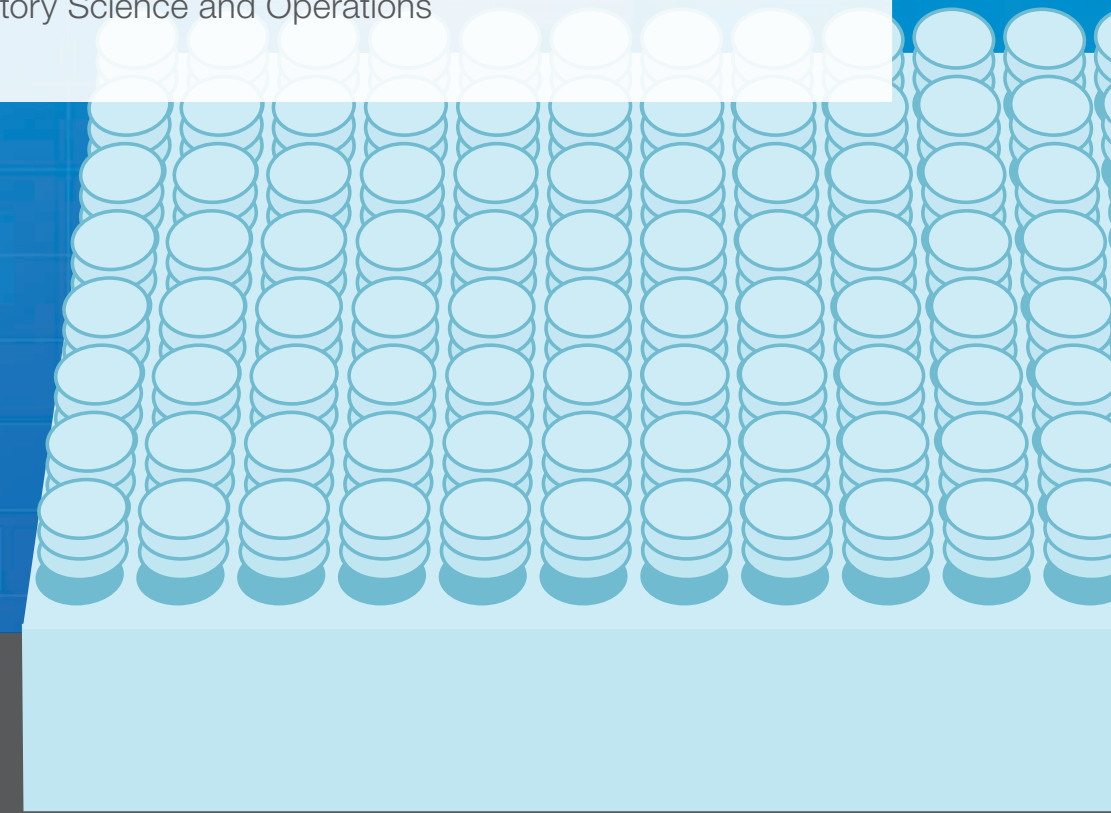


# AVOIDING HEMOLYSIS BLOOD SAMPLE COLLECTION PROCESSING

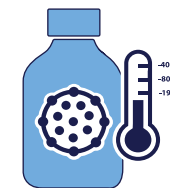
By Abdul Ally, Area Director, Laboratory Science and Operations





