Revolutionising Liquid Biopsy

Unveiling Nucleic Acid BCT Tubes

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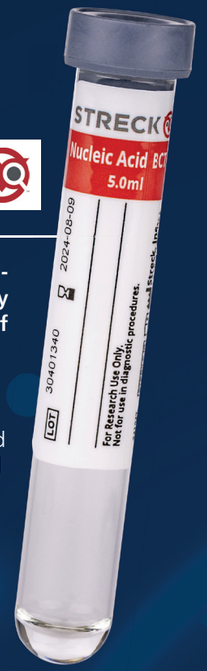


Human blood is a treasure trove of genetic information, hosting crucial details within circulating cell-free DNA (cfDNA) and cell-free RNA (cfRNA). Leveraging the simplicity of a blood draw, liquid biopsy emerges as a powerful tool, unravelling insights from cancer predisposition to the aftermath of traumatic brain injuries.

The Crucial Role of Draw-Time Analysis

In the realm of liquid biopsy, timing is everything. Diseases linked to elevated circulating cfDNA and cfRNA demand precise analysis, necessitating a choice: immediate evaluation or the adoption of a stabilisation tube. Stabilising blood samples, ensures an accurate reflection of what is in the patient's circulation. Without stabilisation, samples deteriorate, releasing genomic DNA (gDNA), extracellular vesicles (EVs), and EV-associated cfRNA due to white and red blood cell breakdown. This degradation introduces non-specific increases in analytes, clouding the original sample's analysis.

Introducing Nucleic Acid BCT

While traditional storage methods, like EDTA tubes, offer brief sample integrity, they fall short in long-term room temperature storage.

The new Nucleic Acid BCT™ (NA-BCT) from Streck, is a novel blood collection tube. It is a game-changer in preserving nucleic acid concentrations, maintaining cfDNA, cfRNA and EV particle levels at draw-time concentrations for up to 7 days at room temperature. This empowers laboratories to shift focus from degradation concerns to data analysis.

Limit Haemolysis

Whereas non-stabilising tubes, such as EDTA, or even current tubes intended for cfDNA usage, suffer from storage time-dependent increases in haemolysis and related decreases in recoverable plasma volume, NA-BCT is designed to limit both [Figure 1].

This is accomplished via an optimised stabilisation solution that maintains the integrity of erythrocytes and WBCs. Blood samples stabilised in NA-BCT have decreased haemolysis compared to equivalent samples stored in other stabilisation tubes or EDTA and better maintain draw-time plasma volume during room temperature sample storage (Figure 1 B, C). This is critical for those assays containing a plasma volume requirement for their analyte extraction workflows. At the same time, retention of draw-time plasma volume directly results in increased extractable analyte yield (e.g., cfDNA yield).

Maintain Draw-time Plasma cfDNA Levels

Total nucleic acid was isolated from plasma using the QIAamp® Circulating Nucleic Acid Kit (Qiagen) according to the manufacturer's "3 mL Plasma" protocol, with the exception that the 60 °C incubation was extended to 60 minutes (Figure 2A). Resultant cfDNA concentration was measured using the Qubit™ dsDNA HS Assay according to kit-included instructions (Thermo-Fisher). Sample quality was assayed using Cell-Free DNA ScreenTape analysis following the manufacturer's protocol (Agilent Technologies). While blood collected into non-stabilising EDTA tubes demonstrates robust time-dependent increases in plasma DNA levels, equivalent donor samples collected into the NA-BCT maintain draw-time plasma cfDNA concentration for up to 7 days when stored at room temperature (Figure 2B). Further, Day-7 cfDNA size and calculated %cfDNA remain similar to draw time for blood collected into Nucleic Acid BCT™, but not donor-matched EDTA [Figure 2 C, D].

Together, these data demonstrate that NA-BCT maintains the draw-time characteristics and concentration of plasma cfDNA for up to 7 days of ambient storage.

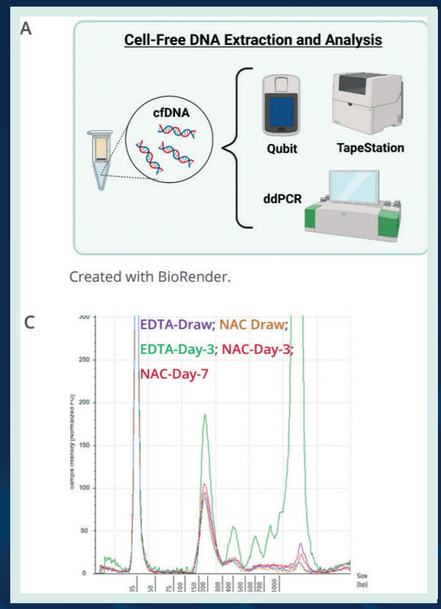


Figure 2. Plasma cfDNA levels are stabilised by Nucleic Acid BCT™ for up to 7 days of ambient storage. (A) Workflow of cfDNA collection and analysis. Cell-free DNA isolated from plasma was assayed for concentration, size and purity using a combination of Qubit™ dsDNA HS Assay, Cell-Free DNA ScreenTape analysis, and Bio-Rad ddPCR. Cell-free DNA concentration (B), size (C), and purity (D) in plasma collected into EDTA or Nucleic Acid BCT™ immediately after draw (Draw) or after 3 (Day-3) or 7 days (Day-7) of ambient temperature storage. [DNA] and %cfDNA are graphed as mean ± STDEV for 6 donors. The single donor electropherogram shown in (C) is representative of what is normally observed for all donors.

Maintain draw-time EV concentration

When samples were collected into NA-BCT, draw-time EV concentration was maintained for up to 7 days at ambient temperature storage. In contrast, EV concentrations increased markedly at days 3 and 7 for equivalent samples collected in EDTA (Figure 3).

