Mechanisms of signal transduction on host-Plasmodium interactions

Célia Regina S. Garcia, Fernanda Koyama, Alexandre Budu, Michael H. Nathanson Robson Sartorello and Desiree Shuck.

1Biomedical Institute – São Paulo University – São Paulo – SP – Brazil, [2]Medical School – Yale University – New Haven – CT – USA, [3]Biosciences Institute – São Paulo University – São Paulo – SP- Brazil.

Plasmodium display serpentine receptors for sensing environmental changes. We have proposed a novel mechanism that allows the intracellular protozoan *Plasmodium* to use thriptophane-related compounds to modulate its cycle in a Ca^{2+}-dependent manner. We also evaluate modulation of UPS (ubiquitin-proteassome) genes by melatonin using real time PCR and selected *Plasmodium* genes. The human malaria parasite *P. falciparum* were treated with melatonin 10µM and 100 nM at 24 hours post invasion. We have found changes in transcription of 4 UPS genes. We have also investigated signal transduction within mammalian cells expressing a *Plasmodium* receptor for activated kinase C.(PfRack). RACK stabilizes PKC in its active form as well as binds to IP_3 receptors and modulate calcium release. We have investigated the function of *Plasmodium falciparum* RACK using a codon-optimized form of the gene to express this protein in mammalian cell lines. Endogenous RACK1 expression was also knocked down. By time-lapse confocal microscopy, we observed that ATP (100µM)-induced Ca^{2+} signals were reduced by 83,3 +3,8% in HEK 293 cells expressing PfRACK and by 89,6 +7,8% in cells expressing PfRACK and treated with siRNA for mammalian RACK1. We also performed experiments using a permeable caged IP3. Upon 2-photon encaging, the IP3 failed to increase calcium in cells expressing PfRACK. Confocal immunolocalization showed partial co-localization of PfRACK with its mammalian ortholog.