HDAC INHIBITORS TARGET ONCOGENIC BRAF AND P53 IN MELANOMA CELLS AND PROMOTE A SWITCH FROM PRO-SURVIVAL AUTOPHAGY TO APOPTOSIS

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Histone deacetylases (HDACs) are often overexpressed in tumor cells causing a predominantly hypo-acetylated chromatin state. Histone deacetylase inhibitors (HDACI) are epigenetic compounds that induce histone acetylation and consequent expression of tumor suppressor and pro-apoptotic genes. Therefore, HDACI have been widely considered very promising antitumor agents. Here, we focus on the effects of HDACI in two melanoma cell lines, SK-Mel 28 and A375, which are characterised by BRAF V600E oncogenic mutation. BRAF is a well-known component of MAPK proliferative signalling that results hyper-stimulated in most melanomas just because of the V600E mutation. Moreover, SK-Mel 28 cells have also an additional oncogenic mutation in p53 which may further contribute in tumor progression. Our results indicate that two general HDAC inhibitors, SAHA and ITF2357, dose dependently reduce the viability of SK-Mel 28 and A375 melanoma cells. Interestingly, both inhibitors markedly decreased oncogenic BRAF protein level in the two cell lines. In SK-Mel 28, we found that oncogenic BRAF has also a nuclear localization and HDACI markedly decrease BRAF in both nuclear and cytoplasmic fractions. Moreover, we evaluated whether HDACI could affect the level of mutated p53 in SK-Mel 28 cells as well. Our results show that the protein level dramatically decreased following HDACI treatment almost disappearing at 16 h while it increased in A375 cells that express a p53 wild type form. In this cell line, a slight decrease in p53 level was observed only after prolonged treatment time (48 h). To understand whether the decrease in p53 levels observed in SK-Mel 28 was due to protein degradation, we pre-treated the cells with bortezomib, a proteasome inhibitor, or bafilomycin A1, an autophagy inhibitor. The results clearly indicated that mutated p53 is mainly degraded by the proteasome since bortezomib completely prevented the decreasing effect of HDACI on p53 level. We also found that HDACI induce a switch from pro-survival autophagy to caspase-dependent apoptosis in melanoma cells. Ongoing studies aim to elucidate the relationship qamon oncogenic BRAF, p53 and pro-survival autophagy in melanoma.