

Cell Signalling Mediated by Oxidants in *Rhodnius prolixus* midgut

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Hematophagy arose independently in many insect orders, and different groups of blood-feeding insects acquired during the course of evolution specific ways to manage and digest blood. Hematophagous insects can ingest in a single meal a huge amount of vertebrate blood, which contains about 10mM of heme bound to hemoglobin polypeptide chains. Heme is a potentially harmful molecule, exerting its toxicity either by promoting the formation of reactive oxygen species or, alternatively, by insertion of free heme into phospholipid membranes eventually leading to cellular lysis. The metabolic adaptations required for a blood diet should maintain cellular homeostasis in a pro-oxidant environment created by an increased heme concentration (Graça-Souza, *et al.* 2006). Recent works are supporting that reactive oxygen species (ROS) could act as signalling molecules in many physiological events. One of these, suggest that MAPK (mitogen activated protein kinase) cascade is regulated by mitochondrial ROS (Zmijewski, *et al.*, 2005). The ERK family (>8 isoforms) belongs to the MAPK family and are typically activated by mitogens and JNKs (c-Jun NH<sub>2</sub> terminal kinase) and p38 MAPKs, both activated by cellular stress. When activated, JNKs and ERKs, phosphorylates Jun and Fos proteins, that are members of AP-1 family. The selective dimerization of AP-1 family members (Jun/Jun or Fos/Jun) regulates the gene expression in cell injury, repair, proliferation and differentiation. (Ramos-Nino, *et al.*, 2002). *Rhodnius prolixus* fifth instar males were fed with tyrode buffer and tyrode buffer added with 5, 50 and 500uM of heme. Two hours after the artificial feeding the anterior midgut was dissected (crop), homogenized and then analyzed by western blotting with different antibodies specific against the MAPK family and against phosphorylated proteins. This work shows for the first time that heme from the diet induces phosphorylation in serine and tyrosine residues of *Rhodnius prolixus* anterior midgut. Additionally, we found that the ERK 2 isoform is present independently of the heme concentration added in the diet, and that the isoform ERK ½ is gradually phosphorylated following the additions of heme in the diet.

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