OP4.36

Amiloride induces alternative splicing of the PP2Acα mRNA in haematopoietic cell lines

Stephanie Gauci[1], Christian Saliba[2], Shawn Baldacchino[1], Anthony Fenech[3], Godfrey Grech[1]

[1]Department of Pathology, Faculty of Medicine and Surgery, University of Malta, [2]Centre for Molecular Medicine and Biobanking, University of Malta, [3]Department of Clinical Pharmacology and Therapeutics, Faculty of Medicine and Surgery, University of Malta

Introduction: Targeting BCRABL1 by imatinib has proven successful for the treatment of Chronic Myeloid Leukemia (CML) patients. Nonetheless, a cumulative 5year failure rate of 17% still exists due to therapeutic resistance. Increased expression of BCRABL1 inhibits PP2A activity, promoting survival and proliferation. Previous studies established that an isoform of the catalytic subunit α of PP2A (PP2Acα) is predominant in 15% of CML. This study investigates alternate splicing of PP2Acα as a novel potential mechanism of therapeutic resistance in CML.

Methods: Several leukaemic cell lines were treated with amiloride, imatinib, rapamycin or FTY720. The differential expression of selected splicing factors was analysed by qRT-PCR. The expression profile of the resulting cellular model was correlated with the splicing factor profile of 14 CML patient samples using the unpaired ttest.

Results: A cellular model was established using TOM-1 cells (BCRABL1+ Bcell precursor leukaemia cell line) treated with amiloride to predominantly express PP2Acα2. Conversely, untreated cell lines, cell lines treated with imatinib, rapamycin and FTY720, and BCRABL1 negative CML samples did not express the PP2Acα2 isoform. 8 out of 15 (53%) splicing factors were differentially expressed in the BCRABL1+ cells, with a pvalue lower than 0.05. All 15 splicing factors analysed were upregulated in the PP2Acα2 mutant isoform CML patient samples.

Conclusion: A cellular model with predominant expression of PP2Acα2 was established. This isoform switch was solely induced by amiloride and correlated with differential splicing factor expression. These results suggest a novel mechanism for BCRABL1 targeted therapy resistance mediated by differential expression of splicing factors.

Disclosure: Funds were made available through the University of Malta.