

SAMPLE TYPE, EXTRACTION CONDITIONS, STORAGE AND SHIPMENT TO THE BIOBANK

These are the guidelines to follow for the donation of samples. The health professional or researcher must notify the CIBERER Biobank (CBK) of the intention to send one or more samples from patients with rare diseases to the Biobank.

Please inform the Biobank staff as far in advance as possible by calling Tel. +34 96 192 63 21 from 8:00 a.m. to 15:00 p.m. Monday through Friday or by sending an email to info-biobank@ciberer.es.

Samples identification

Tubes labeled with a unique identification code and type of sample that it contains, anatomical place of the samples, date and time of collection.

Externally identified packages with the address of the sending Hospital or Laboratory and that of the recipient:

FISABIO-Salud Pública
CIBERER Biobank
Av. Cataluña, 21 - 2nd floor
46020 Valencia
Contact: Dr. Carmen Aguado and Dr. Salvador Martí

The **conditions for obtaining, preserving and sending samples** are detailed below:

SAMPLES SENDING CONDITIONS

The samples must be sent in suitable containers, complying with current legislation on the transport of potentially infectious biological material. The shipment must be made from Monday to Thursday by express courier service with delivery before 10 am the next day to ensure that the samples can be processed the same day they are received.

Peripheral blood samples

Sample processing performance will depend on the number of leukocytes in blood sample, the time elapsed after blood collection, and storage and shipping conditions.

- Plasma collection from peripheral blood: the blood must be collected in tubes with EDTA, citrate or lithium heparin as anticoagulant. After this, keep the sample at room

temperature if it is going to be processed in approximately 30 min or send the samples the same day of the extraction in a vertical position between 4-8 °C, always keep the samples between 4-8 °C until processing.

- Serum collection from peripheral blood: the blood must be collected in blood collection tubes with gel and coagulation activating particles as an additive. There are different tube formats from 5 to 8.5 ml (Vacutainer, Vacuette, etc.). After collecting the sample, it is necessary to gently invert the tube to favour coagulation by putting the blood in contact with the activating particles (4 times maximum). Then, keep the tube in a vertical position. Send the samples the same day of the collection in an upright position and between 4-8 °C, always keep the samples between 4-8 °C until processing.
- Extraction of DNA/RNA from peripheral blood: the blood must be collected with EDTA as an anticoagulant. Send the samples the same day of the extraction at room temperature or at 4-8 °C. If the shipment is to be made in the days after the sample is collected, it must be stored at 4-8 °C until shipment.
- Isolation of mononuclear cells from peripheral blood: blood collection must be performed using ACD or lithium heparin as anticoagulant. The samples must be stored at room temperature and that no more than 24 hours elapsed since their collection until the moment they are received at the CBK. If it is not possible to send it within the first 24 hours, the sample must be stored at room temperature until shipment.
- Immortalization of B lymphocytes: blood collection must be performed using ACD or lithium heparin as anticoagulant. The samples must be stored at room temperature and that no more than 24 hours elapsed since their extraction until the moment they are received at the CBK. If it is not possible to send it within the first 24 hours, the sample must be stored at room temperature until shipment.
- Plasma/Serum Samples: peripheral blood is collected in tubes specifically designed for each sample. After plasma/serum collection, the sample can be kept at 4°C (refrigerated) or 22°C (room temperature) until processing and storage (freezing at -80°C) within 1 to 2 hours of collection. Samples are shipped on dry ice to maintain a temperature of -80°C.

Cell lines

- Cell immortalization: cell lines must be maintained/stored in liquid N₂ (-180 °C). The shipment must be made in dry ice with a sufficient quantity to ensure temperature of -80 °C until the moment of receiving it at the CBK. The amount of dry ice will depend on the type of service contracted for its transport.

Tissue Samples

- Skin biopsy for fibroblast culture: the tissue sample will be placed in a sterile container with DMEM culture medium, 10% fetal bovine serum, 2 mM Glutamine and 1% antibiotic (Penicillin/Streptomycin). It must be sent via express service, as soon as possible, at a 4-8 °C. If it is not possible to send it in the first 24 hours, the sample must be stored at 4-8 °C until shipment. Once received, the Biobank staff will proceed to culture it according to the standardized protocol for it.
- Muscle biopsy for myoblast culture: the tissue sample will be placed in a sterile container with DMEM culture medium, 10% fetal bovine serum, 2mM Glutamine and 1% antibiotic (Penicillin/Streptomycin). It must be sent via express service, as soon as possible, at a 4-8 °C. If it is not possible to send it in the first 24 hours, the sample must be stored at 4-8 °C until shipment. Once received, the Biobank staff will proceed to culture it according to the standardized protocol for it.

Urine samples

- Collect urine in sterile flasks/tubes. Once the urine is collected, it must be sent by express service, as soon as possible, at 4-8 °C. If it is not possible to send it in the first 24 hours, the sample must be stored at 4-8 °C until shipment.

Cerebrospinal fluid

- The cerebrospinal fluid (CSF) is collected in the specific tubes for it: sterile 12 ml tubes with screw cap and without any type of additive. After collecting the sample, it can be kept between at 4 °C in a refrigerator or at 22 °C (room temperature) until processing and storage. There is no current evidence that demonstrates a preference in the temperature at which the sample must be kept prior to processing. Keep the tube in an upright position during transport, as the tube seal is not airtight. It is recommended that the time between CSF extraction and freezing at -80 °C should be within the range of 1 to 2 h after collection.

Stool samples

- Sterile, wide-mouth and screw-capped polypropylene container. Freshly released stool sample must be collected. Avoid contact with water or urine. The volume of the sample should be approximately the size of a walnut. Identify the container with the date, time of collection and a with a donor code. The sample should be kept refrigerated at 4-8 °C until shipment to the biobank. Recommended delivery time is less than or equal to 2 h and no more than 24 h.

SAMPLE ACCEPTANCE CONDITIONS

The samples will be inspected upon arrival to verify their adequacy in terms of the requirements specified in this document. The requested test will not be carried out when the integrity of these has not been preserved until the laboratory, or when there is any possible

circumstance that makes it impossible to correctly carry out the tests and/or the delivery of results.

In other cases, and in general, due to the lack of compliance with the requirements established in this document, as well as the lack of associated information (donation form, service request form, informed consent, etc.), the Biobank staff will contact the user to correct the deficiency or, where appropriate, request a new sample.

The most common causes of sample rejection are the following:

- Unidentified patient (Name/Identification number)
- Inappropriate sample for the required study
- Day and time of sample collection not specified
- Sample origin not specified
- Illegible or incorrectly completed request form
- Fresh tissue samples frozen
- Samples not sent in the required conditions
- Broken tubes/containers
- Evident external contamination
- Coagulated peripheral blood
- Insufficient volume. If there is not enough for several requests, the clinician will be asked to specify their priority.

Likewise, when the quality of the sample not be suitable, the CIBERER Biobank staff will fill in an incident report and the applicant will be informed of this fact. In addition, when appropriate, the applicant will be informed of the extent to which these incidents may have affected the final result.