Q.05 - Effect of the Hypoxia on the Expression of Zinc Transporters in Renal Adenocarcinoma Cell Line

Luana da Silva Ferreira [1], Soraia Barbosa de Oliveira [1], Maria Helena Bellini[1]
[1]Molecular and Cellular Biology of Cancer, Nuclear and Energy Research Institute (São Paulo , Brasil)

INTRODUCTION: Renal cell carcinoma (RCC) is a tumor responsible for about 1 to 3 % of all malignancies. The most common histological variant is the clear cell carcinoma (ccRCC), representing about 45% of all cases of RCC in adults. ccRCC is associated with the VHL gene mutation. The loss of the VHL protein prevents the degradation of HIF subunits, which are involved in critical oncogenic pathways. Zinc is an essential trace element and its cellular homeostasis is regulated by zinc transporters such as ZIPs and ZNTs. The profile of their expression in renal tumor is still unknown. OBJECTIVES: Evaluate the expression profile of zinc transporters in ccRCC in normoxia and hypoxia culturing conditions. MATERIALS AND METHODS: 786-0 tumor cells were cultured in hypoxic conditions inside a hypoxia chamber with an oxygen absorber to the atmosphere of 1% O2, 5% CO2, and 94% N2, and placed in an incubator at 37°C for 24 hours. The Altair PRO Single-Gas Detector was used to measure the percentage of O2. For gene expression analysis, RTq-PCR was used and the results were analyzed by the Delta-Delta ct method. DISCUSSION AND RESULTS: VEGF and HIF2a expression in 786-0 cells were evaluated to confirm the efficacy of hypoxia chamber. There was a significant increase in the VEGF expression of 312.8±2.14% (P< 0.0001) and HIF2a of 593.4±57.21% (P< 0.0092). Besides that, the gene expression analysis revealed a downregulation in the hypoxic environment of the channels ZNT9 of 71.41±0.84% (P< 0.0001), ZIP1 of 17.45±3.68% (P< 0.0418), ZIP4 of 76.3±9.75% (P< 0.0054) and ZIP10 of 44.96±4.31% (P< 0.0001). CONCLUSION: The hypoxia modulates the expression of Zn channels in 786-0 cells indicating that such channels play a role in the pathophysiology of ccRCC.

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Q.06 - NCX Expression in Human Glioblastoma Multiforme Cell Lines and Differential Sensitivity to NCX inhibitors

Vyctória dos Santos Ramos [1], Laura Francisca Leite do Prado de Souza[2], Rayssa de Mello Lopes[2], Tiago Rodrigues[2], Ivarne Luis dos Santos Tersariol[3], Valeria Valente[4]
[1]Centro Interdisciplinar de Investigação Bioquímica, Universidade de Mogi das Cruzes (, Brazil), [2]Centro de Ciências Naturais e Humanas, Universidade Federal do ABC (, Brazil), [3]Departamento de Biologia Molecular, Universidade Federal de São Paulo (, Brazil), [4]Faculdade de Ciências Farmacêuticas, Universidade Estadual Paulista (, Brazil)

INTRODUCTION: Glioblastoma multiforme (GBM), also called astrocytoma grade IV, is a fast-growing and extremely aggressive type of brain tumor. GBM is very difficult to be treated and, besides surgical removal, the combination of the oral alkylating drug temozolomide and radiotherapy are the current available weapons. Since tumor cells often exhibit altered Ca^{2+} homeostasis to support the high proliferative rate, several calcium pumps and channels have been studied in cancer. However, only few studies have investigated the role of Na^{+}/Ca^{2+} exchanger (NCX). This exchanger has particular importance in GBM due to its high expression in cells from central nervous system OBJECTIVES: To evaluate the expression of the three NCX isoforms (NCX1, NCX2, and NCX3) in two different GBM cell lines comparatively and also to test their sensitivity to NCX inhibitors. MATERIALS AND METHODS: RNA sequencing was performed by Illumina sequencers (Genome Analyzer IIx and NextSeq 500) and NCX expression was obtained. GBM cell lines U-343MG and U-251MG ($3x10^4$ /cm²) were grown in supplemented DMEM high glucose medium. Growth curves were obtained by Neubauer chamber counting. The effects of NCX1 inhibitor KB-R7943 on cell viability were evaluated by the MTT reduction assay DISCUSSION AND RESULTS: 343MG cells exhibited a higher proliferative rate compared to U-251MG and consequently higher sensitivity to temozolomide. Conversely, U-251MG cells exhibited a higher NCX1 expression compared to U-343MG cells, which in turn, expressed more NCX3. In accordance, U-251MG cells were more sensitive to the inhibition of NCX1 by KB-R7943 than U-343MG CONCLUSION: Together, these data show differential expression of NCX in GBM cell lines, which modulates the sensitivity to NCX inhibition. Further studies are required to assess functional aspects of NCX in these GBM models.

Keywords: Glioblastoma, Calcium, NCX
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