Role of PI3K Pathway Signaling in Embryonic Cell Lineage BME26 of *Rhipicephalus (Boophilus) microplus*.

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Introduction. Insulin signaling pathway (ISP) has been demonstrated with a high degree of evolutive conservation. Besides its role in glucose metabolism, ISP also participates in events related to embryogenesis of vertebrate and invertebrate models. Metabolic response to insulin is mediated primarily by phosphatidylinositol 3-OH kinase (PI3K), a heterodimeric enzyme with catalytic (p110) and regulatory (p85) subunits. Chemical inhibitors of PI3K are able to block accumulation of glycogen in response to addition of exogenous insulin in BME26 cells. Due to the involvement of PI3K pathway in cell survival, this study aims to evaluate the role of PI3K signaling on the viability and cell morphology BME26. Material and Methods: The effects of chemical inhibition or activation of PI3K, and gene silencing for p85 on cell viability BME26 were evaluated in MTT reduction assay. Results and Discussion: Cells were exposed for 24 hours to irreversible inhibitor wortmannin, concentrations 5-250 nM, showed viability greater than 70% (IC<sub>50</sub> value of 1,239 nM). Same treatment with reversible inhibitor, LY294002, between the concentrations of 1-100 µM, retained viability values above 50% (IC<sub>50</sub> 89.15 µM). Additionally, AKT inhibition (PI3K pathway downstream effector) reduced cell viability in a concentration range of between 24-72 µM (IC<sub>50</sub> 48µM). Cell viability was not altered when BME26 cells were treated with a PI3K activator, 740Y-P (from 0.05 to 50µg/ml), in the absence of fetal calf serum. A cDNA fragment related to p85 in BME26 cells was amplified and cloned. Sequence confirmation contributed in setting the strategy for gene silencing by RNAi in cells. Silencing of p85 subunit promoted small changes in BME26 cells viability. BME26 morphological analysis and the search for sequences homologous to the catalytic subunit of PI3K, and other components of ISP in ticks are under development. Conclusão: Further studies on ISP may extend the current knowledge of this ectoparasite’s physiology.

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