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The effect of pilin protein of *Xenorhabdus nematophila* on cellular and humoral defensive system in *Spodoptera exigua* larvae

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Xenorhabdus nematophila is a symbiotic bacterium of entomopathogenic nematode, *Steinernema carpocapsae*. It produces several toxic proteins which interfere with the immune system of insects. Here, we have shown that purified pilin protein could be involved as a virulence trait of *X. nematophila* during pathogenesis. The pilin protein purified from culture medium of *X. nematophila* and then three concentrations 5, 10, and 15 mg/ml were injected to fifth instar larvae of *Spodoptera exigua* (Hübner). Then fluctuation of total haemocyte counts (THC) and granulocyte percentage (cellular defense) as well, protease, phospholipase A_2 (PLA_2), and phenoloxidase (PO) activities were surveyed at different times. Also, the fold change expression of three main antimicrobial peptides (AMPs) including attacin, cecropin, and spodoptericin were measured. The THC population and number of granulocytes in larvae treated with different concentrations of pilin were less than the negative control. The pilin protein activated PO, PLA2 and protease enzymes. Phenoloxidase was activated in the initial hour post injection (hpi) but at 2 hpi was stable. The activities of PLA_2 and protease reached the maximum level at 12 and 4 hpi, respectively and then decreased later. The expression of attacin, cecropin, and spodoptericin in the larvae treated with pilin protein was up-regulated above that of the control. Attacin and cecropin expression reached the maximum level at 4 hpi and then decreased. While, the spodoptericin expression had an irregular trend. The cecropin was expressed more than other AMPs. The expression of attacin and cecropin, after injection of 10 mg/ml toxin protein were 66 ± 1, and 403±1 fold more than control, respectively. The findings of this research provide the first insight into the role of pilin protein when the bacterial symbiont of *Steinernema carpocapsae* (Weiser) encounters the humoral defense of an insect.

Keywords: Antimicrobial peptides, cellular defense, insect pathology, phenoloxidae, phospholipase A_2, protease