TITLE: PRODUCTION OF CHITINASE BY THE FUNGUS *ASPERGILLUS NIVEUS* IN SOLID-STATE FERMENTATION USING FISHING INDUSTRY WASTE

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ABSTRACT:
Chitin is the second most abundant polysaccharide on the planet, composing the arthropod exoskeletons and fungal cell walls. Despite of the great availability, its effective utilization for biotechnological purposes is reduced. Chitinase hydrolyze chitin in N-acetylglucosamine and have been used in the treatment of industrial waste containing chitin, as found in fishing industries, to obtain compounds with higher added value. According to this, the aim was to find the best conditions for the chitinase production by *Aspergillus niveus* under Solid-State Fermentation (SSF) using shells and heads of shrimp. The SSF was performed using 2.5 g of crushed shells and/or heads of shrimp as solid substrate for different periods of fermentation (1-10 days) at 30°C and 60% relative humidity. The substrate was humidified with different moistening agents (distilled water, tap water, SR salts, Khanna salts, Minimal medium salts and trace elements) in different proportions (1:0.5 to 1:3 w/v). After fermentation, 50 mL of cold distilled water was added and shaken at 150 rpm for 20 min at 4°C for extraction of the extracellular enzymes. The dialyzed filtrate was used as chitinase source and the enzyme activity was determined using 1 mmol L^{-1} of 4-nitrophenyl-β-D-N,N',N"-triacetylchitotriose in 100 mmol L^{-1} sodium acetate (pH 4.0-5.5) and citric acid (pH 3.0-4.0; pH 6.0-6.5) buffers at different pH and temperatures (30-80°C). The best solid substrate for the production of chitinase was the mixture of crushed shell and head shrimp for 144 hours (3.6 U/g of substrate). The deproteinization process of the shells and heads of shrimp drastically reduced chitinase production (1.9 U/g of substrate), demonstrating that the protein is important for fungal growth as a source of carbon and nitrogen. The best moistening agent was tap water in the ratio of 1:2 (w/v) (4.3 U/g of substrate). Chitinase activity present in the crude filtrate was better quantified at 65°C and pH 5.0 with 100 mmol L^{-1} sodium acetate buffer. The 45 kDa protein band observed in 10% SDS-PAGE was similar to the purified *A. niveus* chitinase reported in the literature. In conclusion, the use of fishing industry waste in SSF allowed the production of chitinase, an enzyme with biotechnological potential, by *A. niveus*, indicating an alternative way to minimize the environmental impact of this type of waste.

KEYWORDS: chitinase, enzyme, shrimp shell, filamentous fungi, chitin waste

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