STK405759 as a Novel Tubulin Active Agent for Multiple Myeloma Therapy

Merav Leiba[1], Gabriela Rozic[1], Lena Paukov[1], Jana Jakubikova[2], Adrian Duek[1], Abraham Avigdor[1], Arnon Nagler[1]

[1]*Division of Hematology & Bone Marrow Transplantation, Sheba Medical Center, Tel Hashomer, Israel*

[2]*Department of Medical Oncology, Jerome Lipper Multiple Myeloma Center, Harvard Medical School, Dana-Farber Cancer Institute, USA*

Background: Despite advances in treatment, multiple myeloma (MM) remains incurable due to development of drug resistance in the bone marrow microenvironment. Microtubules (MTs) are dynamic protein biopolymers formed through polymerization of heterodimers of α- and β-tubulins. Disruption of microtubules induces cell cycle arrest in G2-M phase and formation of abnormal mitotic spindles. MTs are also involved in many processes in interphase cells, including intracellular trafficking, cell motility and angiogenesis. The important functions of MT in the cells make them an attractive target for anti-myeloma drug discovery.

Furan metotica is a novel class of anti-mitotic spindle drugs that inhibit kinetochore-microtubule binding and trigger a spindle checkpoint mediated arrest in mitosis, which frequently ends in cell death. We evaluated the activity of STK405759, a member of the furan metotica family, as a novel, potential antitubulin drug for MM treatment in preclinical models.

Methods: Cytotoxic activity of STK405759 was evaluated by XTT assay. Apoptosis and cell cycle were checked by flow cytometry. Tubulin polymerization inhibition was evaluated using a biochemical cell free assay and by evaluating the levels of soluble and polymerized tubulin in MM-treated cells using Western blot analysis. Efficacy and toxicity of the drug were evaluated in a murine MM xenograft model. Histochemistry was used to assay tumor apoptosis.

Results: STK405759 had a potent cytotoxic activity against a wide variety of MM cell lines and patient-derived MM cells, regardless of their sensitivity to conventional therapy or novel agents. In contrast, the viability of normal peripheral blood mononuclear cells derived from healthy donors and MM patients was not inhibited. Importantly, STK405759 induced cell death of RPMI MM cells co-cultured with HS-5 bone marrow stromal cells.

STK405759 inhibited tubulin polymerization in a cell free system and resulted in a decrease in polymerized tubulin in MM treated cells. The STK405759 anti-tubulin activity was supported by demonstration of MM cell cycle arrest followed by activation of an apoptotic default pathway. Activation of pro-caspase-8 and poly (ADP-ribose) polymerase in the cleaved forms, as well as down-regulation of the Mcl-1 anti-apoptotic protein was detected in RPMI treated cells.

Combination studies of STK405759 with bortezomib, lenalidomide or dexamethasone showed significant synergistic and additive cytotoxicity in MM cells. *In vivo* studies revealed decreased MM tumor burden and prolonged survival of STK405759-treated mice compared to controls. STK405759 induced apoptosis of tumors cells from treated mice.

Summary/Conclusion: STK405759 is an active, microtubule-targeting agent with potent anti-myeloma activity. These results provide a rationale for further evaluation of STK405759 as monotherapy or part of combination therapy for treating patients with MM.