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Biobank / Laboratory Standard Operating Procedure (SOP)

This SOP shall govern the procedure for storage of blood, urine, stool and tissue (normal and tumour) in the Biobank, and maintenance / cleanliness of the Laboratory:

Materials / Consumables Required:

- Cryovial - for tissue (breast cancer, colorectal cancer, normal) storage
- Anticoagulant bottle (EDTA) – for plasma preparation
- Plain bottle – for serum preparation,
- Universal bottle – for urine/stool collection
- PCR rack – for arranging sample containing cryovial for onward storage into the -80°C freezer.
- Cold ice pack – for sample transportation from collection point to the laboratory.
- Minus 8°C transport refrigerator to transport samples from location outside OAUTHC
- Others – surgical blade, Needle and syringe (2mls, 5mls 10mls), gloves, goggles, face mask, apron, cotton wool, methylated spirit, savlon, petri dish, micro-pipette/tips, pasteur pipette

Recruitment / Persons responsible for Sample collection

- Patient's recruitment takes place in the clinics, theatres, endoscopy room and/or recruitment hall.
- Informed consent for recruitment into the study and for samples collection [blood, tissues (normal and tumour), urine, stool as may be appropriate for each protocol] often obtained at the point of recruitment.

- Collection of fresh tissues is done by the surgeon during surgery or the pathologist from gross specimen.
- Collection of blood sample is carried out by the research assistant at the point of recruitment.
- Collection of urine / stool sample is done by the patient having being appropriately instructed by the research assistant.

Sample Collection / Processing / Storage

1. Tissue

(a) Normal and Tumour

- Tumour must be taken from the gross tumor tissues
- Normal tissue must be taken from the specimen apparently appearing normal. It will be taken far from the grossly tumorous tissue.
- Transport fresh tissue(s) collected into labelled cryovial from respective collection point to the laboratory using ice pack, transport freezer or dry ice.
- The warm and cold ischemic time should be less than 30 minutes and should be documented on the specimen log.
- Arrange into PCR rack and store in the appropriate labelled compartment of the -80°C freezer.

(b) Preserved RNA tissue

- Collect fresh tissue(s) inside labelled plain bottle.
- Add '*RNA later reagent*' and store in the fridge for not more than 30 days
- The samples should be centrifuged using cold centrifuge within 30 days of samples collection.
- Dispensed RNA preserved tissue into the cryovial and store in the appropriate labelled compartment of the -80°C freezer.

2. Blood / Hematological Samples

(a) Serum:

- Collect 5mls of blood sample into red top bottle labelled SER (plain bottle).
- Allow to clot at room temp for 30-60 minutes.

- Centrifuge for 15 minutes at 1,500 rpm.
- Pipette aliquots of serum supernatant into labelled cryovials / eppendorf tubes.
- Arrange into PCR rack and store in the appropriate labelled compartment of the -80°C freezer.

(b) Plasma / Buffy Coat / Red Blood Cell

- Collect 5mls of blood sample into the purple top bottle labelled PLA (anticoagulant bottle).
- Centrifuge for 15 minutes at 1,500 rpm.
- Pipette aliquots of the first layer **plasma** supernatant into labelled cryovials / eppendorf tubes.
- Then, pipette aliquots of second layer **buffy coat** supernatant into labelled cryovials / eppendorf tubes.
- Then, dispense the last layer **RBC** sediment into labelled cryovials.
- Arrange each into appropriately labelled PCR rack and store in the appropriately labelled compartment of the -80°C freezer.

3. Urine

- Instruct consented patient how to collect midstream urine (MSU) into universal bottle enclosed in a biohazard bag.
- Transport MSU immediately through ice pack from the recruitment point to laboratory.
- Dispensed MSU into labelled cryovial.
- The urine must arrive the Freezer within one hour of passage of the urine
- Arrange into PCR rack and store in the appropriate labelled compartment of the -80°C freezer.

4. Stool

- Give consented patient universal bottle, instruct him/her to scoop stool sample into it and return same within 24 to 48 hours after being screened.
- Transport stool sample collected immediately through ice pack from the recruitment point to the laboratory.
- Dispense stool sample from universal bottle into labelled cryovial.

- Arrange into PCR rack and store in the appropriate labelled compartment of the -80°C freezer.

NB: Protocol-specific sample labelling is done with research number with or without patient's initials.

All samples remain stored at -80°C freezer in the laboratory for as long as it is possible before it is subsequently transferred to MSKCC for analysis.

Laboratory Maintenance / Precautions

- Biological samples / tissues should be properly dispensed into cryovials and deposited inside -80°C freezer.
- Laboratory should be temperature-controlled, precisely between 18-20°C.
- Daily temperature of freezers should be monitored and recorded in the morning (8am), noon (12pm) and evening (4pm).
- Cryovials should be properly labelled with cryo-marker to prevent mix-up of samples.
- Laboratory room and slabs should be disinfected using methylated spirit or alcohol-based disinfectants.
- Used materials (such as gloves, face-mask, cryovials) should be properly disposed into the bio-hazard waste-bin.
- Date and time of tissue collected and shipping records should be filed.
- Unprocessed samples and any known variation or deviation from protocol should be immediately reported to the principal investigator.