Identification of a Novel Molecule for the Treatment of Muscle-Invasive Bladder Cancer

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BACKGROUND

- Bladder cancer (BCa) is one the primary causes of cancer death in U.S. and worldwide
- Cisplatin based therapies are the mainstay of BCa treatment however, the overall survival rate remains dismal in the case of muscle-invasive bladder cancer (MIBC), which constitutes about 30% of all BCa cases
- The goal of this study is to determine the efficacy of two plant derived compound analogues, ASR488 and ASR490, in terms of growth inhibition in MIBC cell lines, TCCSUP and J82

HYPOTHESIS

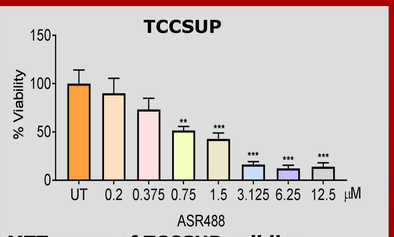
- Several natural compounds and their analogues have demonstrated anti-cancerous attributes by targeting oncogenic signaling pathways, such as Withaferin A
- ASR488, a Withaferin analogue, demonstrated growth inhibition in cancer cells
- We anticipated that ASR488 inhibits cancer cell growth by inducing apoptosis and attempted to delineate the responsible mechanism

CLINICAL IMPACT

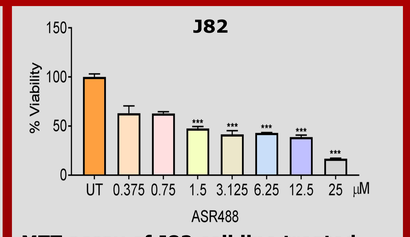
- These results suggest that ASR488 might be a potential therapeutic agent for treatment of MIBC, however additional studies are needed to confirm the mechanistic aspects of growth inhibition and efficacy of the compound as an anti-metastatic agent

RESULTS

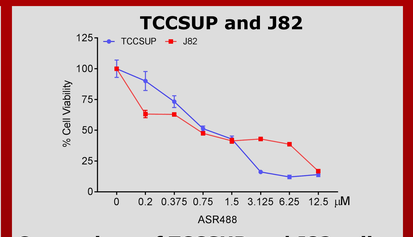
- ASR488 and ASR490 were screened for growth inhibitory potential in BCa cell lines
- ASR488 demonstrated significant growth inhibition in TCCSUP (IC_{50}: 750nm) and J82 (IC_{50}: 800nm) and was chosen for further experimentation
- Significant induction of apoptosis was seen in TCCSUP, which was further confirmed by increased expression of BAX, Cleaved PARP, and Cleaved Caspase-3
- Immunoblotting assays demonstrated an apparent decline in Chk1 expression which indicates its possible phosphorylation and role of ASR488 in inducing DNA damage
- We were able to confirm increased expression of p-Chk1 in ASR488 treated TCCSUP cells



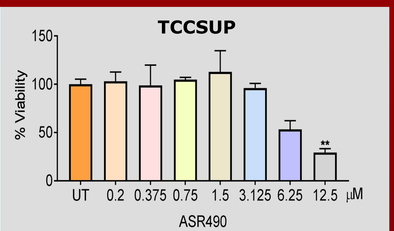
MTT assay of TCCSUP cell line treated with ASR488 (IC_{50} – 750nm)



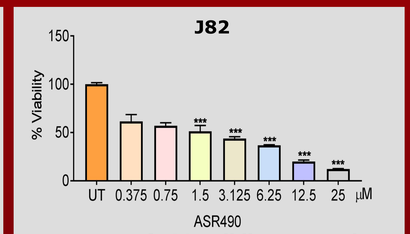
MTT assay of J82 cell line treated with ASR488 (IC_{50} – 800nm)



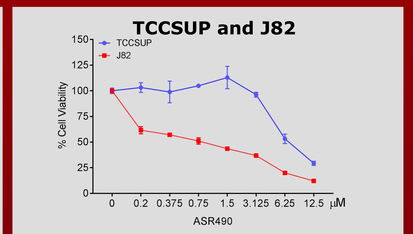
Comparison of TCCSUP and J82 cell lines treated with ASR488



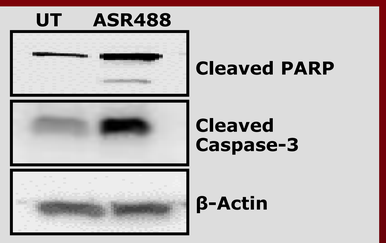
MTT assay of TCCSUP cell line treated with ASR490



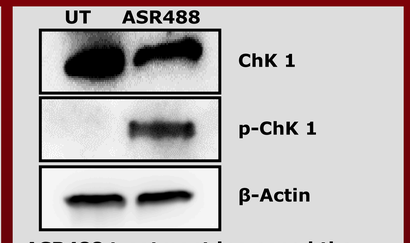
MTT assay of J82 cell line treated with ASR490



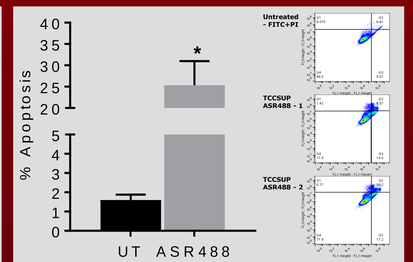
Comparison of TCCSUP and J82 cell lines treated with ASR490



ASR488 treatment increased the expression of pro-apoptotic markers in TCCSUP



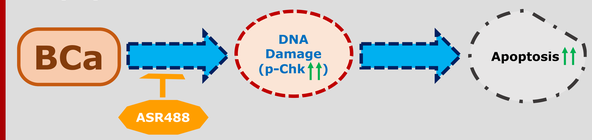
ASR488 treatment increased the expression of DNA damage markers in TCCSUP



ASR488 induced apoptosis in TCCSUP (23.7%; p = 0.0042)

CONCLUSION

- ASR488 treatment significantly inhibits cell growth of Muscle Invasive Bladder Cancer (MIBC) cells
- Initiation of DNA damage signaling was observed in bladder cancer cells following the treatment with ASR488 resulting in apoptotic induction



FUTURE DIRECTIONS

- Use another cell line, UROtsa, in order to measure the toxicity of ASR488 in normal bladder cancer cells and compare it to TCCSUP and J82, both of which are MIBC cell lines
- Screen for DNA damage response markers to confirm the role of DNA damage signaling in cell growth inhibition by ASR488
- Screen combination of existing chemo-therapeutic compounds with ASR488/ASR490 to develop a potent combinatorial treatment strategy for muscle-invasive bladder cancer

ACKNOWLEDGEMENTS

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