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Insect Transferrin Functions as an Antioxidant Protein

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Transferrin performs multifunctional roles in insects as an iron transporter, an antibiotic agent, a vitellogenin, and a juvenile hormone-regulated protein. Here, we show a novel functional role for insect transferrin as an antioxidant protein. Stresses, such as heat shock, fungal challenge, and H_2O_2 exposure, cause upregulation of white-spotted flower chafer *Protaetia brevitarsis* transferrin (*PbTf*) mRNA in the fat body, and they cause increased PbTf protein levels in the hemolymph. RNA interference (RNAi)-mediated *PbTf* reduction causes increased iron and H_2O_2 levels in the hemolymph and results in induction of apoptotic cell death in the fat body during exposure to stress. The observed effect of *PbTf* RNAi indicates that PbTf inhibits stress-induced apoptosis by diminishing the Fenton reaction via the binding of iron, supporting an antioxidant role for PbTf in stress responses.

Key words: Insect, transferrin iron, oxidative stress, stress response, apoptotic cell death, RNA interference, antioxidant protein

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Pr-lynx1, a Modulator of Nicotinic Acetylcholine Receptors in the Insect

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Insect nicotinic acetylcholine receptors (nAChRs) are targets for insecticides. Despite the importance of nAChR as a major target for insecticide action, modulators of nAChR in insects as yet remain unidentified. Here we show the cloning and identification of a nAChR modulator gene in an insect. This gene was isolated by searching the firefly *Pyrocoelia rufa* cDNA library, and the gene itself encodes for a protein of 120 amino acids in length, named *Pr-lynx1*. *Pr-lynx1* shares all the features, including a cysteine-rich consensus motif and common gene structure, of the *Ly-6/neurotoxin* superfamily. The recombinant Pr-lynx1, which is expressed as a 12-kDa polypeptide in baculovirus-infected insect Sf9 cells, is normally present at the cell surface as a GPI-anchored protein. Northern and Western blot analyses revealed that Pr-lynx1 is expressed in various tissues, such as the ganglion, brain, mandibular muscle, proventriculus, leg muscle, and epidermis. This expression pattern is similar to the distribution of nAChRs as assayed by α3 nAChR immunoreactivity. Co-expression of Pr-lynx1 to *Xenopus* oocytes expressing α3β4 nAChRs results in an increase in acetylcholine-evoked macroscopic currents, indicating a functional role of Pr-lynx1 as a protein modulator for nAChRs. This study on *Pr-lynx1* is the first report of a modulator for nAChRs in insect species.

Key words: *Pyrocoelia rufa*, *Ly-6/*neurotoxin superfamily, acetylcholine, nicotinic acetylcholine receptor, GPI-anchored protein, insect