

Guidance for Completing your NBB Tissue Request

Planning Your Tissue Request

- If you are not sure precisely what tissue, tissue regions, or quantities are best for your research project, please submit a request for help to the [NIH NeuroBioBank email address](#) before completing the on-line request.
- The Request Form requires you to provide a **detailed** rationale for 1) number of subjects requested, 2) amount of tissue requested per subject, and 3) number of brain regions requested. Please see the Acceptable Tissue Amount Guidelines (below) to plan your request.
- If you have not performed a pilot study in human tissue, your first request should be for tissue to perform a pilot study using your proposed methods.

Completing Your NBB Tissue Request Form

1. Complete all applicable fields. Required fields must be completed in order for the system to accept the request. Incomplete information will delay the review of your request, especially Specimen Shipping Information.
2. **Request Name** should be a short unique descriptor of your request.
3. **Requested Specimens** lists all of the specimens you selected for your study (using the specimen search). This list must contain all the specimens you are interested in obtaining.
 - a. For each specimen listed, you must enter the amount of each specimen needed for your study using the “Amount Requested” field. Please follow the Acceptable Tissue Amount Guidelines (below) when submitting your request. A detailed rationale must be provided for any deviations from these guidelines.
 - b. “Unaffected Control” are subjects with no known psychiatric or neurological diagnoses.
 - c. If you wish to adjust your list of selected specimens, click the “Edit Requested Specimen(s)” option. The specimen search will appear. You can add/remove specimens. Once you are finished updating your selection, click the “Return to Request” option at the top of the search. The request form will be updated with your new specimen list.
4. **Request Details**
 - a. The “Title of Research Plan” will appear at the top of your MTA.
 - b. Concisely describe the request according to the instructions. If you are requesting multiple anatomical regions per subject, the rationale must be clearly indicated here.
 - c. List each type of assay or method to be used with the requested tissue. Include the amount of tissue needed for each assay/method per subject.
 - i Example: “qPCR, 2mg grey matter per subject.”
 - d. Indicate whether you or your direct collaborators have used the method(s) proposed in this request with human postmortem tissue before.

- i If the answer is “No,” then this request should be for tissue for pilot studies.
- e. Provide a rationale for the number of subjects requested. Acceptable rationales include power analyses, or accepted standards in the field (with appropriate citations).

Determining Your Tissue Needs

The table below provides guidelines for generally acceptable amounts of tissue that can be requested for different types of studies. As different brain banks process and store their samples differently (see SOPs), you may be supplied with more tissue than you request. However, each of the tissue amounts below will provide sufficient material to perform each type of methodology listed. These values were derived in collaboration with researchers who have successfully performed each methodology using human brain tissue.

If your methodology is not listed below, please include a detailed rationale supporting the amount of tissue you are requesting.

If your methodology is below, but you require more tissue than is indicated, please include a detailed rationale supporting your request outside of the acceptable range.

Please note that some brain regions, such as the substantia nigra, hippocampal subfields and thalamic and hypothalamic nuclei, are small, and many investigators are interested in studying these regions. As such, for certain methodologies, the NBB may indicate that a tissue amount which differs from the below guidelines will be provided.

If you need guidance on any issues related to tissue amount guidelines, please contact the NIH NeuroBioBank at neurobiobank@mail.nih.gov.

Method	Accepted Tissue Amount/Region/Subject
FACS sorting with NeuN	≤ 100mg of gray matter
HITS-CLIP	≤ 300mg
Immunoblot	≤ 10mg of tissue per protein examined
Immunohistochemistry	Sufficient tissue for ≤ 6 sections per immunolabeling <i>(May be provided as free floating sections, formalin-fixed paraffin-embedded sections, fixed tissue blocks, or paraffin-embedded tissue blocks dependent upon region requested and brain bank)</i>
Mass Spec:	
Quantitative Mass Spec (DIA, DDA, TMT) with no enrichment	≤ 5mg
Mass Spec with sucrose density gradient based sub-cellular enrichment	≤ 5 - 350 mg depending on specific protocol
Co-IP with Mass Spec	5 - 20 mg
Mass Spec for post-translational modifications	5 - 20 mg
Microarray	≤ 5mg
qPCR	≤ 5mg

Method	Accepted Tissue Amount/Region/Subject
RNASeq	
Total gray or white matter	≤ 20mg
Single nucleus RNASeq (snRNASeq)	≤ 35mg
Single nucleus Assay for Transposase-Accessible Chromatin RNASeq (snATAC RNASeq)	≤40 mg
Slide-Based Assays (e.g., <i>in situ</i> hybridization, laser-capture microdissection)	Sufficient tissue for ≤ 6 sections per condition

Note: If an investigator has not tested the proposed methods in post-mortem human tissue, a pilot study will need to be conducted first for the optimization of methods. The Neurobiobank can provide tissue suitable for such methods piloting.