

## Frequently Asked Questions (FAQs)

### 1. How long are proteins stable in a HEMACollect™•PROTEIN BCT (blood collection tube)?

Proteins in whole blood or plasma isolated from whole blood using a HEMACollect™•PROTEIN BCT are stable for up to 7 days\* at room temperature (20°C-26°C/68°F-79°F) before they need to be aliquoted and subjected to proteomic analysis or long-term storage. For long-term storage, isolate the plasma and freeze at -80°C/-112°F, or as specified in your protocol. During transport, avoid multiple freeze-thaw cycles and temperatures below 4°C/39°F.

\*Stabilization is determined from a representative subset of plasma proteins.

### 2. What types of proteins were evaluated in the HEMACollect™•PROTEIN BCT?

The HEMACollect™•PROTEIN BCT, using ProteoPrecision™ technology, preserves a wide range of plasma proteins while also minimizing hemolysis and platelet activation. The stability and performance of the HEMACollect™•PROTEIN BCT was evaluated with representative proteins by exposing samples to room temperature (RT) or simulated transport with a minimum of 24 hours at each temperature prior to RT storage for up to 7 days total. The direct compatibility of the HEMACollect™•PROTEIN BCT was demonstrated with proteomic technologies.

- **Hemoglobin (Hb):** Samples collected in a HEMACollect™•PROTEIN BCT maintained hemoglobin levels below the accepted threshold of 100 mg/dL, and this low level was maintained over the 7-day stability period.
- **Platelet activation markers:** Two protein markers for platelet activation, CD62P (also known as P-selectin), a surface protein marker, and epidermal growth factor (EGF), a protein released upon platelet activation, were evaluated. The HEMACollect™•PROTEIN BCT effectively reduced platelet activation and maintained plasma EGF levels similar to baseline for up to 7 days.
- **Blood cell protein release marker:** IL-8 is an intracellular cytokine expected to remain low in plasma samples that have not undergone significant cell lysis. IL-8 levels remained stable in samples from a HEMACollect™•PROTEIN BCT, indicating effective preservation of cellular integrity.
- **Protease activity:** Lectin-like oxidized LDL receptor-1 (LOX-1) is a Type II transmembrane protein known to be cleaved by a number of different proteases, including intramembrane, lysosomal and metalloproteases. LOX-1 levels were preserved in samples from HEMACollect™•PROTEIN BCT near baseline, indicating suppression of non-specific protease activity.
- **48-cytokine panel:** The direct compatibility and performance of the HEMACollect™•PROTEIN BCT using the Olink™ Proximity Extension Assay (PEA) was evaluated. After 7 days of storage, 85% of proteins remained stable in samples from the HEMACollect™•PROTEIN BCT within an average log<sub>2</sub> fold change of ±1.

- **100-protein panel:** The direct compatibility and performance of the HEMACollect™•PROTEIN BCT using a custom 100-target SomaLogic™ SomaScan™ panel assay was evaluated. After 7 days of room temperature storage, 93% of proteins in the HEMACollect™•PROTEIN BCT remained stable.
- **General plasma proteins (LC-MS/MS):** The direct compatibility and performance of the HEMACollect™•PROTEIN BCT with common depletion kits and LC-MS/MS workflows were evaluated. Data suggested the HEMACollect™•PROTEIN BCT may preserve labile proteins that degrade rapidly in EDTA and that some proteins over-represented in EDTA samples were linked to a lack of sample homeostasis and cell integrity maintenance (e.g., platelet basic protein, platelet factor 4, hemoglobin subunit beta).

For detailed study results, request a copy of the white paper *The novel HEMACollect™•PROTEIN blood collection tube designed for the extended preservation of plasma proteins*. (DNA Genotek, PD-WP-80). The HEMACollect™•PROTEIN BCT was evaluated for its ability to stabilize a variety of representative plasma proteins. However, users should evaluate the specific proteins for their intended application.

### 3. Does the stabilizing liquid interfere with analysis platforms? Are any plasma purification steps needed before using mass spectrometry, Olink™ or SomaScan™ platforms for downstream analysis?

The HEMACollect™•PROTEIN BCT contains a proprietary stabilizing liquid (ProteoPrecision™ technology) which does not interfere with the downstream analysis of a broad range of proteomic technologies, including common abundant protein depletion kits used with untargeted mass spectrometry workflows, immunoassays and next generation affinity-based methodologies. The HEMACollect™•PROTEIN BCT is directly compatible with the Olink™ and SomaLogic™ platforms. The HEMACollect™•PROTEIN BCT supports a range of proteomic applications without time-consuming sample preparation or cleanup steps, similar to EDTA tubes.

For details on performance with various technologies, request a copy of the white paper *The novel HEMACollect™•PROTEIN blood collection tube designed for the extended preservation of plasma proteins*. (DNA Genotek, PD-WP-80).

### 4. Is there an impact of freezing plasma on bioanalytical capabilities?

The HEMACollect™•PROTEIN BCT significantly reduces the risk of sample degradation by enabling room temperature storage of samples for up to 7 days, thereby minimizing freeze-thaw events. Freeze-thaw cycling of the sample or extreme temperature fluctuations can compromise sample quality. Uncontrolled or multiple freezing and thawing events, which can occur with general blood collection tubes that are not immediately processed and frozen, may negatively impact bioanalytical capabilities and protein integrity.