this eBook

# BIOBANKING & BIOREPOSITORY



GMP Biologics Management



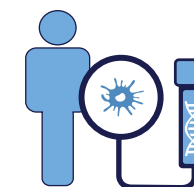
Kit Production



Public Health Research



Qualification/Validation



Advanced Therapy



Laboratory Processing



Biobanking & Biorepository



Cold-Chain Logistics

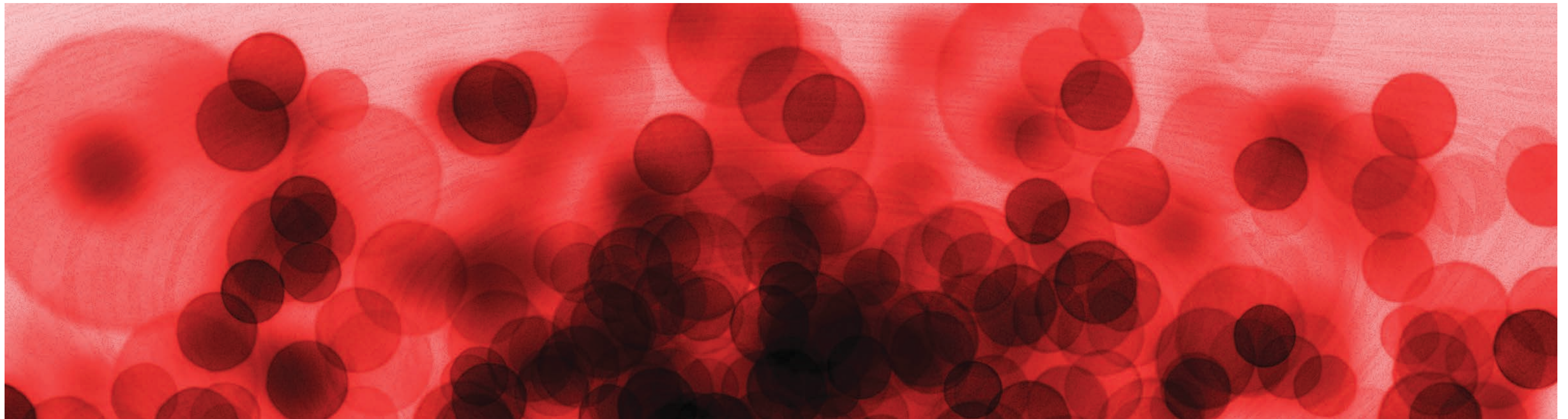
## ABOUT THE AUTHOR



by Abdul Ally  
Abdul Ally is the Area Director of  
Laboratory and Operations Science

He has more than 28 years of experience in molecular biology, laboratory management, and managing contract research operations for clinical research support. Mr. Ally advises clients on specimen collection and processing, and develops custom processes to meet client research specifications. He has published more than 18 articles in peer-reviewed publications, holds four US patents, and has been involved in such highly diverse activities as genomics research and development, managing contract research laboratories under FDA compliance, and developing and fitting out genomics and nucleic acid research core facilities for King Abdullah University for Science and Technology (KAUST) in Thuwal, KSA.

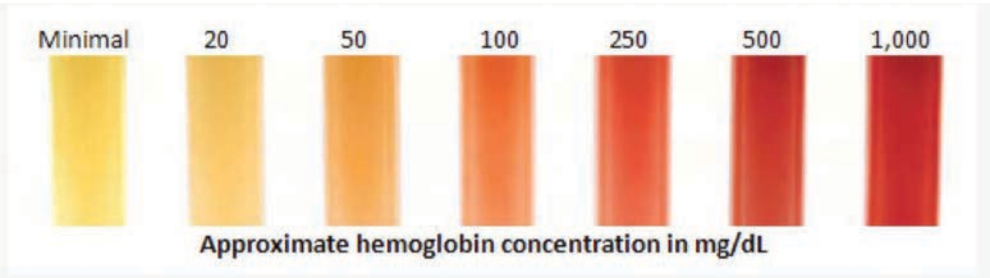




# OVERVIEW

Depending on your point of view, red blood cells, or erythrocytes, are throwaways and a nuisance, ruining a good blood specimen if the cells burst and contaminate the sample. On the other hand, erythrocytes have significant value in specific types of research, and if your objective is biobanking these cells in an intact state, then hemolysis and the need for a re-draw is equally as much to be avoided.

Erythrocyte cell membranes rupture easily, releasing hemoglobin and flooding the sample with potassium and other internal components. Fortunately, breakage of erythrocytes is easy to detect, as the hemoglobin turns the serum or plasma sample from pink to red, depending on the number of cells that have lysed. Hemolysis is a primary driver of the need for re-draw, resulting in wasted time and resources. How can hemolysis be prevented?