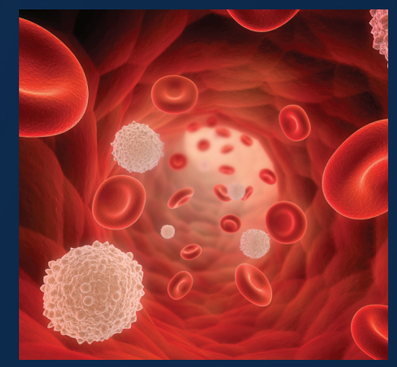
Maintain draw-time cfRNA concentration

To qualitatively compare cfRNA between samples collected in EDTA or NA-BCT, the Bioanalyzer RNA Pico Assay was used per kit included instructions (Agilent). Samples collected into the Nucleic Acid BCT maintained cfRNA concentration for up to 7 days, whereas cfRNA levels increased substantially with time for samples collected into EDTA (Figure 4). Further, samples collected in the Nucleic Acid BCT maintained a draw-time cfRNA size profile for up to 7 days of ambient temperature storage. These data demonstrate that Nucleic Acid BCTs effectively maintain draw-time cfRNA concentration during blood sample storage.

Streck Nucleic Acid BCT stabilises all plasma nucleic acids (cfDNA and cfRNA) and extracellular vesicles for up to 7 days at room temperature. This tube is ideal for labs seeking to reduce haemolysis in their samples, maximise plasma yield, and maintain draw-time concentrations of cfDNA, extracellular vesicles, and cfRNA. NA-BCT is a powerful addition to the liquid biopsy toolkit allowing for combined interrogation of both cfDNA (mutation profiling) and cfRNA (transcriptome analysis and expressed fusion gene detection) in addition to downstream analysis of EVs.

However, the advantages extend far beyond precise data. NA-BCT liberates laboratories from cold storage constraints, slashing shipping and handling costs.

It is perfect for groups relying on off-site laboratories, it extends storage time without compromising analysis. Moreover, it amplifies laboratory efficiency, allowing batches of samples to be processed weekly, contrasting with the immediate attention demanded by traditional tubes.



For more information please visit:
www.alphalabs.co.uk/NABCT

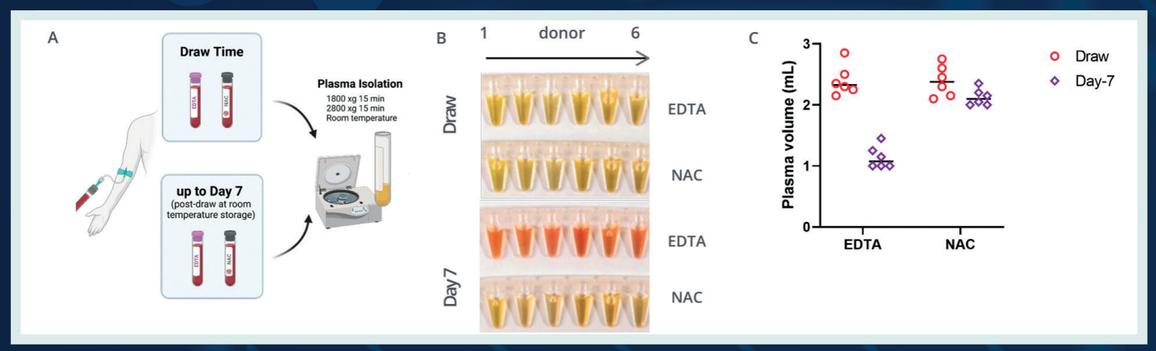


Figure 1. Nucleic Acid BCT™ (NA-BCT) maintains draw-time plasma characteristics. (A) Blood was collected from self-declared healthy donors into EDTA or NA-BCT. Plasma was isolated immediately after draw (Draw) or after 7 days (Day-7) of ambient temperature storage using a generic double spin centrifugation protocol and immediately frozen at -80 °C. (B) Haemolysis of blood samples collected into EDTA or NA-BCT immediately after draw or after 7 days of ambient temperature storage. (C) Plasma volume is maintained to near draw-time levels in NA-BCT.

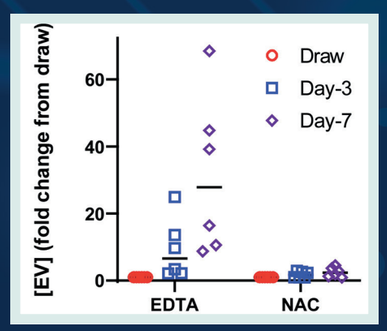


Figure 3. Concentration of EVs from plasma collected into EDTA or Nucleic Acid BCT immediately after draw (Draw) or after 3 (Day-3) or 7 days (Day-7) of ambient temperature storage. n=6 self-declared healthy donors.

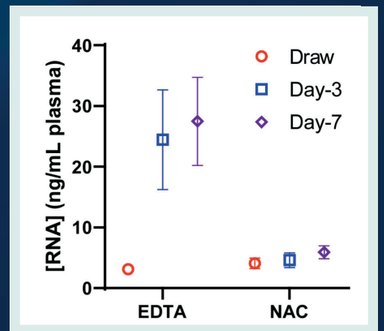


Figure 4. cfRNA concentration in plasma collected into EDTA or NA-BCT immediately after draw (Draw) or after 3 (Day-3) or 7 days (D ay-7) of ambient temperature storage.

References
Nicholas George, Ph.D., Lisa Bartron, MB(ASCP), and Jordan LaRue Nucleic Acid BCT™ maintains draw-time concentration of cell-free DNA, extracellular vesicles, and associated cell-free RNA www.streck.com/wp-content/uploads/2023/06/880237-NABCT-Tech-Note.pdf

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