## STUDY OF LYSOSOMAL-LIKE HYDROLASES DURING <u>PERIPLANETA</u> <u>AMERICANA</u> EARLY EMBRYOGENESIS

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Egg formation at oviparous animals consists on accumulation of a large amount of nutritional reserve to the embryo in yolk granules, which resemble quiescent lysosomes. Some lysosomal enzymes like proteases and acid phosphatases have been described being responsible for yolk mobilization and are activated by acidification. Our previous results showed an acid phosphatase activity at day-6 eggs with maximum level at day-10 during embryogenesis of Periplaneta americana. This activity from day-6 was shown to be directed exclusively against phosphotyrosine while phosphoserine also proved to be suitable substrate at day-10. We also observed two classes of proteases during early embryogenesis: A papain-family cysteine protease, inhibited by E-64, insensitive to CA 074 (typically from cathepsin-L) and activated by acidic pH at day-3; and a neutral serine protease, detected by using aprotinin beginning at day-6. Concomitantly we are also investigating a putative serine protease inhibitor, identified at day-0 eggs modulating yolk proteolysis. We now demonstrated protease activity by using fluorogenic substrate being it localized within both small and large yolk granules at day-6. Our results showed a temporal relationship between protease and acid phospha tase during early embryogenesis although we could not detect interplay between them. Preliminary results showed that vitellins are not the preferential target for tyrosine phosphatase by incubation in vitro. Possible targets are now under investigation.

Supports: CAPES, FAPERJ, PRONEX

Key words: acid phosphatase; embryogenesis; Periplaneta americana; protease.