*If you have questions, please call Ken at any time, day or night.*

*(859) 797-0843*

1. **PURPOSE:** This SOP explains the necessary steps for obtaining cardiovascular tissue removed from but not limited to, LVAD placement or removal, heart transplant, heart organ donation, or blood plasma samples. The IRB authorizes certain personnel to obtain cardiovascular tissue removed during these procedures for research. There are currently three participating labs; Campbell, Smyth, and Abdel-Latif. The procurement of samples is time-sensitive and a biologic material that must be carefully handled. Collection of tissue products must be carefully coordinated and stored.
2. **SCOPE and RESPONSIBILITIES:**   
   1. Scope:
      1. The activities described in this SOP are to ensure efficient and safe collection, transport, dissection, and storage of all cardiovascular human tissue samples received.
      2. Approval for samples are based on IRB #08-0338-F2L, PI: Kenneth Campbell.
   2. Responsibilities:
      1. Principal Investigator
      2. Research Coordinator
      3. Program Coordinator
      4. Program Fellows
      5. Postdoctoral Researchers
      6. Graduate Students
      7. Undergraduate Students
3. **DEFINITIONS and ABBREVIATIONS:**
   1. Definitions:

* Autoclave - a strong heated container used for chemical reactions and other processes using high pressures and temperatures, e.g., steam sterilization.
* Biohazard - a risk to human health or the environment arising from biological work, especially with microorganisms.
* Hashcode – used to identify sample without providing any patient information.
* Liquid nitrogen – nitrogen in a liquid state at an extremely low temperature used as a coolant.
* Procurement - the action of obtaining or procuring something.
* Sterile field - an area immediately around a patient that has been prepared for a surgical procedure.
  1. Abbreviations:
* CT – Cardiothoracic
* Hep B – Hepatitis B
* IRB – Institutional review board
* KODA – Kentucky Organ Donor Affiliates
* LA – Left atrium
* LV – Left ventricle
* LVAD – Left ventricular assist device
* OR – Operating room
* OSHA – Occupational Safety and Health Administration
* PT – Procurement Team
* RA – Right atrium
* RV – Right ventricle

1. **SAFETY AND QUALITY CONTROL:**
   1. Follow appropriate precautions based upon OSHA guidelines, infection control policies, and/or the institutional procedure manual for the handling of bodily fluids.
   2. The Hep B vaccine is optional but highly recommended when working with human tissue.
   3. Always follow sterile procedure when in the OR. Never touch equipment, surgical instruments, patient, and/or personnel that are in the sterile field.
   4. Ensure all necessary containers are labeled with a biohazard sticker.
   5. Ensure use of biohazard bags for the autoclave.
   6. Always keep human tissue samples on ice, dry ice, or in liquid nitrogen for proper preservation.
2. **MATERIALS, REAGENTS, and EQUIPMENT:**

Various materials, reagents, and equipment are required for the procurement of human cardiac tissue samples. Generalized items may include, but are not limited to,

* OR attire – includes OR scrubs, hat or bouffant, mask, shoe covers
* UK ID badge for access to OR
* Portable cooler for transport of tissue
* Exam gloves
* Sharpie marker
* 2ml Cryogenic vials (drawer 13D marked Eppendorf tubes)
* Hepranized tubes
* Underpad (drawer 11E)
* Weigh boats x 2 (shelf 7H)
* Dissecting instruments
  + One large silver scissor
  + One small silver scissor
  + One large black scissor
  + One small black scissor
  + One small forceps
  + One medium forceps
* Liquid nitrogen, dry ice, normal ice (rm MN518)
* Liquid nitrogen dewar (rm MN508)
* Cryrogenic box
* Laddle
* Envelop with hashcode and notecard enclosed
* Black binder of Human Tissue Recovery Log sheet (shelf 1K)
* CIDecon disinfectant
* Conflickt (Decon) disinfectant spray
* Tupperware container for instrument cleaning
* Biohazard bag (drawer 3C)
* Autoclave (rm MS

