WNT DEPENDENCY IN PATIENT-DERIVED PANCREATIC ORGANOID MODELS FOR PRECISION MEDICINE APPROACHES

Haley A. Zlomke MD, Jacquelyn Zimmerman MD PhD, Toni Seppälä MD PhD, Reecha Suri MD, William R. Burns MD, Christopher Shubert MD MHA, Kelly J. Lafaro MD, Christopher L. Wolfgang MD PhD, Jin He MD PhD, Richard A. Burkhart MD

Background: Disease heterogeneity can drive the variable responses seen in pancreatic ductal adenocarcinoma (PDAC) to systemic therapies. Organoid models of disease have been shown to accurately recapitulate heterogeneity in early cultures and may be used to guide patient specific, personalized therapy. Historically WNT stimulation has been required to initiate cultures from fresh patient-derived specimens. Here, we evaluate the effect of exogenous WNT stimulation on PDAC organoid phenotype, tumorigenicity and chemotherapeutic sensitivity.

Methods: Patient derived organoids (PDOs) were established after digestion of fresh tissues obtained by biopsy or surgical specimens from eleven patients on an IRB-approved protocol. Seven were established using historical protocols reliant upon exogenous WNT supplementation. Four were established under two different protocols with and without WNT supplementation in the media. Growth characteristics, immunohistochemical analysis, and chemotherapeutic sensitivity analysis were performed under conditions of WNT supplementation and WNT restriction. Pharmacotyping was performed over clinically relevant dose ranges of five standard of care chemotherapeutics and a putative clinical response was determined by modeling based upon population distribution.

Results: WNT supplementation is not obligatory for the establishment, expansion and characterization of tissues derived for PDAC. In de-novo PDO establishment, WNT can be associated with an increased rate of success in establishment and an increased pace of biomass accumulation in the expansion phase. In lines previously established and expanded with WNT supplementation, the removal of WNT ligand did not significantly alter rates of cell proliferation and growth. No established cultures were lost after withdrawal of our exogenous WNT stimulation. The withdrawal of exogenous WNT can result in phenotypic changes to the culture that are evident under bright-light microscopy and immunohistochemical staining. Chemosensitivity determination can be performed by pharmacotyping in the presence, or absence, of exogenous WNT stimulation. Ex-vivo drug sensitivity, particularly to gemcitabine and irinotecan, can vary with WNT stimulation. Despite this heterogeneity, the putative clinical response of each individual tumor is not altered with WNT manipulation.

Conclusion: Intratumoral heterogeneity is a challenge to capture in real-time from patient-derived models of disease. When using these models to inform clinical care, the role of exogenous WNT stimulation and heterogeneity in culture conditions remains uncertain. Here we show that exogenous WNT can alter phenotype and growth rate. Despite this, pharmacotype was minimally altered with heterogeneous WNT conditioning, suggesting that patient derived organoid technology may be a robust predictor of clinical chemotherapeutic response. Exogenous WNT does not appear to alter the capacity of PDOs to serve as predictive biomarkers of clinical chemotherapeutic response.