Introduction
Many hospitals and research institutes are building up large collections of biological samples, together with associated medical and lifestyle data, as a resource for biochemical research. Typically the objective is to better understand the causes of disease and treatment response.

Many of these biorepositories carry on collecting data from participants over a long time period and so the value of a stored sample becomes greater over time. For many biorepositories long-term, secure sample storage is of absolute importance, with expectations of 25+ years storage not uncommon.

Recent data has shown the importance of minimizing freeze-thaw cycles on samples and many biobanks are now adopting single use tubes to ensure sample quality is maintained throughout the sample life. This application note discusses these issues, highlights the Brooks REMP single use tube system and shows the example of the sample flow being used at the Karolinska Institute, Stockholm, Sweden, for Blood Serum, Urine and Plasma.

Serum, Plasma and Urine Storage
It is generally accepted, that for long term stability of analytes, the following are required:

• Storage at -80°C or below (vapor phase liquid nitrogen -196°C is often used)
• Minimum freeze-thaw cycles (ideally just one)
• No risk of cross-contamination

There is also some evidence that exposure to light should be minimized. However, there are a number of practical constraints that make 100% compliance with these requirements a challenge:
1. Low temperature freezers are inconvenient to use and automated freezers are only available down to -80°C.
2. Multiple freeze-thaw cycles are hard to avoid:
   • Given that many samples will be required several times over the life of the biorepository.
   • In manual freezers where racks/trays of samples must be pulled into room temperature for a human to pick.
3. Samples will need to be thawed and then decapped for a sub-sample to be taken.
4. Storage at -80°C or below generally relies on an ‘O’ ring in a screwcap tube to ensure a good seal. Most tubes and caps do not come with long term storage data.
5. Sub-samples may need to be shipped around the world to be tested (requiring re-freezing)

Figure 1: Typical Biorepository Sample Flow: note that for remote dispatch on dry ice a freeze thaw step is necessary

As more data has become available that shows the impact of freeze thaw cycles on the analytes in blood serum and plasma samples1,2 more biorepositories have been insisting on a process flow that requires only a single freeze-thaw cycle:

Figure 2: Biorepository Sample Flow with Single Use Tubes
The key differences are:

- Multiple low volume aliquots of each sample are taken for single use
- Each aliquot is stored in a 2D barcoded tube in an automation friendly SBS format
- When a sample is requested the sample is picked and dispatched and can remain frozen and sealed right up to the point of analysis
- The sample is exposed only twice (the alternative is to repeatedly take sub-samples from a single storage tube, resulting in multiple cap openings, each one a chance to contaminate the sample)
- Each sample is frozen and thawed only once (The alternative is to repeatedly freeze and thaw the single storage tube as each aliquot is taken)

Figure 3: 96 Heat sealed tubes, each with up to 300μl capacity

Brooks Life Science Systems’ REMP tubes have long led the way in single use tubes. A heat seal is an excellent choice for sealing a single use tube:

- An aluminium/polypropylene laminate or foil is welded to the top of the tube; this seal will stay in place right up to the point of analysis
- The foil seal saves significant amounts of space over other types of caps and seals
- The foil seal adds minimal cost to the tube
- Tubes are only picked once, thus data tracking integrity is inherently good; thus many users can demonstrate secure operations using uncoded tubes which leads to further saving on cost

Figure 4: Typical Process Flow for Storing Blood Serum Samples in Single Use Heat Sealed Tubes: Best Practice for Good Sample Quality.

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References
