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Et al.

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Implantable Microenvironments to Capture Stable-to- Aggressive Tumor Transition

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Clinical stability occurs when cancers reach a state where the disease neither advances nor regresses. Tumors can remain in this state for multiple years before progressing to more aggressive phenotypes. The mechanisms for maintaining a stable state and the factors that contribute to tumor activation are poorly understood. We hypothesized that an implantable biomaterial scaffold would be able to isolate a population of stable tumor cells that could then be used to study the transition to an aggressive phenotype. In this work we developed a tunable and highly controlled, porous acrylamide scaffold and subcutaneously implanted them in immunodeficient (NSG) mice. Prior to implantation scaffolds were seeded with a variety of different cell types. Specifically, human bone marrow stromal cells were supplemented with mouse stromal cells genetically engineered to express human cytokines to promote the generation of different tissue microenvironments in the scaffolds. After implantation the mice received an orthotopic injection of either human breast or prostate cancer cells. The tumors were allowed to generate metastases to the scaffolds and other tissues. Scaffolds were transplanted to non-tumor bearing mice once the tumor burden became exhaustive to the host to allow for further study of the microenvironment. The role of immune cells on the tumor microenvironment was also explored. Human peripheral blood mononuclear cells were isolated from donors and injected intravenously prior to transplantation. Bioluminescent imaging was used to capture tumor growth *in vivo* over a ten week period. The scaffolds were analyzed via immunohistochemical staining on thinly sectioned tissue and intact tissue cleared samples to characterize the tissue microenvironment and its effect on tumor progression. Our work has demonstrated the application of implantable tissue engineered microenvironments to study the phenomena of tumor stability *in vivo* and has uncovered some potentially important factors that drive the transition from stable to aggressive tumors.

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