Nucleic Acid BCT™

STRECK

INSTRUCTIONS FOR USE

Nucleic Acid BCT™ is a direct draw whole blood collection tube intended for the stabilization of draw time concentrations of cell-free DNA, cell-free RNA, and extracellular vesicles. **This product has not been cleared by the U.S. Food and Drug Administration for In Vitro Diagnostic Use. The product is For Research Use Only. Not for use in diagnostic procedures.**

SUMMARY AND PRINCIPLES

Accurate analysis of plasma analytes, such as cell-free DNA, extracellular vesicles (EVs), or EV-associated cell-free RNA can be compromised by delayed blood sample processing, handling, and shipping. In all cases, changes to plasma analyte concentrations may occur through deterioration and breakdown of blood cells.

The preservative reagent contained in Nucleic Acid BCT stabilizes both nucleated blood cells and erythrocytes thus preventing the extraneous release of cellular materials, such as genomic DNA from white blood cells (WBC) or EV-associated RNA from WBCs and immature erythrocytes. Samples collected in Nucleic Acid BCT are stable for up to 7 days at ambient temperature, allowing convenient sample collection, transport, and storage.

REAGENTS

Nucleic Acid BCT contains an anticoagulant and proprietary preservatives in a liquid medium.

PRECAUTIONS

1. **For Research Use Only. Not for use in diagnostic procedures**.
2. Do not freeze specimens in glass Nucleic Acid BCT as breakage could result.
3. Do not draw whole blood into Nucleic Acid BCT past the expiration date printed on label. If sample collection occurs on or before the expiration date printed on the label, whole blood collected into Nucleic Acid BCT should be stored at 18 °C to 25 °C for up to 7 days prior to processing to plasma.
4. Do not use tubes for collection of materials to be injected into patients.
5. Hemolysis immediately after draw can be a sign of improper collection technique and the tube should be discarded and redrawn.
6. Product is intended for use as supplied. Do not dilute or add other components to Nucleic Acid BCT.
7. Overfilling or underfilling of tubes will result in an incorrect blood-to-additive ratio and may lead to incorrect analytic results or poor product performance.
   CAUTION
   a. Glass has the potential for breakage; precautionary measures should be taken during handling.
   b. All biological specimens and materials coming in contact with them are considered biohazards and should be treated as if capable of transmitting infection. Dispose of in accordance with federal, state and local regulations. Avoid contact with skin and mucous membranes.
   c. Unused tubes should be disposed with infectious medical waste.
   d. Remove and reinsert stopper by either gently rocking the stopper from side to side or by grasping with a simultaneous twisting and pulling action. A "thumb roll" procedure for stopper removal is NOT recommended as tube breakage and injury may result.
8. SDS can be obtained at streck.com or by calling 800-843-0912.

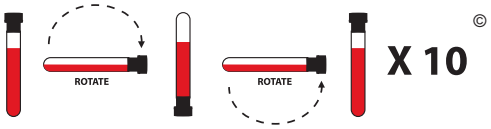
STORAGE AND STABILITY

1. When stored at 2 °C to 30 °C, unfilled Nucleic Acid BCT is stable through expiration date.
2. Blood samples collected in Nucleic Acid BCT are stable for up to 7 days when stored at room temperature.
3. Do not freeze unfilled Nucleic Acid BCT.
4. Ship tubes filled with blood in coolers equilibrated to room temperature to limit exposure to temperature extremes.

INSTRUCTIONS FOR USE

For a video demonstration, visit streck.com/mixing.

1. Collect specimen by venipuncture according to CLSI PRE02[1].
   Prevention of Backflow - Since Nucleic Acid BCT contains chemical additives, it is important to avoid possible backflow from the tube.
   To guard against backflow, observe the following precautions:
   a. Keep patient's arm in the downward position during the collection procedure.
   b. Hold the tube with the stopper in the uppermost position so that the tube contents do not touch the stopper or the end of the needle during sample collection.
   c. Release tourniquet once blood starts to flow in the tube, or within 2 minutes of application.
2. Follow recommendations for order of draw outlined in CLSI PRE02[1]. Nucleic Acid BCT can be drawn after the EDTA tube and before the fluoride oxalate (glycolytic inhibitor) tube. If a Nucleic Acid BCT tube immediately follows a heparin tube in the draw order, Streck recommends collecting a non-additive or EDTA tube as a waste tube prior to collection in the Nucleic Acid BCT.
3. Fill tube completely.
4. Remove tube from adapter and immediately mix by gentle inversion 10 times. Inadequate or delayed mixing may result in incorrect analytical results or poor product performance. One inversion is a complete turn of the wrist, 180 degrees, and back per the figure below:



