

Characterization of Metabolic Disorders Associated with Dengue Virus NS1 Protein Expression in Hepatocytes

Andrade, I.S.¹; de-Souza-Ferreira, E.²; Rocha, D.C.P.¹; Pagani, T.D.¹; Costa, G.S.²; Zuma, A.A.¹; Reed, C.³; Wolkowicz, R.³; De Souza, W.¹; Almeida, F.C.L.²; Galina, A.²; Silva, E.M.¹; Mohana-Borges, R.¹

¹Instituto de Biofísica Carlos Chagas Filho, Universidade Federal do Rio de Janeiro, Brazil; ²Instituto de Bioquímica Médica Leopoldo De Meis, Universidade Federal do Rio de Janeiro, Brazil; ³Department of Cell and Molecular Biology San Diego State University, USA.

Introduction: Dengue is an important arthropod-borne viral infection of humans. This disease has wide effect on multiple organ systems, the most common being the liver. Dengue virus (DENV) is composed of 10.7kb RNA translated as a single polyprotein, which is cleaved in three structural proteins and seven nonstructural (NS) proteins. NS1 has approximately 50 kDa and exists in oligomeric forms, which are found in different cellular locations and as secreted hexameric lipoparticle. Previously, we demonstrated that NS1 is an important modulator of cellular energy metabolism by increasing the glyceraldehyde-3-phosphate dehydrogenase (GAPDH) activity. NS1 is related to dengue pathogenesis response, then it is interesting to verify how this protein is capable of modulating the hepatocytes metabolism. **Objectives:** Our aim is to evaluate the influence of NS1 in metabolism and mitochondrial function. **Material and Methods:** We used Huh7.5.1 transduced with *ns1* DENV2 16881 gene and induced with doxycycline for 48h to express NS1 protein. These cells and negative control were evaluated by oxygraphy of both intact cells and in digitonin-permeabilized, to check mitochondrial function. The activity of glycolytic enzymes were measured in vitro. Mitochondrial morphology was observed by transmission electron microscopy. The hydrophilic cell metabolites were extracted from the aqueous phase of chloroform/methanol/water and identified by ¹H-NMR. **Results and Discussion:** We observed reduced oxygen consumption of intact hepatocytes expressing NS1 but no difference when permeabilized by accessing mitochondrial respiration while providing typical mitochondrial substrates (pyruvate+malate and succinate). No significant difference was observed in hexokinase, phosphoglucose isomerase, phosphofructokinase and pyruvate kinase activity in NS1-expressing cells. Mitochondrial morphology appears more elongated in cells expressing NS1, indicating potential changes in its shape. NMR analysis revealed that metabolites such as lactate and urea were decreased in NS1-expressing cells, whereas guanine is increased. **Conclusions:** NS1 may be considered an important molecule associated with metabolic abnormalities associated with dengue.

Key words: Dengue, NS1, metabolism.