

## A Functional Role of the *Apriona germari* Beetle Transferrin

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In a search to identify gene involved in the beetle self-defense responses, we cloned a novel transferrin gene, AgTf, from the mulberry longicorn beetle, *Apriona germari*, which is firstly characterized in coleopteran insect. The AgTf gene spans 8244 bp and consists of six introns and seven exons coding for 722 amino acid residues. Like known cockroach and termite transferrins, AgTf appears to have residues comprising iron-binding sites in both N- and C-terminal lobes. The AgTf expression was detected in both fat body and epidermis, although the AgTf expression in the epidermis was relatively less than in the fat body, and no expression was found in the midgut. To elucidate the functional role of transferrin in the *A. germari* beetle self-defense responses, AgTf up-regulation was analyzed under the conditions that promote an increase of the levels of reactive oxygen species (ROS). Expression of AgTf in both fat body and epidermis of *A. germari* larva was up-regulated by iron overload, microorganism challenge, and H<sub>2</sub>O<sub>2</sub> or paraquat exposure with intrahemocelic injection. The expression level of AgTf in both fat body and epidermis was up-regulated when *A. germari* larva was exposed at low (4°C) and high (37°C) temperatures, suggesting that the AgTf seems to play a self-defense role against temperature shock. To understand further functional roles of the AgTf in *A. germari*, we have elucidated the effects of reduced endogenous AgTf mRNA levels in *A. germari* larvae via RNA interference (RNAi). The RNAi-mediated AgTf reduction resulted in the increase of the DNA fragmentation and cell death of *A. germari* larvae to heat stress. These results suggest that the AgTf plays an important role in protecting apoptotic cell death against oxidative damage caused by heat stress.