5. After collection, transport and store tubes within the recommended temperature range.

Note:

1. For best results, a 21G or 22G needle is advised. Slower fill times may be observed when using a smaller gauge needle.
2. When using a winged (butterfly) collection set for venipuncture and the Nucleic Acid BCT is the first tube drawn, a non-additive or EDTA discard tube should be partially drawn first in order to eliminate air or "dead space" from the tubing.
3. As in the case with most clinical laboratory specimens, hemolysis, icterus and lipemia may affect the results obtained on blood samples preserved with Nucleic Acid BCT.

PLASMA ISOLATION

Step 1. To separate plasma, centrifuge whole blood at 1800 x g for 15 minutes at room temperature.
Step 2. Remove the upper plasma layer and transfer to a new conical tube (not provided).
Step 3. Centrifuge the plasma at 2800 x g for 15 minutes at room temperature .
Note: Nucleic Acid BCT has been validated for a maximum centrifugation of 3,000 x g for 10 minutes. Exceeding these limits may result in breakage.

DNA EXTRACTION

Extraction of cell-free plasma DNA can be accomplished using most commercially available kits that include a Proteinase K treatment step.
For optimal results, include a Proteinase K treatment step (≥ 30 mAU/mL digest) at 60 °C in the presence of chaotropic salts for 1 hour when extracting cell-free DNA.

CELL-FREE RNA EXTRACTION

Extraction of cell-free RNA can be accomplished using the following protocol and kits. Other protocols and kits require validation from the end user.

The Nucleic Acid BCT is compatible with the following commercially available nucleic acid isolation kits when used according to the manufacturer's instructions for use: QIAamp® Circulating Nucleic Acid Kit (Qiagen), MagMAX™ Cell-Free Total Nucleic Acid Isolation Kit (ThermoFisher), and Plasma/Serum Circulating and Exosomal RNA Purification Kit (Slurry Format, Norgen). A DNase1 digest step is advised to deplete contaminating genomic or cell-free DNA.
Note: When using the QIAamp Circulating Nucleic Acid Kit, the provided plasma protocol was utilized with extension of the 60 °C incubation time from 30 to 60 minutes.

EXTRACELLULAR VESICLES/EXOSOME ISOLATION:

Isolation of extracellular vesicles can be accomplished using filter-based (Qiagen exoEasy), size-exclusion-based (Cell Guidance Systems Exo-spin™), or precipitation-based (Thermo-Fisher Total Exosome Isolation Kit) methods.

FREEZING AND THAWING PLASMA

1. To Freeze: For long-term storage, after the second spin, collect and transfer the plasma to a cryogenic tube (not provided) and freeze at -20 °C or -80 °C.
2. To Thaw: Thaw cryogenic tubes at appropriate temperature as specified in your protocol.

LIMITATIONS

1. For single use only.
2. Tube is designed for direct draw with a standard needle holder and single use collection. Collection using other means, such as a syringe, or collection and transfer from other devices is not advised.
3. Specimen transport via pneumatic tube system is not advised.
4. Organic phase extraction methods, such as phenol-chloroform, are not advised.
5. Exosomes isolated from Nucleic Acid BCT may no longer be suitable for functional studies.

REFERENCES

1. Clinical and Laboratory Standards Institute, PRE02, Collection of diagnostic venous blood specimens. Approved Standard – Eighth Edition.
2. ISO 6710, Single-use containers for human venous blood specimen collection.

ORDERING INFORMATION

Please call our Customer Service Department at 800-228-6090 for assistance. Additional information can be found online at streck.com.

TECHNICAL SUPPORT

Please call Streck Technical Services at 800-843-0912 for assistance. Additional information can be found online at streck.com.

GLOSSARY OF SYMBOLS

See the Instructions (IFU) tab under Resources on the product page at streck.com.

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