We determined that targeted inhibition of HCK down regulates the features of tumor dormancy and drug tolerance, eliminating cells with a hybrid mesenchymal-epithelial phenotype, diminishing polyploidy and hyper-G2 endoreduplication, and suppressing features of drug resistance and tumor cell dormancy. Finally, using an ex vivo human tumor model that predicts clinical response and recreates the native tumor microenvironment, CANscript, we demonstrated that the unique signatures of tumor dormancy in refractory TNBC patients are tied to predictive clinical outcome. Conclusion: Expression of HCK drives cancer cells to drug tolerant cancer cells, subpopulation, resulting in cancer drug resistance and tumor cell dormancy, or quiescence. Targeted disruption of HCK will overcome these features of dormancy, which could majorly alter the course of treatment for patients with resistant disease. Using CANscript, we determined that nanoconjugated inhibitors can be studied ex vivo, and may therefore be a powerful tool to predict the pharmacodynamics and clinical response to therapy.

**Abstract**

**Background:** Triple negative breast cancer (TNBC) is an aggressive basal-like malignancy, which occurs more frequently than any other subtype of breast cancer. We recently discovered that TNBC can overcome drug pressure by switching to a hybrid basal-like (CD44HI) and epithelial-like (CD24+44-) cell state, which is accompanied by a transient period of dormancy, or quiescence. These dormant, ‘hybrid’ cells highly express the phosphorylated SRC family kinase, hematopoietic cell kinase (HCK).

**Methods:** Here, we used a panel of novel HCK inhibitors to study the effect of signaling-disruption in drug tolerance and tumor cell dormancy. Using flow cytometry and immunofluorescent microscopy we examined how HCK inhibition perturbs common features associated with dormancy such as proliferation and cell cycle status, intracellular levels of reactive oxygen species, and state of glucose metabolism. Next, we engineered molecular conjugates of HCK inhibitors using bio-compatible agents, which preferentially target dormant, drug tolerant cells. Finally, using an ex vivo human tumor model that predicts clinical response and recreates the native tumor microenvironment, CANscript, we demonstrated that the unique signatures of tumor dormancy in refractory TNBC patients are tied to predictive clinical outcome.

**Conclusion:** Expression of HCK drives cancer cells to drug tolerant cancer cells, subpopulation, resulting in cancer drug resistance and tumor cell dormancy, or quiescence. Targeted disruption of HCK will overcome these features of dormancy, which could majorly alter the course of treatment for patients with resistant disease. Using CANscript, we determined that nanoconjugated inhibitors can be studied ex vivo, and may therefore be a powerful tool to predict the pharmacodynamics and clinical response to therapy.