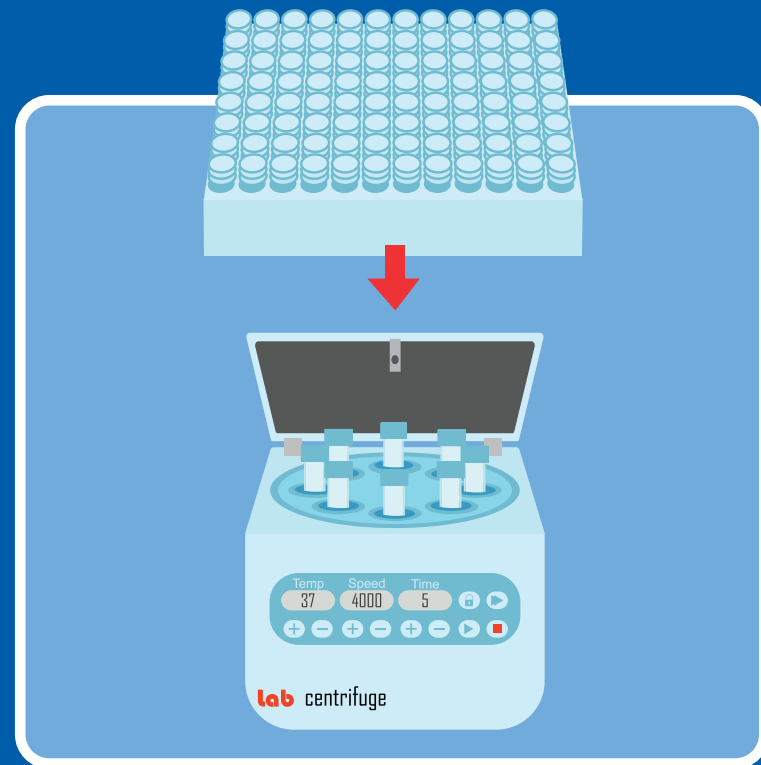


In-vivo hemolysis can occur with certain disease conditions (autoimmune hemolytic anemia) or as a reaction to a transfusion. However, most often the hemolysis is the result of improper specimen collection and handling; prevention rests primarily with those doing the collection and initial processing of the specimen. Planning specimen collection carefully, establishing a correct process, and training in the process is critical.

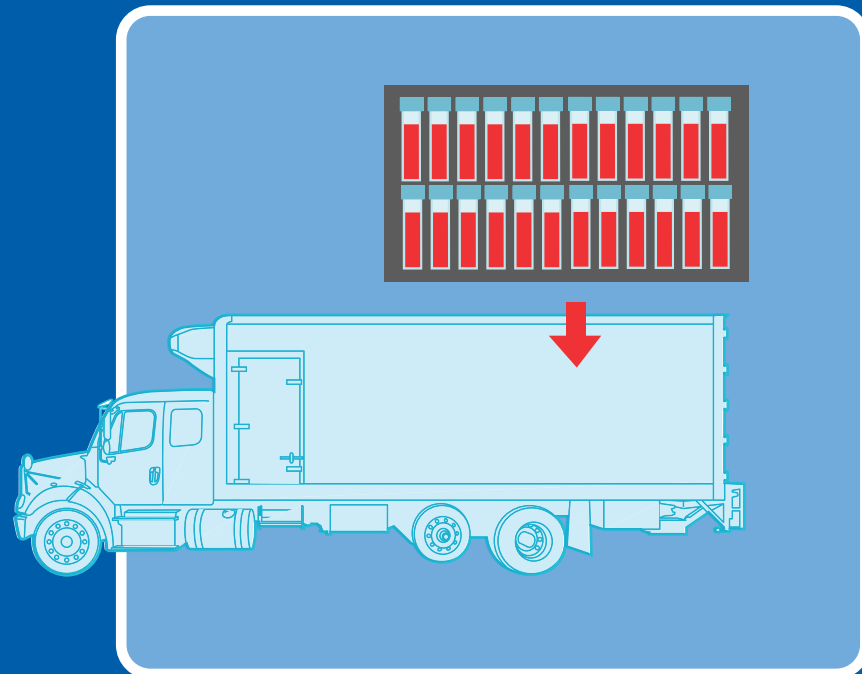
# PREVENTING HEMOLYSIS DURING SPECIMEN COLLECTION



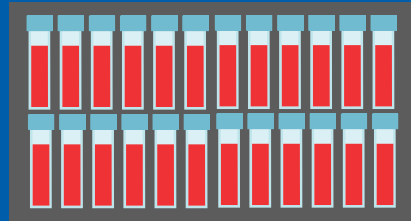
- Use a 20-22 gauge needle for routine collection; too small a needle results in excess vacuum force, while too large a needle can cause shear stress on the cell walls.
- Draw the sample from the antecubital region of the arm; drawing from other sites—sometimes a necessity in the emergency room—has been shown to result in a higher degree of hemolysis.
- Warm up the puncture site; warming increases the blood flow and prevents the need to “milk” the site, a significant cause of hemolysis.
- Do not leave the tourniquet on for longer than one minute; prolonged tourniquet time causes the interstitial fluid to leak into the tissue, promoting hemolysis.
- Alcohol damages cell walls; allow the venipuncture site to completely air dry after cleaning it with alcohol.
- Place the needle correctly in the vein; if the bevel of the needle is crowded by the inner wall of the vein, the partial occlusion exerts a dramatic shear force on the cells. This is typically indicated by too slow a blood flow.
- When using a syringe, pull the plunger gently; pulling too quickly exerts excess pressure—well beyond that of a standardized evacuated tube—and will shear the cell walls.
- Similarly, pushing hard on the syringe plunger while transferring blood to another tube exerts a destructive level of pressure, and can also cause loss of the sample if the stopper comes off.
- Avoid drawing from catheters and lines; these are designed to deliver fluids to the patient, not drawn from the patient. Drawing blood samples from these systems involve shear forces and turbulence that makes hemolysis unavoidable.
- Fill tubes to correct volume; under-filling of tubes containing anticoagulant results in a higher than recommended concentration of the additive, which promotes hemolysis. Use a smaller tube for difficult draws.



