

## Standard Operating Procedure (SOP) 003V8.0

Acquisition of Serum from Whole Blood  
SPREC SER-SST-A-A-N-B-A [3]

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### Materials:

**Blood collection sets:** BD (Becton, Dickinson and Company) Vacutainer™ Blood Collection Set, 21 gauge butterfly (Fisher cat. # 02-664-1)

**SST Collection tube:** BD Vacutainer™ Venous Blood Collection Tubes: SST\* Serum Separation Tubes Red/Gray top 8.5ml (Fisher cat. #02-683-96)

**Centrifuge:** Eppendorf 5702 or 5702R

**Cryostorage tubes:** Corning 2.0ml Cryogenic Vials. (Fisher cat. # 0337421)

**Repeater Pipet:** Eppendorf Repeater Plus Pipette (Fisher cat. # 21-380-9)

**Combitips:** (Fisher cat. #13-683-705)

**Labelling:** All tubes are to have bar code stickers placed on the tube prior to venipuncture. Bar code packets are assigned during the donor registration process.

**Position for venipuncture:** sitting

**Order of the Blood Draw:** Blood collection tubes must be drawn in a specific order to avoid cross-contamination of additives between tubes. [5] The order of draw is 1) SST, 2) EDTA 9ml (SOP 001V8.0), and 3) EDTA 2ml (SOP 001V8.0). A total of three tubes of blood are drawn during the collection process.

**Temperature for collection and processing:** Blood samples to be separated into serum are drawn into an SST and allowed to clot at room temperature. [4]

**Processing:** Blood is drawn into the Serum Separator tube (SST) and gently mixed by inverting eight times immediately after drawing. Tube is incubated upright in a tube rack at room temperature for 45 minutes ( $\pm 10$  min.) after the blood has been drawn to ensure complete coagulation. Following incubation and clotting, the Serum Separator tube is centrifuged at 1200rcf for 10 min. A repeater pipet is used to aliquot 600ul of the top serum layer directly into each of five pre-labeled cryogenic vials. If serum volume is low, fewer aliquots are collected. If serum volume exceeds 3ml, existing 5 vials are topped off. Vials are capped and immediately placed into cryoboxes on dry ice.

**Storage of Serum:**

Best Practice recommends that separation into serum and placement of serum into frozen storage should occur within 2 hours of the blood draw [1]. Freeze-thaw is not optimal [2] and therefore, serum should be aliquoted. Serum aliquots are logged into cryoboxes and placed on dry ice for transport to the storage facility. Serum is stored at  $-80^{\circ}\text{C}$ .

**Standardization:** All variables including the time the whole blood is at room temperature prior to separation, time stored at  $-80^{\circ}\text{C}$  as serum prior to shipment and/or utilization, volume of aliquots and color of serum will be entered into the database.

**Oversight:** All adverse and unexpected events will be recorded in the database and will be addressed by the Executive Committee. This includes all phases of the process: donation, storage and retrieval, processing, and utilization.

## **References:**

1. Leyland-Jones, Brian R. et al., *Recommendations for Collection and Handling of Specimens from Group Breast Cancer Clinical trials, from Onsite Collection through Shipping to the Central Bank*. Journal of Clinical Oncology 2008 26:34, 5638-5644
2. Mitchell BL, Yasuie Y, Lia CI, et al. (2005). Impact of Freeze-thaw Cycles and Storage Time on Plasma Samples Used in Mass Spectrometry Based Biomarker Discovery Projects. *Cancer Informatics* 1:98-104.
3. Sabine Lehmann et.al. International Society for Biological and Environmental Repositories (ISBER) Working Group on Biospecimen Science. Standard preanalytical Coding for Biospecimens: Review and Implementation of the Sample PREanalytical Code (SPREC). *Biopreservation and Biobanking* Vol. 10 No.4, 2012
4. Tuck, Melissa K et al. "Standard operating procedures for serum and plasma collection: early detection research network consensus statement standard operating procedure integration working group." *Journal of proteome research* vol. 8,1 (2009): 113-7. doi:10.1021/pr800545q
5. WHO Guidelines on Drawing Blood: Best Practices in Phlebotomy. Geneva: World Health Organization; 2010. 2, Best practices in phlebotomy. Available from: <https://www.ncbi.nlm.nih.gov/books/NBK138665/>

## **Bibliography**

- Leyland-Jones, Brian R. et al. Recommendations for Collection and Handling of Specimens from Group Breast Cancer Clinical trials, from Onsite Collection through Shipping to the Central Bank. *J Clin Oncol*. 2008 Dec 1;26(34):5638-44. doi: 10.1200/JCO.2007.15.1712. Epub 2008 Oct 27.
- Mitchell BL, Yasuie Y, Lia CI, et al. (2005). Impact of Freeze-thaw Cycles and Storage Time on Plasma Samples Used in Mass Spectrometry Based Biomarker Discovery Projects. *Cancer Informatics* 1:98-104.

## **Electronic Resources**

- Arzoumanian, Lena. Tech Talk Vol.4, No.2  
[https://fritsmafactor.com/sites/default/files/attachments/2012/01/techtalk\\_november2005\\_vs7436.pdf](https://fritsmafactor.com/sites/default/files/attachments/2012/01/techtalk_november2005_vs7436.pdf)
- Holland Lab/Berkeley <https://www.hollandlabucb.org/>
- <http://library.med.utah.edu/WebPath/TUTORIAL/PHLEB/PHLEB.html>
- [http://www.geisingermedicallabs.com/catalog/blood\\_specimens.shtml](http://www.geisingermedicallabs.com/catalog/blood_specimens.shtml)