



**TITLE:** SELECTION OF HEAT-RESISTANT MICROORGANISMS PRODUCERS OF EXTRACELLULAR HYDROLYTIC ENZYMES FOR ENRICHMENT OF ORGANOMINERAL FERTILIZERS

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**ABSTRACT:**

Organomineral fertilizer market has expanded in Brazil in recent years, these are usually composed of minerals associated with biologically processed waste such as peat, bone meal, cattle manure and swine. Especially, attention is currently focused on the enrichment of organomineral fertilizers with microorganisms capable to produce extracellular enzymes to hydrolyze the complex insoluble organic materials and convert agricultural wastes into value-added fertilizer products. During the granulation process, high temperatures are used, that limit the survival of the microorganisms and causes enzymatic denaturation. The objective of this work was to isolate microorganisms from the composting process and to select heat-resistant isolates that produce hydrolytic enzymes. The samples were collected in Capim Branco city, MG in composting beds with different formulations and process steps. Isolation of the microorganisms was performed by diluting the samples in saline (10-1) followed by heat shock at 80°C for 30 minutes under 120 rpm stirring. After incubation, serial dilution of the samples was performed and 0.1 ml aliquots of each dilution were transferred to Petri plates containing solid medium by surface sowing technique and subsequently incubated at 46 ± 2°C for 48 hours. The different morphotypes observed on the plates were isolated and submitted to tests of resistance at temperatures of 60 and 100°C for 2 and 4 hours in liquid medium. Forty-two heat-resistant microorganisms were selected for evaluation of the enzymatic activity. The media amido agar was used to identify the production of amylase, sodium phytate for the production of phytase and carboxymethylcellulose

medium for the production of cellulase. The enzymatic activity was determined by means of the enzymatic index (EI) obtained from the relationship between the mean diameter of the halo hydrolysis (mm) and the mean diameter of the colony (mm), being considered as a producer of hydrolytic enzyme the microorganism with  $IE \geq 2.0$ . The values obtained for IE were submitted to analysis of variance with three replicates, and the means were compared by the Skott-Knott test at 5% probability ( $p < 0.05$ ). As a result, 17 amylase producing microorganisms, 2 phytase producers and 7 cellulase producers were obtained. These microorganisms have potential for their application in high-temperature biotechnological processes for the production of granulated organomineral fertilizers.

**Keywords:** Biotechnological potential, Composting, Thermophiles.

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