1. **PROCEDURE:**
   1. To gain appropriate access to be a part of the Procurement Team, contact the Biorepository Program Coordinator.
   2. Procurement Team phone:
      1. During standard operating hours the Program Coordinator will carry the phone and retrieve any samples. After hours, weekends, and/or the absence of the Program Coordinator, approved lab personnel will be “on-call” specimen(s) procurement.
      2. The phone should be monitored and answered 24 hours a day, 7 days a week.
      3. Personnel monitoring the phone should remain within 30 minutes of the hospital to avoid any delay in obtaining specimen(s).
      4. Answer **ALL** calls starting with (859) area code or pre-stored contacts.
   3. Preparation for collection of tissue sample:
      1. All personnel that might encounter PHI must have prior IRB approval. HIPAA guidelines must be followed when handling PHI.
      2. A member of the OR circulating nurse will call the Procurement Team phone (859) 619-0994 to alert of upcoming procedure. Please ask what type of procedure is being performed so the team will know what type of sample to prepare for and what time the procedure is expected to begin. You may also ask the nurse to call back when the surgeon has made the first cut.
      3. Prepare a specimen bag filled with pre-labeled cryogenic vials and hashcode envelope if one is not pre-made (e.g., D213A; LVAD; LV2)
      4. The minimum tissue types for collection are:

