

Function and Evolution of Salivary Components in *Rhodnius prolixus*

John F. Andersen

Laboratory of Malaria and Vector Research
National Institutes of Health/NIAID, Bethesda, MD 20892 USA

The salivas of blood-feeding arthropods contain complex mixtures of proteins which are essential for feeding. In our work we strive to detail the specific functions of individual proteins, to establish relationships between these proteins, and to understand mechanisms of action. Many of these proteins show good potential as therapeutic agents, and some are currently being evaluated in clinical trials. The triatomine insect *Rhodnius prolixus* is a vector of Chagas' disease and contains a rich salivary mixture of novel biologically active proteins. Many of the proteins found in the mixture belong to the lipocalin family. This protein group is notable for its ability to tolerate a large number of amino acid substitutions without disruption of the protein fold. Analysis of the salivary transcriptome of *R. prolixus* suggests that lipocalins have diversified through gene duplication events leading to over 20 expressed forms in the saliva. The lipocalin fold consists of an eight-stranded beta barrel structure with peripheral helical and coil regions. The cavity formed in the center of the barrel creates a binding pocket for a variety of ligands. Three general functional types of *R. prolixus* lipocalin have been identified. The nitrophorins bind a heme molecule in the central cavity and serve as nitric oxide transporters and histamine-binding proteins. RPA11 binds ADP and other nucleotides in the central cavity and serves as a platelet aggregation inhibitor. A third functional type, amine-binding protein, binds a variety of biogenic amines including serotonin and epinephrine and serves to limit platelet aggregation and regulate vascular tone. In addition to the binding of small-molecule ligands, some lipocalins have taken on functions involving protein-protein interactions. One of the nitrophorins binds to coagulation factor IX (and IXa) via its outer surface and inhibits the intrinsic factor Xase complex thereby preventing blood clotting. Here, I will describe the mechanistic details, evolutionary relationships and interactive effects between salivary components that allow this insect to efficiently feed on blood.