

Characterization of Heme Aggregates in the Midgut of *Aedes aegypti*

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Digestion of hemoglobin in blood-feeding organisms results in the release of large amounts of free heme in their midgut lumen. The matrix peritrophic from *Aedes aegypti* can bind heme molecules generated during blood digestion, however the identity of the binding sites and the mechanisms by which heme aggregates is formed is unknown. The aim of the study was to characterize and identify heme aggregates in the midgut of *Aedes aegypti*. The heme aggregation process during the course of blood digestion was monitored by means of midgut fractionation in sucrose gradient, the fractions were analyzed by SDS-PAGE and light absorption spectrometry. Twelve hours after blood meal (ABM), heme-bound to hemoglobin was found at the top of the gradient. During the course of digestion, the amount of heme-bound to hemoglobin decreased and the band containing heme associated to peritrophic matrix aggregates increased at higher sucrose concentrations. SDS-PAGE analysis of peritrophic matrix-containing fractions showed the presence of several proteins, such as *Aedes aegypti* intestinal mucin 1, which has been shown to bind heme. Possible heme aggregates were identified by Field Emission Scanning Electronic Microscopy in midgut 24h ABM. Heme aggregates predominantly in beads-like structures with variable size ranging from 50 to 100 nm. These results suggest that heme aggregation is an integral part of blood digestion, probably mediated by peritrophic matrix proteins.

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