Non Controlled Version

**Note: Controlled Versions of this document are subjected to change without notice*

Snap Freezing Protocol For Tissue

I. Purpose

To provide details regarding the snap freezing and storage of tissue samples.

1. Preparation of Workstation and BSC (Biological Safety Cabinet)

1.1. Follow standard practices of operating a BSC and thoroughly wipe it down before use.

1.2. Clean and prepare and a workstation close to the BSC. The following equipment and reagents are required:

a. Styrofoam box containing dry ice
b. Vessel containing liquid nitrogen
c. Tweezers or forceps to transfer tubes/vials
d. Styrofoam box containing wet ice
e. Protease inhibitor*
f. P10 pipette and tips
g. Appropriate PPE (personal protective equipment) to handle dry ice and liquid nitrogen

*A protease inhibitor is mandatory to add to the tissue prior to snap freezing if the sample is to be used for downstream protein studies (e.g. ChIP).

2. Tissue Sample Handling and Addition of Protease Inhibitors in BSC

2.1. Cut, tube transfer, manipulate, add protease inhibitor or handle the tissue sample in the BSC if required.

2.2. For ChIP (Chromatin Immunoprecipitation) studies, 100-200mg of tissue is required.

2.3. Ensure the tube/vessel is completely closed, wipe with 70% ethanol. Label the tube.

3. Snap Freezing Tissue and Transfer to Storage

3.1. Immediately submerge the sample tube into liquid nitrogen for snap freezing of the tissue.

3.2. Snap freezing is immediate. Leave the tube submerged for ~30 sec to completely snap freeze the tissue.

3.3. Transfer the tube quickly to the Styrofoam box containing dry ice.

3.4. Transfer the tube to a -80°C freezer for long-term storage.