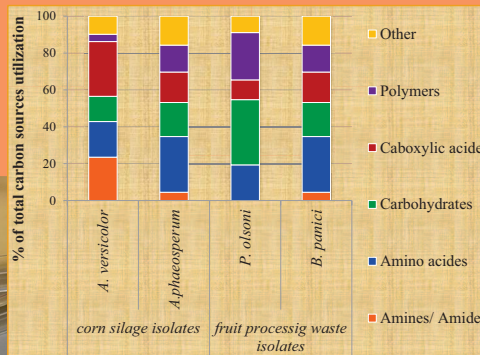
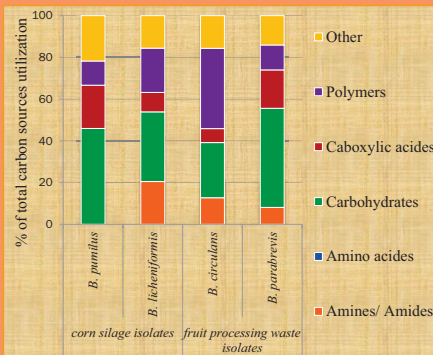
Microorganisms were isolated from soil, corn silage and fruit processing waste. The bacteria and fungi were cultured on agar medium with appropriate soil or waste extract and Martin medium, respectively. Screening of cellulose producers was done on minimal medium with 2% of cellulose addition. After plates inoculation, these were incubated at 26°C for 120 hours and flooded with Gram's iodine for every 24 hours and the zone of clearance around the colony were observed and measured.

Microorganisms identification was done using sequencing analysis (MicroSEQ) with universal primers for bacteria (16S-rDNA) and fungi (D2-LSU). Metabolic characterization of microorganisms was done using BIOLOG™ system. The plates GEN-III and FF were used respectively for bacteria and fungi characterization. The metabolic pattern of particular guilds group (carbohydrates, amino acids, amines and amides, carboxylic acids, polymers and miscellaneous) was assessed for all microorganisms.

Specimen	# Samples	Bas calling	Filter	Ass embly	Specim en Score	Top Match	% Match	Consensus Length	Library Entry Length	Comments
Fungal strain 1	2	✓	✓	✓	28	Bipolaris panici-milacei (DAOM=171971)	99.97	180	279	
Fungal strain 2	2	✓	✓	✓	26	Penicillium ochroni (CBS=232.6)	97.52	246	281	fruit processing waste isolates
Bacterial strain 1	2	✓	✓	✓	33	Brevibacillus parabravis* (ATCC=10027)	99.97	489	484	
Bacterial strain 2	2	✓	✓	✓	32	Bacillus circulans (ATCC=4513)	98.85	441	493	

Specimen	# Samples	Bas calling	Filter	Ass embly	Specim en Score	Top Match	% Match	Consensus Length	Library Entry Length	Comments
Fungal strain 1	2	✓	✓	✓	24	Arthrinium phaeospermum (CBS=142.55)	94.10	145	278	
Fungal strain 2	2	✓	✓	✓	25	Aspergillus versicolor (CBS=117.34)	99.98	178	279	corn silage isolates
Bacterial strain 1	2	✓	✓	✓	32	Bacillus pumilus (ATCC=70617)	99.98	469	495	
Bacterial strain 2	2	✓	✓	✓	35	Bacillus licheniformis (ATCC=14580)	99.85	483	497	

Specimen	# Samples	Bas calling	Filter	Ass embly	Specim en Score	Top Match	% Match	Consensus Length	Library Entry Length	Comments
Fungal strain 1	2	✓	✓	✓	39	Mucor circinelloides (CBS=195.68)	99.70	369	384	
Fungal strain 2	2	✓	✓	✓	43	Trichoderma aureoviride (DAOM=175924)	99.22	277	277	soil isolates
Fungal strain 3	2	✓	✓	✓	42	Fusarium sporotrichoides (DSM=62423)	99.63	278	278	



RESULTS

Molecular identification indicated that investigated microorganisms belonged to the following bacteria and fungi genus: *Bacillus*, *Brevibacillus*, *Mucor*, *Fusarium*, *Aspergillus*, *Trichoderma* and *Penicillium*. Tested strains have potential in degradation of cellulolytic compounds, which could be useful in the first stage of methane fermentation process.