The Enemy Within: Impairment of Heme Crystallization By a Quinoline Drug Drives Biochemical, Cellular and Physiological Shifts on an Insect Vector

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Introduction: Hematophagous organisms digest hemoglobin and release large amounts of heme in their digestive tracts. When free, heme is toxic and promotes redox imbalance and membrane destabilization. Thus, blood-feeding organisms require protective mechanisms to deal with excessive heme. Hemozoin (Hz) is a heme crystal produced by Plasmodium sp, Schistosoma sp and triatomine insects as the main heme detoxification mechanism. Previous evidence demonstrated that quinoline drugs are antimalarial and antischistosomal agents by inhibiting Hz formation. Here, we investigated the effect of quinidine, an antimalarial quinoline, in R. prolixus. Material and methods: Hz was extracted from posterior midgut of R. prolixus by differential solubilization. Urate and hemoglobin levels were determined by colorimetric assays. Lipid peroxidation was determined by the TBARS assay in hemolymph. Total heme levels were measured by the alkaline-pyridine method. Reactive species in posterior midgut were analyzed by fluorescence microscopy using the dihydroethidium (DHE) probe. The hemolymph levels of Rhodnius heme binding protein (RHBP) were determined spectrophotometricaly and transcript levels assessed by real time PCR in fat bodies. Posterior midguts were fixed with glutaraldeyde and submitted to transmission electron microscopy routine. Results and discussion: Quinidine did not affect blood ingestion or digestion, but severely affected Hz formation. Heme levels in the hemolymph were increased by quinidine, which promoted lipid peroxidation, reactive species generation and reductions in urate levels. As a compensatory mechanism, protein and transcript levels of RHBP increased by quinidine. Ultrastructural analyses of posterior midgut indicate general loss of cellular organization with reduced densities of mitochondria, together with the presence of myelin figures as well as numerous electron dense structures (haemoxysomes), indicating autophagic pathways. Although there were no changes on insect viability after quidinine treatment, the most striking physiological observation was a decrease in eggs laying. Conclusion: Impairment of heme crystallization within the Rhodnius midgut results in severe physiological changes involving redox imbalance and autophagy.

Key words: hemozoin, heme, Rhodnius prolixus, quinoline drugs

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