



*Obafemi Awolowo University Teaching Hospitals Complex – ARGO Biobank Center
P.M.B. 5538
Ile-Ife, Osun State
Nigeria*

Tissue Harvesting and Routine Freezing

1.0 PURPOSE

The African Research Group for Oncology (ARGO) collects tissues from consented patients’ samples. Tissue samples are obtained by qualified personnel and are only collected for the ARGO only if present in excess to that required for pathological assessment and diagnosis. The purpose of this document is to outline standardized procedures for the ARGO researchers to follow during tissue harvesting.

2.0 SCOPE

This Standard Operating Procedure (SOP) describes how tissues are harvested and processed for routine banking by the ARGO researchers. Refer to the safety manual for information about handling human biohazardous materials and standard precautions.

3.0 ROLES AND RESPONSIBILITIES

This SOP applies to all ARGO personnel that are responsible for harvesting tissue. Applicable staff may include the following roles:

ARGO /Designated Personnel	Responsibility/Role
Surgeon / Physician	Obtains patient consent and tissue
Pathologist	Diagnose of tissue malignancy, gross of tissue and resection of excess tissue for the ARGO research purposes.
Research Assistant	Harvests, processes, stores tissue samples. Transports tissue samples.

4.0 MATERIALS, EQUIPMENT AND FORMS

Materials and Equipment
Container with ice
Appropriate container for resected tissue (Petri dishes)
Markers and pens
Disposable forceps
Clean scalpels for trimming tissue
Cryovials for storage of frozen tissue
PPE
Sufficient appropriate labels for collection tubes
Needle/sharps disposal unit
Weight Scales

5.0 PROCEDURES

This procedure is intended to ensure that tissue samples will be collected from patients in a timely and efficient manner while eliminating the risks of contamination. Samples that has been adequately processed is vital in obtaining products with high integrity and quality for genomic and proteomic techniques.

5.1 Tissue Harvesting

- 5.1.1 ARGO staff will give the physician or pathologist all the necessary tools to harvest tissue with any protocol specific requirements. For example, media or sterile tools for sterile collection.
- 5.1.2 The physician or pathologist will place the sample in the petri dish or media in a container of ice prepared by ARGO staff.
- 5.1.3 ARGO staff will ensure that the resected tissue never desiccates or is contaminated by surrounding tissue or other samples. If appropriate, change scalpel blades between dissecting tumor tissue and surrounding uninvolved tissue.
- 5.1.4 The ARGO staff will ensure that all samples are maintained and processed on ice over a metal ice plate and processed immediately to ensure preservation of the tissue. Metal ice plates are used to facilitate the cutting of samples into smaller aliquots.
- 5.1.5 Treat all tissue as potentially infectious.
- 5.1.6 Select tumor tissue for banking without compromising the tissue that will be sent to research investigators.
- 5.1.7 Attempt to preserve and store normal adjacent tissue as well.

- 5.1.8 If possible, allow for the banking of multiple samples from one specimen. For larger tumors and resections, tumor should be harvested from different spatial areas.
- 5.1.9 An accompany paraffin block should be generated for each tissue type. Exceptions will be made for very small tissue samples.
- 5.1.10 Timing is critical. The elapsed time between resection and freezing of a given sample should be kept at a minimum. Document all relevant time points in the database.

5.2 Freezing

- 5.2.1 Freezing is performed by ARGO Research Assistants.
- 5.2.2 All non-protocol routine banking of samples will be frozen by placing in the -80°C freezer. A representative sample of both tumor and normal will be used to generate paraffin blocks, if needed.
- 5.2.3 For the frozen tissue samples, attempt to have no more than **six** cryovials for each distinct tissue type for a given sample, unless otherwise indicated.
- 5.2.4 To avoid freeze/thaw cycles, it is recommended to have about **3 pieces** of tissue per cryovial. The tumor and normal samples should be cut into smaller **0.8cm x 0.8cm** pieces.
- 5.2.5 If there is abundant tumor, attempt to harvest a larger amount of tumor (depending on size and availability). For very large tumors, the first two cryovials should be minimized to 3 pieces and any subsequent cryovials can be filled to maximum.
- 5.2.6 Samples and cryovials are to be kept on ice during processing until frozen in liquid nitrogen.
- 5.2.7 Document the weight of all aliquots in milligrams in the database.
- 5.2.8 Accession the samples in the database and print appropriate labels on the cryovials.
- 5.2.9 All cryovials are suitable for placement in the -80°C.
- 5.2.10 Assign a location in the storage box and document the location in database. Place the sample at the assigned location.
- 5.2.11 Snap frozen tissue is suitable for preparation of DNA, RNA and protein. Do not use samples that had contact with formalin at any point in the process.
- 5.2.12 All routine samples not for distribution should be frozen and kept in the -80°C freezer.

6.0 APPLICABLE REFERENCES, REGULATIONS AND GUIDELINES

- 6.1 Best Practices for Repositories I. Collection, Storage and Retrieval of Human Biological Materials for Research. International Society for Biological and Environmental

Repositories (ISBER)
http://www.isber.org/Search/search.asp?zoom_query=best+practices+for+repositories

6.2 US National Biospecimen Network Blueprint
<http://biospecimens.cancer.gov/resources/publications/reports/nbn.asp>

7.0 REVISION HISTORY

SOP Number	Date Reviewed	Reviewed By	Comments