Therapeutic targeting autophagy to sensitize cancer immunotherapy in prostate cancer

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Although cancer immunotherapy has revolutionized cancer treatment, patient response to immunotherapy remains varied. Despite progress, the mechanisms limiting cancer immunotherapy are not yet fully understood. A low number of tumor infiltrating T cells (cold tumor) is one of the limiting factors for cancer immunotherapy. Agents that enhance immunotherapy by shifting cold tumors to hot tumors will greatly benefit cancer immunotherapy. In prostate cancer, the majority of tumors are known to be cold and hence, cancer immunotherapy is not the ideal treatment option. Here, we use a prostate cancer model as an example to demonstrate that modulating the tumor microenvironment through altering autophagy will change the tumor cytokine secretion profile, which in turn attracts immune lymphocytes into the tumor microenvironment. In tandem, we have identified a candidate compound known as ESK981 for such a purpose. ESK981 is a potent anti-cancer multi-tyrosine kinase inhibitor with novel autophagy-inducing property. As a Phase-I cleared small molecule, ESK981 has been proven to be useful in multiple types of cancer by enhancing immunotherapy.

Method
A small molecule library was used for screening autophagy activity and cytokine secretion. Various types of human cancer cell lines (prostate, renal, bladder, breast etc.) and multiple syngeneic mouse lines were examined for autophagy activity as well as an *in vitro* response to interferon stimulation with or without ESK981. A syngeneic mouse prostate cancer was used for the *in vivo* examination of autophagy as well as the anti-tumor effect by ESK981 monotherapy and/or in combination with anti-PD-1 therapy.

Conclusion
We have discovered a robust, novel autophagy-modulating small molecule, named ESK981, for the treatment of various cancer types as a monotherapy. In addition, we have demonstrated that autophagy has an essential role in the anti-tumor effect of immunotherapy, especially for anti-PD-1 in syngeneic

prostate cancer model. Therefore, the use of a small molecule such as ESK981 to target autophagy can enhance immunological infiltration induced by cancer immunotherapy, such as immune checkpoint blockade, for non-immunogenic tumors.

Conflict of Interest Statement

A.M.C. is a co-founder and serves on the Scientific Advisory Board of Esanik Therapeutics, Inc. which owns to the rights to the clinical development of ESK981. Esanik Therapeutics, Inc. did not fund or approve the conduct of this study.

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