

### PHYTASE PRODUCTION BY Aspergillus niger AND Aspergillus terreus IN SOLID STATE FERMENTATION USING WHEAT BRAN

**en** años

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#### INTRODUCTION

Animal nutrition industry demands for phytases with high specific activity and stability under high temperature conditions (feed pellet production) and under acidic conditions (monogastric animals), for this reason continues to stimulate the search for new enzyme sources [1]. Most of scientific work has been focused on Aspergillus spp. phytases due to their biochemical properties found [2]. An alternative for phytase production is solid state fermentation (SSF), it offers several advantages over submerged fermentation (SmF), as hiperglycosylated proteins, making them more stable, higher yields and productivities and can be used agroindustrial residues as substrate-support [3]. Wheat bran (agroindustrial residue) is one of the most used in phytase production by SSF, due to their high phytic acid concentration (1-2%) compared to other residues. It is important to give an adequate pretreatment to the residue in order to be used for phytase production avoiding contamination and preventing phytic acid hydrolysis [4]. Filamentous fungi (164) belonging to CIATEJ collection, isolated from different agroindustrial wastes and unconventional environments (mescal, cocoa fermentation, coffee pulp, etc.), were grown in a media using phytic acid as sole source of carbon and phosphorus in order to find phytase-producing candidates. A screening method using 15 mL tubes fermentation system was performed to assay different fermentation conditions at the same time, obtaining rapid and reliable results. The best fermentation conditions found using 15 mL tubes as reactor were: wheat bran without wash and UV exposed for 2h and culture media A; reaching a maximum phytase activity of 1.91±0.25 U/gdm at 192h of fermentation (Fig. 1-A). The other wheat bran pretreatments as well as culture media B do not present significant phytase activity (Fig. 1-B). Maximum phytase activity reached using columns reactors were: 4.2±0.71 U/gdm at 96h of culture for Aspergillus niger and 0.49±0.005 U/gdm at 96h for Aspergillus terreus (Fig. 2).

# **METHODOLOGY** Substrate-Support Pretreatments: Washed Wheat bran

### Impregnation Media:

Medium A	g/L
Sucrose	10
MnSO <sub>4</sub>	0.1
MgSO <sub>4</sub>	0.5
Fe <sub>2</sub> (SO <sub>4</sub> ) <sub>2</sub>	0.1
KCI	0.5
Yeast	5
extract	

	Medium B	g/L
	Glucose	5
	Starch	5
	MnSO <sub>4</sub>	0.1
	MgSO <sub>4</sub>	0.5
	Fe <sub>2</sub> (SO <sub>4</sub> ) <sub>2</sub>	0.1
	KCI	0.5
	NH₄NO₃	2
ı	CaCl	0.5

## **RESULTS**

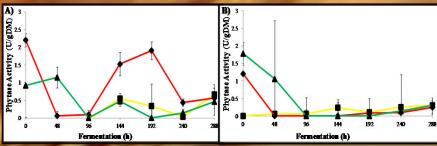


Figure 1. A. niger fermentation using the three different pretreatments: 1 = ; wheat bran without wash and autoclaved, 2 👆; wheat bran washed and autoclaved, and 3 📥 ; wheat bran without wash and UV exposed for 2h; and the two impregnation media: A) medium A and B) medium B.

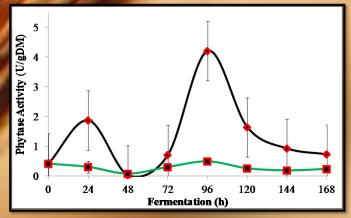


Figure 2. Phytase production in column reactors by A.niger - and A. terreus - using wheat bran as substrate-support and impregnation media A.

### Conditions of SSF:

UV exposed for 2 h



Autoclaved, 15 min at 121°C

15 mL Tubes 12 days of fermentation

7 Days of fermentation

### **CONCLUTIONS:**

The use of 15 mL tubes fermentation system; show to be an easy and practical way to assay different fermentation conditions at the same time, obtaining rapid and reliable results. UV sterilization is an excellent process for using wheat bran as substrate for phytase production. Moreover the column reactor fermentation increased significantly the phytase productivity. The fermentation conditions obtained in this research can be used to find the best phytase producing strain for future experiments.

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