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Sera-Xtracta Cell-Free DNA Kit

FAQs

Applications and automation

1. **What is Sera-Xtracta™ Cell-Free DNA Kit?** Sera-Xtracta Cell-Free DNA Kit is designed for the rapid extraction and purification of cell-free DNA from a range of sample types including plasma, serum, and urine. The protocols are rapid and have been developed to select for cfDNA while minimizing higher molecular weight genomic DNA contamination.
2. **Are there any limitations on the sample types that can be extracted with Sera-Xtracta Cell-Free DNA Kit?** The kit is recommended for the isolation of cell-free DNA from plasma (isolated from fresh, or frozen and thawed blood), serum and urine with an elution volume of 30 µL for a standard input of 2.0 mL plasma.
3. **How should blood collected in standard collection tubes be processed to optimize cell-free DNA extraction?** Processing for plasma and serum should be conducted as soon as possible after collection (no later than 4 hours). The blood should at no point be frozen otherwise, cellular degradation will result in the plasma being contaminated with an excess of genomic DNA and alteration of the small-fragment DNA population in the plasma. Blood samples should ideally be collected in blood vacutainers specifically intended for stabilization of cfDNA content (e.g. Cell-Free DNA BCT™ [Streck]).
4. **Is the Sera-Xtracta Cell-Free DNA Kit compatible with all blood collection/cfDNA stabilizing tubes?** The performance of Sera-Xtracta Cell-Free DNA kit was evaluated using blood collected in Cell-Free DNA BCT collection tubes (Streck), PAXgene™ Blood ccfDNA Tubes (Qiagen) and Cell-Free DNA collection tubes (Roche). The recovery of the main cfDNA peak is indistinguishable between the three blood collection tubes types.
5. **What is the yield level provided by the kit?** Levels of cfDNA can be highly variable from sample to sample, making yield specifications less meaningful. The Sera-Xtracta Cell-Free DNA Kit has been optimized to bind fragments in the range of 50–500 bp, and estimated recoveries of > 70% based on spike-in experiments have been observed.
6. **What type of particles are used in the Sera-Xtracta Cell-Free DNA Kit?** Sera-Xtracta Cell-Free DNA Kit includes silica-coated magnetic beads.
7. **What is the protocol duration?** The isolation procedure can be completed in less than 2 hours to yield cell-free DNA with a purity and quality that is compatible with most molecular biology techniques, including Bioanalyzer™ (Agilent Technologies) analysis, PCR, digital PCR, genotyping and next-generation sequencing (NGS).
8. **Can Sera-Xtracta Cell-Free DNA Kit be easily automated?** Yes. Use of paramagnetic beads allows use on any open automation platform. When automating, pay attention to the settling time and employ intermittent mixing if necessary.

   Note: the initial addition of binding mix to plasma, serum, and urine may need to be added manually, depending on automation platform capabilities.

Storage and handling

1. **How should Sera-Xtracta Cell-Free DNA Kit be stored?** All kit components should be stored at room temperature (15°C–30°C). All kit components can be stored at room temperature. Optional storage of liquid Proteinase K at 2°C–8°C.
2. **Why is Proteinase K included in the kit/protocol?** Proteinase K is required to isolate cfDNA from blood collected in stabilizing Streck™ tubes and is beneficial in removing protein that may be associated with the fragmented cfDNA. It is active even when enzyme inhibitors such as EDTA and detergents are present in samples, so is suited for use in such protocols.
3. **Is a pulse spin in a microcentrifuge necessary before each magnetic settling step?** A pulse spin in a microcentrifuge is strongly recommended before magnetic settling, to ensure all the liquid sample in the tube is collected together in a single bulk volume at the bottom of the tube. Isolated droplets on the tube walls or trapped under the tube lid will not settle on the magnet properly.
4. **Can the purified cell-free DNA be stored at room temperature?** Purified cell-free DNA may be stored at 2°C–8°C for a short period, if being used directly for analysis and/or downstream molecular biology applications. For longer term storage, store purified isolates at -20°C or less.
5. **Can DNase-free water be used for eluting samples?** Although DNase-free water alone can be used for elution, for longer term, we recommend storage in the elution buffer provided to minimize acid hydrolysis.
6. **Can the elution buffer be substituted?** The elution buffer provided is satisfactory for eluting cfDNA for most downstream applications; however, the end user may substitute another tris-based buffer for this if required.
7. **How many reactions can be performed with one Sera-Xtracta Cell-Free DNA Kit?** Each kit is designed to provide 96 purifications based on sample input volume of 2.0 mL. Processing of different volumes is possible, but will affect the total number of purifications that can be achieved. See the full IFU for further direction.

Physical characteristics

1. **Where can I find the Certificate of Analysis (CofA) for my lot of Sera-Xtracta Cell-Free DNA?** Enter your specific lot number to retrieve the CofA at cytiva.com/certificates.

2. **What is included in the Sera-Xtracta Cell-Free DNA Kit?**

Component	Quantity
Proteinase K	2 × 3 mL
20% SDS	12 mL
Magnetic silica bead suspension	1.6 mL
Binding buffer	2 × 150 mL
Wash buffer 1*	85 mL
Wash buffer 2*	35 mL
Elution buffer	10 mL

*End user is required to add indicated volumes of ethanol before first use.

3. **What additional reagents do I need to provide?** The end user is required to supply:
   - Absolute ethanol, for completion of the two wash buffer formulations before first use.
   - Isopropanol, for use in the cfDNA binding step, alongside the magnetic beads and binding buffer.

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