1.0 Purpose
The purpose of SOP 6.14 is to provide in detail how to perform an intra-medullary tibia injection in mice. Cancer stem cells and mesenchymal stem cells (MSCs) obtained from normal human breast tissue, normal human hip bones and/or cancer stem cell lines may be injected into NOD/SCID mice to model cancer stem cell and MSC interactions in our xenograft models.

2.0 Scope
SOP 6.14 is intended to cover all resources, personnel and equipment in the BCR laboratory.

3.0 Procedure
3.1 A few hours before transplantation, irradiate mice that are at least 8 weeks old (and males if possible as they are easier to use with this type of injection) with 300 cGy from an x-ray irradiator (Mark I, Model 25, J.L. Shepherd).
3.2 Use isoflurane to anesthetize the mice (ketamine mix is not needed for such a short procedure).
3.3 Using Nair, remove all of the hair surrounding the knee joint.
3.4 Flex the knee at 90° and have the proximal side of the tibia drawn to the anterior.
3.5 Insert a 26-gauge needle into the joint surface of the tibia through the patellar tendon and then down into the bone marrow cavity with steady pressure, to create a hole/passage.
3.6 Subsequently change to a 28-gauge needle in order to transfer the MSCs.
3.7 Use approximately 1x10⁶ DsRed and Luciferase labeled MSCs suspended in 10-50 uL PBS or matrigel (containing a cell concentration of 2x10⁷ per mL so that you are injecting one million cells per injection) and transplant directly through the bored bone holes into the tibia bone marrow cavity of NOD/SCID mice using a Hamilton/insulin syringe equipped with a 28-gauge needle.
3.8 Inject 10 uL of PBS to the right tibia of the control group.

4.0 Applicable References


Irradiation SOP

5.0 Change Description

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