**Title:** Investigating a new model of breast cancer metastasis with cellular MRI

**Trainee Name:** Natasha Knier

**Supervisor(s):** Dr. Paula Foster

**Structured Abstract:**

**Introduction:** Breast cancer is a leading cause of death in women due to the propensity of breast tumours to spread to distant sites such as the lymph nodes, lungs, liver, bone, and brain. The incidence of brain metastases is increasing, with the frequency of cases being reported as high as 50%. Studying tumour growth, dormancy, and cell apoptosis of this disease is challenging due to the limited experimental models available, and typical methods employed are limited to post-mortem examination. Previously, our group has used iron-based cellular MRI technology to track the fate of cancer cells in vivo, primarily using human mammary carcinoma cell lines (such as 231BR) in nude mice. Here, we use these cellular MRI techniques to characterize the growth of brain metastases of the 231BR cell line in the nude and severely immune-compromised NOD/SCid/Il2rg-/- (NSG) mouse.

**Methods:** NSG (n=10) and nude mice (n=10) received intracardiac injections of $1.5 \times 10^5$ iron-labeled GFP expressing 231BR cells. Images of the mouse brain and body were acquired with a clinical GE 3T MR system at Day 0, Day 21 (NSG endpoint), and Day 32 (nude endpoint). Day 0 brain images were assessed for signal voids, representing arrested iron-labeled 231BR cells; the % of black pixels was determined. Endpoint images were assessed for retained signal voids and brain metastases. Brain metastases were counted, and the % of brain occupied by metastases was determined, representing brain tumour burden. Body images were assessed for metastases.

**Results:** On day 0, iron-labeled cells were visualized throughout the brain in all mice as signal voids. The % of these signal voids decreased significantly between day 0 and endpoint for both strains, indicating that the clearance and retention of cells in the brain is similar between strains. At endpoint, brain metastases were evident in all mice as regions of hyperintensity. There was no difference in the number of brain metastases in NSG mice (M=69.00±15.12) compared to nude mice (M=58.33±33.70), however, tumour volumes in nude mice (M=0.38mm$^3$±0.50) were significantly larger than in NSGs (M=0.033mm$^3$±0.036, p<0.0001). Tumour burden (% of the brain with metastases at endpoint) was not significantly different between strains; however, body images indicated that the NSG mice had numerous metastases located in the liver, lungs, and lymph nodes.

**Discussion:** Characterization of the NSG and nude mouse as an experimental platform to study breast cancer brain metastasis is necessary to study the fate of this disease in vivo. Here we show that the 231BR cell line grew differently in NSG mice compared to nude mice. While both are immune-compromised strains, NSG mice are more severely deficient, which led to increased brain tumour burden and numerous liver and lung metastases that are not present in the nude model. This work demonstrates the valuable role that imaging can play toward credentialing these important preclinical models.