For more information, request a copy of the white paper *The novel HEMACollect™•PROTEIN blood collection tube designed for the extended preservation of plasma proteins*. (DNA Genotek, PD-WP-80).

**5. Can plasma from whole blood collected in the presence of other additives be added to a HEMAcollect™•PROTEIN BCT to achieve protein stability?**

The HEMAcollect™•PROTEIN BCT with its ProteoPrecision™ technology is specifically designed to stabilize proteins directly at the point of collection when whole blood is drawn into it. Adding blood or plasma from other tubes (e.g., EDTA) will not confer the stability benefits of the HEMAcollect™•PROTEIN BCT.

**6. Is it suitable for other analytes like cfDNA?**

The HEMAcollect™•PROTEIN BCT has been specifically designed and tested for its ability to preserve plasma proteins for proteomic research.

**7. What are the cost implications?**

The HEMAcollect™•PROTEIN BCT is a cost-effective alternative to general BCTs by

- eliminating cold-chain shipping, enabling room temperature storage and ambient temperature transport and facilitating efficient batch processing.
- reducing the risk of sample rejection, thereby minimizing the need for repeat sample collection and processing.
- simplifying logistics and geographic reach, especially in regions with limited cold-chain infrastructure.

**8. Which proteomic analysis technologies is the HEMAcollect™•PROTEIN BCT compatible with?**

The HEMAcollect™•PROTEIN BCT is designed to enable downstream analysis across a broad range of technologies, including traditional and affinity-based methods. The HEMAcollect™•PROTEIN BCT integrates seamlessly into existing workflows without requiring additional steps or protocol changes and helps generate high-quality data across diverse platforms.

- **ELISA-based immunoassays:** ELISA assays were conducted on plasma isolates to evaluate critical markers of preanalytical variability: ex vivo platelet activation (CD62P, EGF), blood cell protein release (IL-8) and protease activity (LOX-1).
- **Mass spectrometry (LC-MS/MS):** Samples derived from a HEMAcollect™•PROTEIN BCT are compatible with common LC-MS/MS workflows and upfront abundant protein depletion kits (e.g., Thermo Fisher Scientific High-Select™ Top 14, Norgen ProteoSpin Abundant Serum Depletion Kit). The number of total proteins detected following depletion is comparable between samples collected in a HEMAcollect™•PROTEIN BCT and an EDTA BCT.

- **Olink™ Proximity Extension Assay (PEA):** Plasma derived from the HEMAcollect™•PROTEIN BCT is directly compatible with Olink™ PEA and demonstrated superior stabilization performance relative to EDTA-collected samples on the Olink™ Target 48 Cytokine Panel. After 7 days of storage, 85% (34/40) of proteins in samples from a HEMAcollect™•PROTEIN BCT remained stable within an average log<sub>2</sub> fold change of ±1 across donors, compared with only 47.5% (19/40) of proteins in EDTA-collected samples.

- **SomaLogic™ SomaScan:** The HEMAcollect™•PROTEIN BCT-derived plasma exhibited improved stabilization compared with EDTA-derived plasma on a custom 100-protein SomaScan™ panel. After 7 days of room temperature storage, 93% of proteins in a HEMAcollect™•PROTEIN BCT remained stable (within ±1 log<sub>2</sub> fold change), whereas only 76% remained stable in EDTA. After simulated transport followed by room temperature storage for 7 days, the HEMAcollect™•PROTEIN BCT maintained 93% stability compared with 69% in EDTA. This level of stability demonstrates robust performance and direct compatibility for high-throughput SomaScan™ assays, without requiring upfront desalting of plasma.

**9. What are the instructions for handling the HEMAcollect™•PROTEIN BCT in the lab?**

- The HEMAcollect™•PROTEIN BCT is made of plastic with an internal silica-based gas barrier coating which offers better resistance against breakage compared to glass — an important element in automated systems. It also maintains a vacuum over time.
- The uncapped tube is 100 mm in height and has an outer diameter of 16 mm. The tube will fit in most racks and centrifuge rotors designed for standard 10-mL BCTs.
- To isolate plasma from the HEMAcollect™•PROTEIN BCT, centrifuge the tube at 1,900 × g for 15 minutes at a temperature between 4°C–25°C (39°F–77°F). Carefully remove the upper plasma layer and transfer the plasma to a new conical tube (not provided). Centrifuge the plasma at 2,800 × g for 15 minutes at a temperature between 4°C–25°C (39°F–77°F). Transfer the plasma into low protein-binding tubes (not provided). For the long-term storage of plasma, store in low protein-binding tubes at -80°C/-112°F, or as specified in your protocol.

**10. How is the HEMAcollect™•PROTEIN BCT workflow different than EDTA BCTs for blood collection?**

There are no major differences in the blood collection workflows and minimal training is needed for phlebotomists. Detailed instructions can be found in the HEMAcollect™•PROTEIN BCT instructions for use and best practices guide.

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The HEMAcollect™•PROTEIN BCT is for research use only and not for use in diagnostic procedures. Some DNA Genotek Inc. products are not available in all geographic regions.

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Customers must review the labels of sample collection devices and ensure compliance with their intended requirements.

To request samples or a quote, or if you have any questions or concerns, visit [www.dnagenotek.com](http://www.dnagenotek.com) or email [info@dnagenotek.com](mailto:info@dnagenotek.com). For technical support, email [support@dnagenotek.com](mailto:support@dnagenotek.com).