

Prospection and Heterologous Expression of Antimicrobial Peptide from Anticarsia gemmatalis

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INTRODUCTION: Insects represent the group of animals with a greater number of species. There is a diversity of peptides that are produced in several groups of studied insects, including antimicrobial peptides. They are small and well-structured molecules and cationic at physiological pH. Our project aims to identify and characterize new antimicrobial peptide derived from Anticarsia gemmatalis, starting from searches in cDNA libraries of this important pest. OBJECTIVES: Identify and characterize a new antimicrobial peptide from gut's cDNA libraries of A. gemmatalis. MATERIAL AND METHODS: We obtained a cDNA library from an RNA pool, after dissection of larvae gut. After large scale sequencing of the cDNA, contigs were assembled. We found two contigs with a defensin hit, comparing with a Manduca sexta's defensin. The sequence of the designed defensin was composed of contigs 4448 and 4241. The sequence was inserted into an expression vector pET26b, and this recombinant DNA was expressed in bacteria after the induction with IPTG. Then, we performed extraction of periplasmatic proteins, cell lysis and some purification techniques in order to obtain the peptide in its pure form and in the soluble fraction. **RESULTS AND DISCUSSION**: It was observed the peptide expression in *E.coli* BL21(DE3). This peptide was purified by affinity chromatography, but in small amount in the soluble fraction. Western blotting's preliminary analysis indicated the absence of the peptide in the soluble fraction, probably being expressed in inclusion bodies. CONCLUSION: It's possible to store and transform 4448/4241 peptide in bacterial cells. Besides, it is possible that this peptide was located in inclusion bodies as well as in the soluble fraction of the extract after cell lysis. The purification was the best result of this work, but it was yet preliminary. We expect to submit this sample to some other purification protocols to improve the peptide production.

Keywords: Anticarsia gemmatalis; Antimicrobial peptides; Defensin.

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