1. Is fasting required for the blood draw?
   Fasting is not required for the blood draw at any of the ARTFL-LEFFTDS visits. However, we understand that some patients may be asked to also donate CSF, which would ideally include an 8 hour fast. The collection form for both blood and CSF include a question for information about when the patient ate their last meal. This will allow us to account for both fasted and nonfasted samples.

2. CSF should be collected, preferably fasted. Is that an 8 hour fast? Longer?
   An 8 hour fast is ideal, but please collect CSF even if the patient has not fasted. Please record the last time a meal was eaten by the subject.

3. What is the difference between the gravitational and pull methods of extracting CSF?
   The gravitational method of CSF extraction does not utilize the 6mL luer lock syringes that are provided within the LP Trays supplied by NCRAD. The fluid is simply collected directly into the 50 mL conical tube provided in the kit. Both of these methods have been approved for these research studies.

4. What are the different types of needles provided by NCRAD for the CSF draw?
   There are two different needle types provided by NCRAD, and a BD Spinal Needle Introducer will accompany each kind. The Supplemental Kit contains a BD Whitacre Needle 24G, 0.55 x 90mm; whereas the LP tray already comes equipped with a Sprotte needle, 24G x 90mm. Please see the below depiction of needle types for a CSF draw.
5. What is the time frame for completing all the lab work (CSF and blood draws) if the study visit has to be done over several days? We understand that an ARTFL or LEFFTDS study visit may take place over the course of several days. It is completely acceptable to draw samples anytime across this time frame. However, please keep in mind that the sodium heparin tube cannot be drawn on a Friday. For example, if a patient is seen for a Cycle 1, Visit 1 of the LEFFTDS study and the patient is seen on Thursday and Friday, please draw the sodium heparin sample on Thursday and immediately send the sample ambient to NCRAD. Other samples for that subject may be drawn on Friday, frozen per the protocol, and sent with the next batch shipment to NCRAD.

6. Within the CSF form, there are questions asking for the total number of CSF aliquots and amount of CSF collected, should we include the volume of CSF that was sent to the local lab or those aliquots kept for the local PI? Within the CSF Sample and Shipment Notification Form, please only include information for the CSF aliquots that are being delivered to NCRAD.
7. Within the Biological Sample and CSF shipment notification forms, there is a question asking for the specimen number of the residual aliquot. What is this and where do I find this information? The residual aliquot is any aliquot containing a volume less than 0.5 mLs. NCRAD asks that the blood processing team document which aliquot contains a smaller volume by indicating the sample number located on the label. The sample number, pictured below, is preprinted on the aliquot labels provided by NCRAD.

![Sample Number Label](image)

8. Would it be possible to centrifuge the CSF samples at 4ºC (instead of room temperature) since the aliquot tubes are pre-chilled and the CSF samples are frozen immediately after centrifugation? Located in Section 7.4, Step 6 in the Manual of Procedures. Centrifuging the samples at 4ºC within this step has been approved.

9. A subject who will be enrolled in both ARTFL & LEFFTDS will only have to have the sample drawn once. In this scenario, subjects who have already been baselined in ARTFL and later enrolled in LEFFTDS would only need to have the PAXgene™ and sodium heparin tube drawn. How do we then link the samples back to
the EDTA that was drawn for ARTFL? Do we use the same RAVE ID? Or will there be a different RAVE ID per study per subject?
Each patient will only be assigned one RAVE ID between the ARTFL and LEFFTDS studies. Linking the patient between the two studies will take place in the Medidata Rave database system.

10. What FedEx account number should be used for sending shipments of specimens to NCRAD?
Sites sending shipments of samples back to NCRAD should use their site FedEx account number. Please also include any information within the “Reference” section that is required by your institution for tracking. The FedEx return label provided in your kits will already indicate that the sender will be providing payment.

11. Will there be someone available at NCRAD to receive Saturday Deliveries?
NCRAD is unable to accept Saturday deliveries. Therefore, the sodium heparin tube for PBMC’s should not be drawn on Friday. If a LEFFTDS patient is scheduled for their Cycle 1, Visit on a Friday – please make sure that this visit spans more than one day and draw the PBMC sample either on Thursday or Monday. Be sure to send the collected PBMC sample the day it is drawn.