Nicotinamide activates the p53/miR-34a/SIRT1 tumor suppressor network leading to apoptosis of chronic lymphocytic leukemia cells

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Nicotinamide, the amide form of the B-complex vitamin niacin, is the main precursor of NAD^+ in most mammalian cells. Tumor transformation is generally accompanied by profound alterations in total cellular NAD^+ levels, nicotinamide concentrations and consequently in the activities of NAD^+-dependent enzymes. Among these enzymes NAD^+-dependent, sirtuins are class III histone deacetylases. SIRT1, the best characterized member, plays important roles in gene silencing, aging and longevity deacetylating histone and non-histone proteins, including p53. Several pieces of evidence indicate that SIRT1 may work as an oncogene by suppressing p53 functions, leading to genome instability and resistance to apoptosis. For these reasons, there is intense investigation in designing molecular tools that inhibit sirtuins.

Because of a longstanding experience in the use of nicotinamide for the treatment of pellagra and because of the lack of reported side effects, nicotinamide could be in the pipeline of SIRT1 inhibitors to be tested in clinical settings. Our hypothesis is that nicotinamide affects human B cell

homeostasis and that the effects might be more marked in a neoplastic context. Chronic lymphocytic leukemia (CLL), selected as disease model, is characterized by the slowly progressive expansion of a population of mature monoclonal B lymphocytes, intrinsically resistant to apoptosis, limiting the therapeutic efficacy of many drugs.

The results of this study confirm that CLL cells are characterized by increased expression (mRNA and protein) and function of SIRT1, compared to normal B lymphocytes from peripheral blood of age- and sex-matched donors. Nicotinamide treatment directly leads to significant enzymatic activity inhibition of SIRT1 evident only in leukemic cells. Functional block of this enzyme is followed by re-activation of the p53 pathway marked by increased expression of p53-dependent genes, such as p21, NOXA and BAX and decreased of Mcl-1. The endpoint is a p53-dependent block of the cell cycle and induction of apoptosis in CLL cells with a wild type p53. Conversely CLL cells with a mutated/deleted p53 protein are partially resistant to nicotinamide-mediated effects.

Our hypothesis is that the link between SIRT1 and p53 is represented by miR-34a, a direct transcriptional target of p53 that can bind SIRT1 mRNA and trigger its degradation. In line with this hypothesis, a combined treatment of CLL cells with nicotinamide and etoposide (a DNA-damaging chemotherapeutic) was followed by i) up-regulation of miR-34a expression, ii) marked down-modulation of SIRT1 expression and function, and iii) induction of expression and acetylation of p53. This positive feedback loop was operative in the CLL model, with the final outcome of a tumor cell apoptosis significantly enhanced.

In conclusion this work suggests that nicotinamide represents a potentially useful adjunct in the treatment of selected forms of CLL, also considering that normal B lymphocytes are markedly less sensitive to nicotinamide effects.