Snap Freezing of Tissue

Section:	**Biobank - OHIRC**	**SOP No:**	**B-3-007.00**
Issued By:	**Biobank**	**Approved By:**	**Senior Management**
Supersedes:	**N/A**	**Effective Date:**	**October 12, 2018**

1. PURPOSE

This SOP outlines standardized procedures for UOHI personnel to follow for snap freezing of tissue samples for the Biobank. This SOP is intended to ensure that tissue samples obtained from consented participants will be processed in a consistent, safe and efficient manner while eliminating the risks of contamination. This SOP does not cover detailed safety procedures for handling tissue; personnel must follow institutional bio-safety guidelines. This SOP has been adapted for the UOHI Biobank use from the CTRNet Standard Operating Procedure: Snap Freezing of Tissue.

2. INTRODUCTION

Tissue samples are a precious resource in the Biobank and procedures must be followed to obtain products with high integrity and quality. Tissue samples are harvested by qualified clinical personnel from patients who have been through the informed consent process and agreed to participate in the UOHI Biobank program ("participants"). Snap frozen tissue is suitable for preparation of DNA, RNA and protein.

3. ROLES AND RESPONSIBILITIES

Personnel	Responsibility / Role
Biobank Laboratory Technician / Biobank Manager / Research Coordinator / Research Assistant	Transport, Process and Store Tissue and Derivatives

4. SUPPLIES

- Gloves (non-latex recommended) worn to protect personnel handling tissue specimens
- Liquid Nitrogen
- Container (Dewar vacuum flask) for liquid nitrogen
- Safety glasses / insulated gloves for personnel handling liquid nitrogen tank and storage container
- Clean forceps
- Clean scalpel / scissors for cutting/trimming tissue
- Suitable container to place specimen for cutting/trimming (clean flat dish)
- Isopropyl alcohol for storing clean forceps and scalpels
- Screw top cryovials (2D Matrix or marked with indelible ink) for storage of frozen tissue. Labels will not adhere.
- Clean underpads for bench surface
- Green soap for disinfecting scissors and forceps

5. PROCEDURES

5.1 Timing for Tissue Processing

5.1.1 Communicate with responsible personnel to advise when tissue will be collected and to arrange for timely processing.

5.1.2 Fresh tissue should be frozen as soon as possible. Optimally, tissue should be frozen within 30 minutes from resection.

5.2 Verification of Information on Tubes

5.2.1 Verify participant 1D barcode number on accompanying Tissue Collection and Processing Worksheet and ensure that it corresponds with the information on labels on tissue collection tubes.

5.3 Snap Freezing of Tissue with Liquid Nitrogen

5.3.1 Freezing is performed by the laboratory technician/technologist or trained personnel designated by the biobank.

5.3.2 Do not place the sample in contact with formalin or serum at any point in the process.

5.3.3 Ensure that the resected tissue never desiccates or is contaminated by surrounding tissue or other samples. Use clean scalpel / scissors and forceps between samples to avoid cross contamination between samples.

5.3.4 Do not freeze the tissue directly on ice.

5.3.5 Have materials and equipment ready. Have as many 2D-matrix or indelible ink marked cryovials as needed ready.

5.3.6 Use clean forceps to remove tissue specimen from collection tube. Rinse in new tube with phosphate buffered saline. Place specimen on clean cutting surface dish. Cut specimen into 2 to 3 pieces or more as appropriate. If the sample is too large in size, longer freezing time will result in ruined morphology.

5.3.7 Place each tissue specimen into an empty cryovial, close the cryovial, and immediately submerge the cryovial in liquid nitrogen. The specimen should freeze within 30-60 seconds.

5.3.8 Once snap frozen, transfer the samples for storage (on dry ice if not close by) to the designated -80 °C freezer at UOHI equipped with audible or remote alarm, for eventual transfer to vapour phase LN2 tanks at OHRI (refer to SOP B-3-003 for details).

5.4 Accessioning of Samples

5.1 Complete the Tissue Collection and Processing Worksheet with the date and time of tissue processing and storage. Document the exact location of tubes in freezer.

5.2 Accession tissue samples into biorepository inventory database system. Utilize scanner (Hamilton or hand scanner) to accession the 2D labels embedded in bottom of cryovials and 1D labels permanently affixed to cryoracks.

6. ACRONYMS

OHIRC - Ottawa Heart Institute Research Corporation
SOP- Standard Operating Procedure
UOHI - University of Ottawa Heart Institute

7. RELATED REGULATIONS AND/OR POLICIES

References:

7.1 Tri-Council Policy Statement 2; Ethical Conduct for Research Involving Humans; Medical Research Council of Canada; Natural Sciences and Engineering Council of Canada; Social Sciences and Humanities Research Council of Canada, December 2010
http://www.pre.ethics.gc.ca/eng/policy-politique/initiatives/tcps2-eptc2/Default/

7.2 ICH Guidance E6: Good Clinical Practice: Consolidated guideline
http://www.hc-sc.gc.ca/dhp-mps/prodpharma/applic-demande/guide-ld/ich/efficac/e6-eng.php

7.3 Declaration of Helsinki. http://www.wma.net/en/30publications/10policies/b3/index.html

7.4 CTRNET Standard Operating Procedures, Canadian Tumour Repository Network
http://www.ctrnet.ca/operating-procedures

7.5 OTRN OICR Standard Operating Procedures for Biorepositories, Ontario Tumor Repository Network/Ontario Institute for Cancer Research
http://oicr.on.ca/search/apachesolr_search/SOPs%20for%20Biorepositories

Associated OHIRC SOPs:

UOHI Biobank SOP No: B-3-006.00 –Tissue Collection and Transportation

8. APPENDICES

8.1 Appendix A – Tissue Collection and Processing Worksheet Example

9. HISTORY

SOP #	Effective Date	Review Date	Summary of Changes
B-3-007.00			

Appendix A – Tissue Collection and Processing Worksheet Example

Subject's DOB: _ _ _ / _ _ _ _
Month Year

Bar Code number: _____

Collection Date: _ _ /_ _ _ / _ _ _ _ Time: _ _ _ _
Day Month Year hh:mm

1) Atrial tissue	2) Other tissue	3) Other tissue	4) Biopsy	5) Biopsy	6) Biopsy

Source of Tissue: OR _____ Cath Lab:_____ Other: _____

Source details: ☐ Atrial cannula site ☐ Myocardial biopsy ☐ Skin punch biopsy Other:_____

CRF Completed by: _____

Samples Received in Laboratory: _ _ /_ _ _ / _ _ _ _ Time: _ _ _ _
Day Month Year hh:mm

Atrial Tissue		
Processing Time	Stored in – 80° C freezer Time	Number of Tissue Aliquots
_ _ _ _ hh:mm	_ _ _ _ hh:mm	_____

Other Tissue		
Processing Time	Stored in – 80° C freezer Time	Number of Tissue Aliquots
_ _ _ _ hh:mm	_ _ _ _ hh:mm	_____

Myocardial Biopsy		
Processing Time	Stored in – 80° C freezer Time	Number of Tissue Aliquots
_ _ _ _ hh:mm	_ _ _ _ hh:mm	_____

Skin Punch Biopsy		
Processing Time	Stored in – 80° C freezer Time	Number of Tissue Aliquots
_ _ _ _ hh:mm	_ _ _ _ hh:mm	_____

Completed by: _____