P4.11

Expression of protein phosphatase 2 (PP2A) inhibitory subunits in breast cancer cell lines

Maria Pia Grixti¹, Christian Saliba², Shawn Baldacchino¹, Christian Scerri³, Godfrey Grech¹ Department of Pathology, Faculty of Medicine and Surgery, University of Malta, ²Centre for Molecular Medicine and Biobanking, University of Malta, ³Department of Physiology

Biobanking, University of Malta, ³Department of Physiology & Biochemistry, Faculty of Medicine and Surgery, University of Malta

Introduction: PP2A plays an integral role in the regulation of a number of major signalling pathways involved in the maintenance of normal cell division and survival. PP2A endogenous inhibitory subunits, namely SET, CIP2A and IGBP1, commonly found overexpressed in cancer, can suppress this PP2A activity resulting in cell proliferation and survival. In this study we investigated the expression of PP2A inhibitory subunits in breast cancer cell lines and overexpressed SET, CIP2A and IGBP1 in selected breast cancer cell lines.

Malta Medical Journal - Volume 27 - Supplement - December 2015

Methods: Twelve human breast cancer cell lines representing different breast tumour subtypes and a non-tumorigenic epithelial breast cell line were cultured. RT-PCR was used to quantify the transcript levels of PP2A inhibitory subunits. The SET, CIP2A and IGBP1 coding sequence were cloned in a mammalian expression vector. A transfection protocol was optimised to transfect selected cell lines. Western blot was used to quantify the protein levels of PP2A downstream effectors.

Results: Expression analysis showed that the MCF-7 and MDA-MB-453 breast cancer cell lines have the lowest endogenous levels of PP2A inhibitory subunits. Successful transfection of these cell lines with SET, CIP2A and IGBP1 constructs was confirmed by measuring the GFP expression using fluorescent microscopy and western blot analysis.

Conclusion: Overexpression of the PP2A inhibitory subunits allows investigation of differential expression, using a breast cancer cell model. Further studies include the isolation of polysome bound RNA followed by RNA sequencing, to identify potential therapeutic targets in breast cancer subtypes with high SET, CIP2A or IGBP1 expression.