# HEMOLYSIS AND SPECIMEN HANDLING



- Mix additives with the specimens gently; vigorous mixing or shaking can break the cells.
- For tubes with a clot activator, gently invert the specimen five times to ensure complete mixing, and allow the activator to work for a full 30 minutes with the tube in a vertical position.
- For serum tubes without a clot activator, invert—just allow the sample to clot for 60 minutes with the tube in a vertical position.
- Sodium citrate tubes for coagulation testing should be inverted only three or four times.
- All other anticoagulant tubes should be gently inverted eight to 10 times.
- Clotting time cannot be rushed; centrifugation of the sample too soon will result in hemolysis.
- Don't centrifuge specimens at a higher speed or for longer than necessary.
- Protect the specimens during shipping; exposure to inappropriate temperatures and significant jarring will cause hemolysis in transit.



# PRESERVING RBCS FOR BIOBANKING AND DOWNSTREAM RESEARCH

Given their lack of nuclear genetic material, erythrocytes tend to have less value than other cells in blood, but can be useful for specific research, including malaria and genetic abnormalities in fetal nucleated erythrocytes. These cells can be frozen in 40 percent (w/v) glycerol and stored at  $-80^{\circ}\text{C}$ . Red blood cells have also been frozen in solutions of 24 percent (w/w) hydroxyethyl starch and stored frozen at temperatures

above  $-75^{\circ}\text{C}$ , how-ever, warmer storage temperatures allow hemolysis of the cells. Preserving erythrocytes by spray drying, freeze drying, and other means, for storage at room temperature has also been investigated and is a possibility.

Whether preserving or discarding red blood cells, careful collection and processing will prevent you seeing red.



## ADDITIONAL RESOURCES

Whether preserving or discarding red blood cells, careful collection and processing will prevent you seeing red. It's important to understand the proper mechanisms required to maintain your samples integrity throughout the handling process. To learn more about biosample management, download our eBook

**Standardizing Biosample Management: Why Use Collection Kits?**

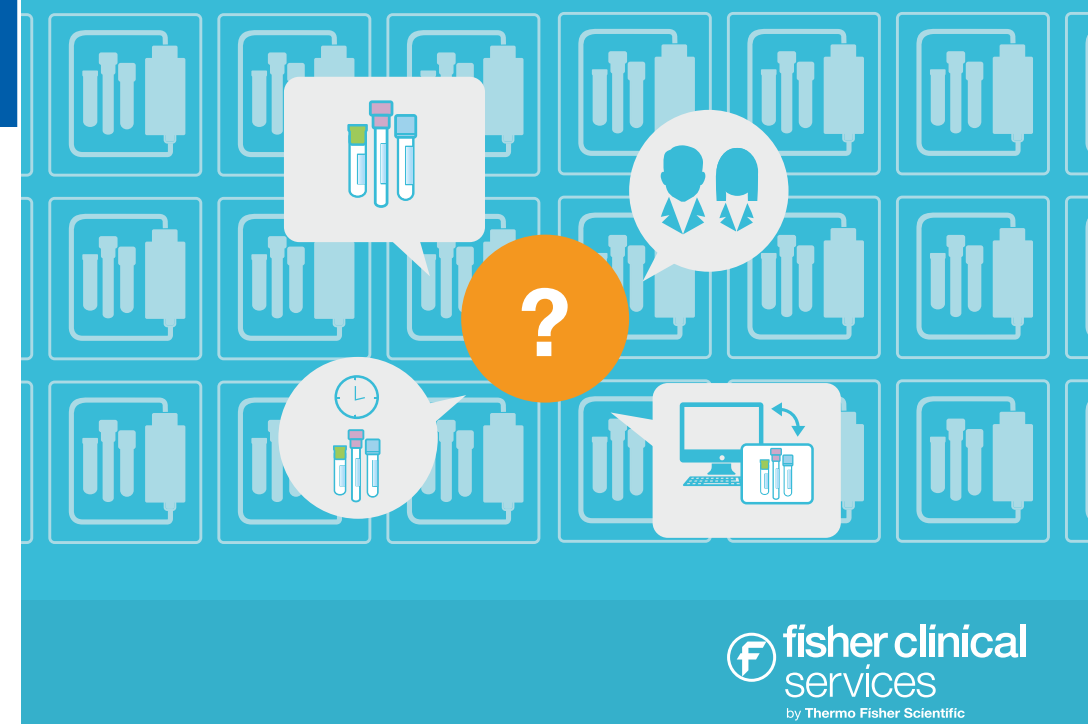
### Standardizing Biosample Management:

#### Why Use Collection Kits?



Scott A. Hixon, *Area Director of Technical Services*

Ian E. Sutherland, MS, *Area Director of Resource Planning*



Find out more at [fisherbioservices.com](https://fisherbioservices.com)

#### Talk with an expert:

Toll Free (US Only) 888-462-7246

Direct: +1 301-315-8460

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