* LVAD
  + 1 x Epi Fat
  + 2 x LV (you may need 6 x LV, 2 each for transmural, but at least use 2)
* Transplant
  + 2 x Epi Fat
  + 8 x LV Epi
  + 8 x LV Mid
  + 8 x LV Endo
  + 8 x Septum
  + 8 x RV
  + 3 x RA
  + 3 x LA if applicable
* Donor heart
  + 20 x LV Epi
  + 20 x LV Mid
  + 20 x LV Endo
  + 20 x RV
  + 12 x Septum
  + 2 x Epi Fat
  + 6 x RA
  + 6 x LA
    1. Setup lab counter space with underpad, dissecting tool kit (include one small silver scissor, one large silver scissor, one small black scissor, one large black scissor, one small forcep, and one medium forcep), and two weigh boats.
    2. Take cooler with biohazard sticker to MS518 (entry code 749518) and fill bottom with normal ice.
    3. Fill liquid nitrogen bucket with liquid nitrogen.
    4. Change into OR scrubs. Scrubs can be removed and returned from scrub machines using UK ID badge.
  1. Collection of specimen(s):
     1. Enter the designated OR room with OR scrubs, hat and/or bouffant, face mask, and shoe covers. Hat, mask, and shoe covers are located at the entrance hallways of the OR.
     2. Locate the circulating RN to make them aware of your arrival. Ask for a patient ID sticker.
     3. Do not go within 2 feet of any sterile area or personnel.
     4. The specimen will be placed in an orange container by the surgeon or surgical tech. The nurse will take the container from the sterile field and give specimen(s) to procurement team. Wear exam gloves if you handle the sample.
     5. Ask the nurse the time when cardiopulmonary bypass was turned on and/or the crossclamp time.
     6. Remove mask after exiting OR.
  2. Transporting specimen(s):
     1. Do not use public elevators for biohazard material. You ***must*** use service elevators or stairwells.
     2. Take sample to the Campbell lab (MN508) for dissection.
  3. Dissecting heart tissue:
     1. Once you have returned to the lab place patient ID sticker on the notecard inside the hashcode envelope. Lock envelope in drug drawer.
     2. Before dissection take a picture of the specimen.
     3. Place ice in one weigh boat. Place the other weigh boat on top. Use this weigh boat to sit sample while you dissect to keep cool. Work as quickly as possible to minimize sample degradation.
     4. Ventricular tissue should be processed first. Separate ventricular tissue into epicardial, midmyocardial, and endocardial. As you dissect, place layers in a cryogenic vial and drop in liquid nitrogen bucket. Vials should be labeled with hashcode, procedure, and tissue.
     5. Process the atrial tissue in the same way.
     6. If you were given aorta, place in cryogenic vial. Label the vial and drop into liquid nitrogen.
     7. Blood samples should be processed last.
     8. After dissection of organ donor heart, any remaining tissue need to be taken to surgical pathology. Heart transplant tissue needs to be returned to the OR.
  4. Storage of specimens:
     1. Remove all of the frozen tubes, vials, etc. from the liquid nitrogen using a ladle and place in a cryogenic transfer storage box.
     2. Place cryogenic transfer box in an open storage slot in one of the liquid nitrogen dewars.
     3. Fill out the Human Tissue Recovery Log in the black binder (shelf 1K). Be sure to write the location of the transfer box.
  5. Cleaning, disinfecting equipment:
     1. Place dissecting tools in tupperware. Fill half way with Decon and the rest with water. Let soak for 10 minutes.
     2. Dump ice from cooler in sink.
     3. Spray countertop and cooler (inside and out) with Conflickt disinfectant spray. Leave for 10 minutes.
     4. After 10 minutes, dump the fluid from the tupperware in the sink. Rinse the tools and tupperware with water. Lay a paper towel on the countertop. Place tools on the paper towel to dry. If possible, open scissors to ensure proper drying.
     5. After 10 minutes, wipe down countertop and cooler.
     6. Take tupperware to MN507 for cleaning. Follow lab dish cleaning protocol.
     7. Place all material used for dissection in a biohazard bag (shoe covers, gloves, underpad, weigh boats, mask, hat, paper towels).
     8. Take biohazard bag to autoclave.
  6. Autoclave biohazardous material:
     1. After collecting all of your biohazard waste, including bench liners (blue pads) and paper towels, *loosely* the bag and place simply into one of the *white* rectangular autoclave bins.  (Keep the seal loose so that any steam coming from the bag will be able to flow out.  If it is tight, your bag may explode.  This is not pleasant.)  You may also place the bag into the RED cylindrical container but do not forget about them!  (The RED container is probably best if the BetaStar Autoclave is out of service.)
     2. Consider adding a piece of autoclave-indicator tape (also in drawer 3C) to the side of the bag.  The indicator tape changes color when exposed to high temps for a certain period of time.
     3. If it is after hours, immediately bring up the white bin to MS 6XX, place it into the BetaStar autoclave and autoclave it using the BIOBAG40 protocol.  Make sure to sign the BetaStar autoclave log sheet with your name, lab, our room number (MN508), the time of the protocol and which protocol (BIOBAG40).  Also make sure to say "yes" on the biohazard column.
     4. If the autoclave is in use, put your name, lab, and room number on the next open line of the log sheet to "claim your place in line".   (Sometimes this doesn't help, but it doesn't hurt.)  Recheck the autoclave ~20 min after the running cycle is expected to finish.
     5. 60 minutes after beginning the BIOBAG40 cycle go back upstairs to remove the waste. Dump the waste into one of the provided trash bins in the room, then bring the empty bin back to MN508.
     6. Note that the bin may be HOT! consider bringing up one of the leather gloves from drawer 9B.
  7. Return OR scrubs to scrub machine.

1. **REFERENCES:**
   1. University of Kentucky Cardiovascular Biorepository IRB approved consent form
   2. UK Healthcare Policy and Procedure – Department of Perioperative Services -Dress Code (#OR 03-01)
   3. University of Kentucky HIPAA in Health Research (<http://www.research.uky.edu/ori/HIPAA/main%20page.htm>)
   4. UK Biosafety Manual (4.0-University of Kentucky Biological Safety Requirements, 5.3-Exposure Incidents, 10.0-Decontamination and Disposal of Biohazardous Waste)