

MOLECULAR DETERMINANTS CORE SAMPLE COLLECTION & SHIPPING GUIDELINES



GENERAL CONSIDERATIONS

Consistency in sample collection and handling is most important. Minimize operational variability wherever possible (e.g. collection technique, time of sampling...). Materials collected for experimental work other than metabolomics studies may be suitable for metabolomics analysis, depending on the collection procedure and consistency of treatment across all samples. **Notify us of any preservatives or additives that have been applied during the collection process.** Whenever possible avoid the use of any preservatives or additives.

- Minimum volumes/quantities required per requested analysis are listed below. Limiting volumes/quantities will limit the amounts of possible analysis. Lower sample volumes may be acceptable depending on the nature of your study. Contact us if you are considering a lower sample volume.

- The Molecular Determinants Core does not currently support certain samples (plants, bones, insects...). For sample types other than the ones listed below, please contact us.

TUBE TYPES & LABELLING

- Use polypropylene tubes, **cryovials** (up to 2ml), **Micronic tubes** (0.5-1.4ml push caps or screwcaps) or **centrifuge tubes** (Eppendorfs 1.5-2ml not 0.5ml) for sample storage/shipment.



- Please try to have **all samples in the same tube type** (not half of them in cryovials and half of them in Eppendorf).
- Tubes should **not be more than half full**.



- Samples in different tubes than the ones listed above, with inconsistencies of tubes type within samples or with too much volume will require samples transfer (**extra charge will apply**)

- All tubes must be **clearly labeled with unique sample identifiers**. Use permanent markers (e.g. Sharpie) to directly label tubes while warm; cold tubes will not hold the ink. Note that stick-on labels tend to fall off when frozen, so unless specialized freezer-safe labels are available, direct marking is preferred. Samples may be labeled with 2D barcodes, but please also include an identifier on the tube that can be read by eye.

FREEZING

- If pre-processing of sample is required prior to sending/freezing the samples, always keep samples and tubes at a maximum of 4°C during processing.

- Freeze samples as soon as practical. Flash-freezing of samples in liquid nitrogen, immediate placement of tubes in a -80°C freezer or dry ice/ethanol bath are ideal and recommended across all sample types. Note, however, that ethanol may wash off labeling.

- Store at -80°C until shipment.

PLASMA & SERUM

Minimum volumes: 100 µL per requested analysis.

Both serum and plasma are obtained from blood. Do not freeze blood before collecting plasma/serum. This would lead to massive sample hemolysis and contamination of serum/plasma with cellular metabolites/proteins. When collecting blood, it is also very important to treat all of the samples the same to avoid variation. (Same tubes, same anticoagulant...)

- Plasma: Collect whole blood in tubes containing anti-coagulant and follow tube manufacturer's processing instructions.

- Serum: Collect whole blood in serum separator tubes and follow tube manufacturer's processing instructions.

- Preferred anticoagulants: No additives (red caps) or EDTA (purple caps, K2, K3, avoid Na & Li). For Metabolomics studies, avoid Citrate (blue caps). For Lipidomics studies, avoid Heparin (greens caps).

- **Always notify us which anticoagulant was used**
- **Never use multiple/different anticoagulants within the same experiment/study.**



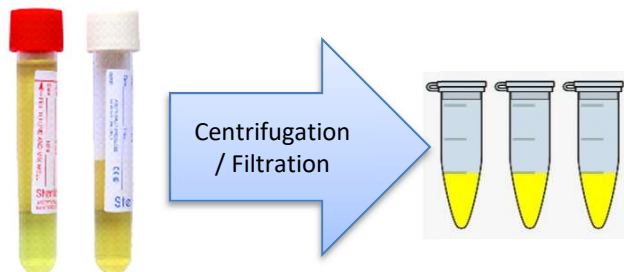
URINES

Minimum volume: 100 µL per requested analysis (+20uL if osmolality measurement needed for normalization).

24-hour whole urine collections kept at 4°C during collection period are highly recommended. If 24-hour collections are not practical or indicated, then first catch is preferred.

• Always notify us if preservatives were used during collection

• Following collection, measure volume and if possible, centrifuge to precipitate sediment (or filter) and transfer the supernatant into a clean tube taking care not to disturb the bottom of the tube whether there is a visible pellet or not. Discard the pellet.



• Aliquot the urine into chilled tubes. Freeze samples as soon as practical. Store at -80°C within one week. Store samples at -80°C until shipment.

SOFT TISSUES

Minimum weight: for collected solid tissues (e.g. biopsy material), the amount of tissue/sample can vary depending upon study objectives and tissue type. A minimum of 50-100 mg is recommended. Contact us if you are considering smaller amounts

• Special tube containing homogenizing beads can be provided on demand for your sample collection (recommended especially if sample is small, this will prevent sample loss during transfer from your tube to the tube of homogenizing beads we use for tissue grinding)

• Weight for each tissue sample must be provided for normalization. If not the MDC will determine sample weights but **extra charge will apply.**



CEREBROSPINAL FLUID & SALIVA

Minimum volumes: 100 µL per requested analysis (+20uL if osmolality measurement needed for normalization).

CELL CULTURES

Minimum quantities each sample should optimally contain or be extracted from 3 million cells, preferably 5 million cells. Cell size will heavily impact this requirement; (smaller cells require higher cell counts). Lower number of cell will drastically impact sensitivity.

• Cell count or protein determination will not be performed on individual samples by the MDC; hence each sample sent to the MDC should either contain the same amount of cells or an

accurate cell count/protein quantification for each sample for normalization purposes.

• Cultures of adherent cells may either be submitted as cell pellets, or as in-plate extractions, per individual project as advised in consultation with the MDC.

• If adherent cells are submitted as pellets, gentle trypsinization may result in a more reproducible yield of cells and reduced cell lysis.

• Cells must be thoroughly separated from media prior to being extracted or flash frozen and shipped to the Molecular Determinants Core. Triple washing with PBS is recommended.

• When removing media by centrifugation, avoid spinning conditions that will lyse the cells. You can save 200 µL cell culture media and several aliquots of fresh media for metabolomics analysis.

SAMPLE LIST

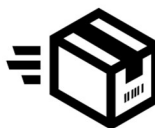
• Please include all available sample information when you submit the samples. Send an electronic document containing sample ID information, preferably in advance of sending the samples, and include a hard copy of the list of samples with the shipment.

• **You must NOT include personally identifying information for human samples.**

SHIPPING

• Clients located in the Baltimore area can deliver in person on the Baltimore campus from. Please contact our Client Lead Susan Aja to coordinate sample drop off.

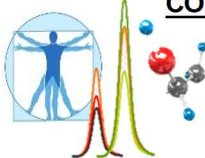
• Samples may be shipped directly to MDC for overnight delivery between Tuesday - Thursday (9 am - 4 pm). Please coordinate with the Client Lead before sending your samples.



Molecular Determinants Core (MDC)
Johns Hopkins All Children's Hospital
Research and Education Building
3rd Floor, Room 3400
600 5th Street South
Saint Petersburg, FL 33701

• All Samples should be shipped frozen overnight in an insulated container on 5-10 kg of dry ice. The MDC is located in Florida, assume ambient temperatures above 90°F April-October, so additional dry ice is strongly recommended. 5 kg is the recommended minimum amount of dry ice for domestic shipping. 10 kg is the recommended minimum amount of dry ice for international shipping. The MDC will not accept any responsibility for packages that are damaged or lost in transit.

• MDC recommends insuring the package for the full value of experiment costs incurred to generate of the samples.



CONTACT INFORMATION

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On the Web: <https://www.hopkinsallchildrens.org/cribmdc>