

Bone Marrow Aspirate

For Research or For Further Processing Use

Product Description

The bone marrow is the site of new blood cell production. It is rich in hematopoietic progenitor cells and mesenchymal stromal cells, as well as newly developed white blood cell and red blood cell populations such as nucleated red blood cells. Many cell types can be isolated from collected bone marrow for use in research and clinical therapeutic applications. Bone marrow is collected from living or cadaveric donors, based on request, via repeated iliac crest puncture into ACD or heparin anticoagulant.

Product Source

Bone marrow is collected from one side of the iliac crest using a 11G bone marrow biopsy needle. Collection occurs in a licensed Operating Room using conscious sedation anesthesia. Up to 200mL of bone marrow is collected aseptically into syringes containing sodium heparin. A sodium dextrose solution anti-coagulant may be substitute upon request.

Product Processing

All product is collected to order and therefore be processed using custom specifications. After collection, marrow filled syringes are transported for processing using sterile technique under a certified laminar flow hood. All syringes are pooled into a single transfer bag and well mixed. Filtration is available upon request. Processed bone marrow is aliquoted into 25mL, 50mL or 100mL volumes. Any combination of the aliquots maybe ordered, including ordering two 100mL aliquots.

Product Testing

Donors are tested for relevant communicable diseases as provided in the current version of the US Code of Federal Regulations. Additional testing can be performed to meet EU GMP requirement. Sterility testing is performed on a pooled product sample.

Product Storage

If product is cryopreserved, store at -80°C or colder until use. Use immediately upon thaw.

Product that is not cryopreserved should be stored at ambient or refrigerated temperature for up to 24 hours.

Recommended Purification of Mononuclear Cells from Bone Marrow Aspirate

Materials

Room temperature PBS without Mg^{2+}/Ca^{2+}
Room temperature Ficoll-Paque Density Gradient
Wash buffer (PBS + 5mM EDTA + 2% human serum)
50 mL conical tubes
Serological pipets
Swing bucket, tabletop centrifuge
Media specific for desired downstream research applications
Hemocytometer or automated cell counter

Protocol

Bone marrow aspirate is sterile upon receipt. To maintain sterility, perform all harvesting steps inside a biological safety cabinet, practice sterile technique, and use only sterile supplies and media.

1. Remove product from bag and place into 50 mL conical tubes, 25 mL/tube maximum. Warm product to room temperature if stored at 4°C.

2. Dilute product 1:1 with PBS by doubling the volume of bone marrow in each tube.
3. Place 15 mL of Ficoll into clean 50 mL conical tubes, one each for 35 mL of diluted product you wish to process.
4. Carefully layer 35 mL of the diluted buffy coat on top of the Ficoll layer, taking extreme care to not break the plane of the Ficoll and cause mixing of the two components.
5. Centrifuge set to 2200 RCF, 20 minutes, 20°C with break OFF.
 - a. If temp is too cold or too hot, separation of mononuclear and polymorphonuclear cells will not occur properly.
 - b. If the break is left on, mixing will occur between the Ficoll and mononuclear cells when the centrifuge stops.
6. Carefully remove the tubes from the centrifuge. Using a serological pipet, remove the white interface between the Ficoll and media/plasma layers. Transfer the interface to a clean 50 mL tube. Use 1 tube for each Ficoll tube used.
7. Wash the cells by filling the tubes with Wash Buffer and centrifuging for 10 minutes at 1500 RCF, 20 °C, break ON. Repeat wash.
8. Pool cells into a single tube and wash one more time by filling the tube with PBS and centrifuging.
9. Resuspend cells in desired media. Remove a sample and count the enriched mononuclear cells.

Warning

This product is composed of human-derived materials. Always wear appropriate personal protective equipment when handling this product and treat it as potentially infectious, using Universal Precautions, regardless of the results of infectious disease testing.

Limitations and Publications

This product is for research use or therapeutic use only, not for resale. Nothing produced directly from this product may be sold. When publishing scientific results obtained using this product, acknowledge supplier as Bio-Sharing.org.