

# ANALYSIS OF THE GENETIC DIVERSITY AND STRUCTURE OF Helicoverpa armigera POPULATIONS FROM NORTH AND NORTHEAST OF BRAZIL



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## INTRODUCTION

The cotton bollworm, Helicoverpa armigera (Hübner), was recently introduced in Brazil. During the 2012/2013 harvest, producers reported reduced yields up to 35% on major crops. Most producers increased the number of pesticide applications in more than 15%, and the costs in cotton fields, for example, jumped from US\$ 400 to US\$ 800 per hectare.

The extensive economic losses reached US\$ 1 billion until July 2013 only in Western Bahia, triggering a phytosanitary crisis. So far, the exact distribution and extension of economic damages of this exotic pest in Brazil is still not known.

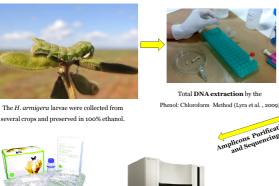
This study aimed to explore the genetic diversity and population structure of three populations of this heliothine invader that were found infesting crops in the states of Roraima, Piauí and Bahia.

## **MATERIAL and METHODS**

A total of 65 H. armigera larvae were collected from infested farms from the states of Roraima (n=14), Piauí (n=39), and Bahia (n=12). Larvae were collected directly from host plants and preserved in 100% ethanol at -20 °C.

Total DNA was extracted from the last three abdominal segments of the larvae using the phenol: chloroform method, modified by Lyra et al. (2009). Partial sequences of the COI and COII genes were amplified using the primers described by Li et al. (2011) and Liu & Beckenbach(1992), following the PCR conditions described by Li et al. (2011).

Amplicons were sequenced by the ABI3730xl DNA Analyzer. Sequences were aligned through the algorithm implemented in Clustal  $\Omega$ . Haplotype frequencies, haplotype diversity ( $\hat{H}$ ) and nucleotide diversity ( $\pi$ ) were estimated using Arlequin v.3.5 software. Genetic differences between populations were determined through pairwise F<sub>ST</sub> statistics.





Illustra<sup>TM</sup>GFX<sup>TM</sup> k for purification









ABI3730xl DNA Analyzer sequence

COI (658 bp) and COII (554 bp) Sequences

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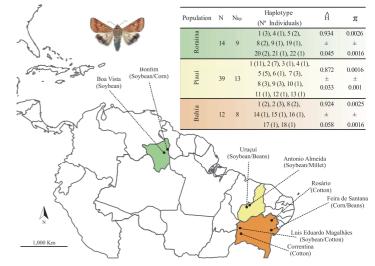


Figure 1. Genetic diversity and distribution of haplotypes from 3 Helicoverpa armigera populations.

#### **RESULTS and DISCUSSION**

The COI and COII fragments from the 65 specimens from Roraima, Piauí and Bahia were concatenated, generating a final 1212 bp sequence for each individual, and a total of 22 different haplotypes were defined (Figure 1).

Many haplotypes were shared among the three Brazilian states, suggesting a significant gene flow among these regions. In general, high haplotype diversity ( $\hat{H}$ -mean > 0.85) and low nucleotide diversity  $(\pi$ -mean < 0.003) were observed for the locations analyzed. In general, this scenario is characteristic of species that have undergone a process of recent population expansion.

AMOVA detected no genetic structure ( $F_{ST} = 7.10^{-5}$ , P = 0.44), with 99.9% of variation accounted for at the within population level and with only 0.01% variation observed among populations. Pairwise F<sub>ST</sub> values were low and not significant ( $F_{ST}$  < 0.03; P > 0.05), demonstrating that these populations are not structured (Table 1).

Table 1. Pairwise FST estimates among three investigated heliothine populations from Brazi	1
based on combined genes COI and COII.	

POPULATIONS	Piauí	Bahia	Roraima
Piauí	0.0000		
Bahia	0.0255 (P =0.15)	0.0000	
Roraima	- 0.0056 ( $P = 0.48$ )	- 0.041 ( <i>P</i> = 0.91)	0.0000

#### CONCLUSIONS

The invasive moth H. armigera is not restricted to crop fields from Northeast and Central Brazil, but has already crossed the Amazon basin.

Surveys in Venezuela and Colombia should start as soon as possible in order to monitor its entry or spread in Central America and the USA.