## **TUCUMAN BIOLOGY SOCIETY**

(Asociación de Biología de Tucumán)

### Abstracts from the

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#### 157.

# DETERMINATION OF $LD_{50}$ AND $LT_{50}$ OF Bacillus NATIVE STRAINS TOXIC TO Spodoptera frugiperda (LEP.: NOCTUIDAE)

<u>Alvarez A, Pera LM, Virla E, Baigorí MD.</u> PROIMI. Belgrano y Caseros, 4000 Tucumán. Tel: 4344888. E-mail: alvarez analia@hotmail.com

Introduction: Spodoptera frugiperda (Sf) is one of the most important corn pests in tropical and subtropical America. Bacillus thuringiensis (Bt) is an entomopathogenic spore-forming bacterium that produces parasporal crystalline proteinaceous inclusions (Cry). **Objective:** The aim of this work was to determine the lethal dose (LD<sub>50</sub>) and the lethal time (LT<sub>50</sub>) of native crystalliferous Bacillus strains against first instar larvae of Sf. Materials and methods: Insects come from a laboratory colony. The artificial diet was immersed in serial dilutions of Cry protein suspension. LD<sub>50</sub> and LT<sub>50</sub> were calculated by Probit analysis. Results and conclusions: The  $LD_{so}$  (CFU) calculated for B. sp RT, B. sp LSM, B. sp LQ and Bt kurstaki (HD1) were 8.99 X 10<sup>6</sup>, 5.60 x 10<sup>7</sup>, 2.70 X 10<sup>7</sup> and 1.04 X 10<sup>7</sup>, respectively. Under our assays conditions, B. sp RT shows the lower LT<sub>50</sub> for doses of about 6 X 10<sup>7</sup>cfu. These results show that Bacillus sp RT is an interesting strain for further development for Sf control programmes.

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### POLYMORPHISM ANALYSIS OF ESTERASES PATTERNS FROM Bacillus STRAINS

Loto F, Baigorí M, Pera L.

PROIMI-CONICET Belgrano y Caseros, 4000 Tucumán. Tel: 4344888. E-mail: lymb@arnet.com.ar

**Introduction:** Electrophoretic analysis of esterases (EC 3.1.1.1) patterns is one of the easiest ways to evaluate enzyme variants, providing valuable information in the study of species differentiation. Objective: The aim of this work was to analyze the electrophoretic profiles of esterases from Bacillus strains. Materials and methods: Microorganisms isolated from different sources were grown on LB agar during 48h. Extracellular esterases were extracted from the medium and chromatographied under nondenaturing conditions. Activities were detected using α-naphtyl acetate as substrate. Esterases were classified according to the band migration rate. They were recorded based on presence/absence in the nPAGE. The degree of similarity was estimated by simple matching coefficients. Clusters were then constructed using the UPGMA method. Results and conclusions: Fifteen different bands from fifty isolations were detected. A numerical classification reveals three main clusters with eighteen different enzymatic profiles. There was no correlation between the enzymatic profile and the source of strain isolation

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### 158.

### INFLUENCE OF CULTURE CONDITIONS ON A MYCE-LIUM-BOUND LIPASE PRODUCTION FROM Aspergillus niger MYA 135

Colín V, Baigorí M, Pera L.

PROIMI-CONICET Belgrano y Caseros, 4000 Tucumán. Tel: 4344888. E-mail: lymb@arnet.com.ar

**Introduction:** Lipases (EC 3.1.1.3) are enzymes that hydrolyse the ester bonds of water insoluble substrates at the interface between the substrate and the water. This reaction is reversible. Lipases may also catalyze ester synthesis and transesterification in reaction mixtures with low water contents. Objective: The aim of this work was to study the influence of culture conditions on a mycelium-bound lipase production from Aspergillus niger MYA 135. Materials and methods: The fermentation medium comprised (in g/l): sucrose 10; NH<sub>4</sub>NO<sub>3</sub> 2; KH<sub>2</sub>PO<sub>4</sub> 1; MgSO<sub>4</sub>.7H<sub>2</sub>O 0.2; CuSO<sub>4</sub>.5H<sub>2</sub>O 0.06. The effect of modification in the environmental condition on lipase production was tested by changing the initial pH of the medium as well as by the addition of CaCl<sub>2</sub>, FeCl<sub>3</sub> or Tween (20, 40, 60, and 80). Lipase activity was determined using 0.01 g of wet mycelium and p-nitrophenyl palmitate as substrate. Results and conclusions: The assayed culture conditions scientifically influences the lipase production. The highest specific activity (36.6 mU/g of DW) was obtained with either initial pH 8 or in presence of 0.5% Tween 60.

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### 160.

# PARTIAL CHARACTERIZATION OF ESTERASES AND LIPASES FROM A. NIGER MYA 135

Romero C, Pera L, Baigorí M.

PROIMI-CONICET Belgrano y Caseros, 4000 Tucumán. Tel: 4344888. E-mail: baigori@proimi.org.ar

Introduction: Esterases (EC 3.1.1.1) and lipases (EC 3.1.1.3) catalyze hydrolysis and synthesis of esters. Their applications include hydrolysis of fats and oils, resolution of racemic mixtures, etc. Objective: The aim of this work was to study the effects of different conditions on the stability of esterases and lipases from Aspergillus niger MYA 135. Materials and methods: Culture supernatant was preincubated at several pH values and temperatures. Residual activities were analyzed by nPAGE using either α-naphtyl acetate or α-naphtyl mirystate as substrates. Activities already separated by nPAGE were also used to evaluate their stabilities in organic solvents. Results and conclusions: Enzymes were active within the pH and the temperature range tested. Except at pH 9 and at 55°C where some bands were not detected. Enzymes preincubated in either 2-propanol, 2,3-butanodiol or acetone showing the same pattern that the control without incubation. Some bands were not detected after preincubation with propanol, n-hexane, hexanol or n-heptane. No band was observed after preincubation with butanol. One of the bands shows a good stability pattern which justifies its application in biocatalysis.

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