SUSAN G. KOMEN FOR THE CURE | TISSUE BANK AT THE IU SIMON CANCER CENTER

Standard Operating Procedure (SOP) 005V4.0

Processing and Storage of Breast Tissue
SPREC TIS-BPS-N-B-SNP-A-C (1)
SPREC TIS-BPS-N-B-NBF-G-P (1)

Effective Date: Sept. 1, 2012
Author: Theresa Mathieson M.S. Biospecimen Manager

Approved by:

Materials:
Cryostorage tubes: Corning 2.0ml Cryogenic Vials. (Fisher cat. #0337421)
Bleach: Any commercially available
Polypropylene Containers: (Fisher cat. #14 955 103)
Liquid Nitrogen: (ICTSI-SSF R3 C156)
Forceps: (Fisher cat. #08-887)
Embedding (Tissue) Cassettes: (Fisher cat. #B1000729BL)
10% Neutral Buffered Formalin (Fisher 245-684)
Safety Goggles: (Fisher cat. #19-181-504)
Stiff Bristled Brushes: (Fisher #03-635)
Emesis Basin or a receptacle to hold diluted bleach and forceps:

Tissue Processing:
Within 5 minutes (or less) of procurement one breast tissue core is placed into an embedding cassette and the cassette closed. Tissue cassettes are labeled in pencil prior to tissue acquisition. The cassette is placed into 10% buffered formalin and stored in the formalin at room temperature until the end of the collection event. At the end of the collection event, the cassettes are delivered to Research Pathology where the tissue will be embedded in paraffin per Pathology protocol. A 5μ section will be obtained for hematoxylin and eosin staining. A photomicrograph of the stained sectioned will be entered into the database.

Universal Precautions are MANDATORY. Eye protection is mandatory every time liquid nitrogen is handled to protect against injury due to splashing. Standard laboratory personal protective equipment (e.g. closed toe shoes, full cover of legs and feet, and goggles) will be worn when handling coolants.

Flash freezing in liquid nitrogen provides excellent specimen integrity and a wide array of options for tissue analysis. **Within 5 minutes (or less) of procurement,** enough liquid nitrogen (LN_2) to cover the specimen is transferred to a new polypropylene container. Using

a clean forceps. the remaining cores are immediately placed into LN2 and snap frozen for 30-60 seconds. The frozen cores are placed in labeled, chilled cryovials. All cryovials are to have bar-coded labels placed on the vial prior to tissue acquisition.
The samples are scanned with a barcode reader, logged into cryoboxes and placed on dry ice for transport to the storage facility.

Used forceps are placed in a bleach solution (1/10 bleach/water) in an emesis basin. When time allows or a clean forceps is needed, the forceps in the bleach solution are gently scrubbed with a brush, rinsed in water and allowed to air dry.

Temperature of specimens following acquisition but prior to storage: Cryovials containing the specimens are to be placed and remain on dry ice until all specimens have been procured.

Storage of Tissue: Once all the samples are procured, the cryovials are to be transported on dry ice to the LN_2 freezer. Tissue samples are stored in liquid nitrogen vapor, (-166.2°C to -195.1°C from top to bottom of tower).

Temperature for collection and processing: All tissue procurement and processing events are done at room temperature.

Standardization: All variables including the time between excision and snap-freezing, time stored on dry ice prior to placement in the LN_2 freezer, time stored prior to shipment and/or utilization will be entered into the database.

Oversight: All adverse and unexpected events will be recorded in the database and will be addressed by the Internal Advisory Committee. This includes all phases of the process: donation, storage and retrieval, processing, and utilization.

References:

1. Sabine Lehmann et.al. International Society for Biological and Environmental Repositories (ISBER) Working Group on Biospecimen Science. Standard preanalytical Coding for Biospecimens: Review and Implementation of the Sample PREanalytical Code (SPREC).Biopreservation and Biobanking Vol. 10 No.4, 2012

Bibliography

1. Karlsson JO, Toner M. *Long-term storage of tissues by cryopreservation: critical issues.* Biomaterials. 1996; **17**(3):243-56

2. Morente MM, Mager R, Alonso S, Pezzella F, Spatz A, Knox K, Kerr D, Dinjens WN, Oosterhuis JW, Lam KH, Oomen MH, van Damme B, van de Vijver M, van Boven H, Kerjaschki D, Pammer J, Lopez-Guerrero JA, Llombart Bosch A, Carbone A, Gloghini A, Teodorovic I, Isabelle M, Passioukov A, Lejeune S, Therasse P, van Veen EB, Ratcliffe C, Riegman PH. *TuBaFrost 2: Standardising tissue collection and quality control procedures for a European virtual frozen tissue bank network.* Eur J Cancer. 2006; **42**(16):2684-91.

3. Qualman SJ, France M, Grizzle WE, LiVolsi VA, Moskaluk CA, Ramirez NC, Washington MK. *Establishing a tumour bank: banking, informatics and ethics.* Br J Cancer. 2004; **90**(6):1115-9

4. Villalba R, Eisman M, Fornes G, Mesa A, De Torres P, Ariza MJ Gutierrez Guzman A, Gomez Villagran JL. *Implementation of a Quality Plan (ISO 9002) In a Regional Tissue Bank.* Cell Tissue Bank. 2001; **2**(1):45-49.

5. Leyland-Jones, Brian R. et al., *Recommendations for Collection and Handling of Specimens from Group Breast Cancer Clinical trials, from Onsite Collection through Shipping to the Central Bank.*2007 Unpublished.

Electronic Resources

First-Generation Guidelines for NCI-Supported Biorepositories:
http://biospecimens.cancer.gov/global/pdfs/NCI_Best_Practices_060507.pdf
http://ctep.cancer.gov/forms/guidelines_fresh_tissue.pdf
http://www.cnio.es/ing/programas/progTumor11.asp#protocolo08 (CNIO, Spanish National Cancer Centre, Madrid)
http://epi.grants.cancer.gov/CFR/bio_fresh_tissue_app.html#bypass
www.pathology.washington.edu/clinical/neuropath/docs/Muscle%20Frozen.doc
http://www.metametrix.com/content/HowToOrder/SpecimenCollection
http://www.isber.org/Pubs/BestPractices2008.pdf