

PMT3-E-2 — SPECIMEN PROCESSING AND SAMPLE MANIFEST FOR THE PMT3-STUDY

Specimens

There are 3 different specimen types associated with PMT3 study:

1. Li-Heparin blood plasma free metanephrines and catecholamines
2. 24-h-Urinary Collection-containers for urinary metanephrines and catecholamines
3. EDTA blood for DNA

Unique identifiers must be used for all specimens (see SOP PMT3-C-1), which are provided in the sample log and are affixed as sticky labels.

1. Processing of blood for plasma metanephrines and catecholamines

Blood for plasma free metanephrines and catecholamines should be kept on ice or ice packs after collection and should be centrifuged immediately to separate the plasma. Plasma should be divided into at least four specimens of at least 1.2 mL each, labeled with the unique identifier sticker that includes the handwritten date of collection, and frozen (-80°C) or assayed immediately.

2. Processing of urine collections for metanephrines and catecholamines

Urine samples collected under the protocol for metanephrines and catecholamines should be processed as complete 24-h-urinary collections. All collection boxes are immediately placed on ice once they are delivered. The urine samples should generally arrive as a single collection with a single start time and end time label, for which aliquoting of urine is simplified. Start and end times of collections should be noted and any discrepancies reported in the single sample receipt log.

Urine volume should be measured. Volume and dates of collections are recorded in the inventory log and on the unique patient ID labels. Three aliquots of 2 to 2.5 mL are stored at -80°C in the appropriately designated storage box.

3. Processing of serum samples for DNA

EDTA blood samples for DNA testing are collected and stored pseudonymized in coded tubes without any other attached information that would allow identification of the individual from whom the sample is collected. The samples should not be centrifuged, have serum extracted, or have anything further done to it. The samples should be kept frozen (-80°C) and ideally should be shipped immediately.

Genes to be tested include exons 10, 11, 13 and 16 of the RET proto-oncogene and all exons of VHL (exons 1-3), SDHB (exons 1-8), SDHD(exons 1-4) and TMEM127 (exons 1-4) genes. Large deletions of SDHD, SDHB and TMEM127 genes will additionally be evaluated by quantitative multiplex PCR of short fluorescent fragments (QMPSF). Multiplex-ligation-dependent probe amplification (MLPA) is carried out for the VHL genes.

4. The Sample Manifest

When the samples are collected, the biomaterial records should be described on Sample Manifest (F1. PMT3_Sample-Manifest-Template). The required biomaterials are the date of each samples are collected, the number of aliquots of each samples, and 24hr urine volume. Failure to collect those data or biomaterials, or samples without appropriately completed Manifest, might invalidate use of the data/samples for the study and thus potential for authorship. An electronic copy should be included or sent separately.