Cagnetta A, Caffa I, Acharya C, Soncini D, Acharya P, Adamia S, Pierri I, Bergamaschi M, Garuti A, Fraternali G, Mastracci L, Provenzani A, Zucal C, Damonte G, Salis A, Montecucco F, Patrone F, Ballestrero A, Bruzzone S, Gobbi M, Nencioni A, Cea M. (2015). APO866 increases anti-tumor activity of cyclosporin-A by inducing mitochondrial and endoplasmic reticulum stress in leukemia cells. *Clin Cancer Res.* 21(17):3934-45.

**PURPOSE:**

The nicotinamide phosphoribosyltransferase (NAMPT) inhibitor, APO866, has been previously shown to have antileukemic activity in preclinical models, but its cytotoxicity in primary leukemia cells is frequently limited. The success of current antileukemic treatments is reduced by the occurrence of multidrug resistance, which, in turn, is mediated by membrane transport proteins, such as P-glycoprotein-1 (Pgp). Here, we evaluated the antileukemic effects of APO866 in combination with Pgp inhibitors and studied the mechanisms underlying the interaction between these two types of agents.

**EXPERIMENTAL DESIGN:**

The effects of APO866 with or without Pgp inhibitors were tested on the viability of leukemia cell lines, primary leukemia cells (AML, n = 6; B-CLL, n = 19), and healthy leukocytes. Intracellular nicotinamide adenine dinucleotide (NAD(+)) and ATP levels, mitochondrial transmembrane potential ($\Delta\Psi(m)$), markers of apoptosis and of endoplasmic reticulum (ER) stress were evaluated.

**RESULTS:**

The combination of APO866 with Pgp inhibitors resulted in a synergistic cytotoxic effect in leukemia cells, while sparing normal CD34(+) progenitor cells and peripheral blood mononuclear cells. Combining Pgp inhibitors with APO866 led to increased intracellular APO866 levels, compounded NAD(+) and ATP shortage, and induced $\Delta\Psi(m)$ dissipation. Notably, APO866, Pgp inhibitors and, to a much higher extent, their combination induced ER stress and ER stress inhibition strongly reduced the activity of these treatments.

**CONCLUSIONS:**

APO866 and Pgp inhibitors show a strong synergistic cooperation in leukemia cells, including acute myelogenous leukemia (AML) and B-cell chronic lymphocytic leukemia (B-CLL) samples. Further evaluations of the combination of these agents in clinical setting should be considered.