



PREANALYTIC PULSE

Troubleshooting Hemolysis

Hemolysis is problematic for laboratories and remains one of the most common causes of specimen rejection. Hemolysis is the rupture of red blood cells and the release of intracellular contents. This phenomenon can impact laboratory testing in several ways. For example, potassium values can be erroneously elevated. The release of hemoglobin imparts a red color to the serum or plasma that can interfere with reagent reactions and photometric reading of the reaction mixture.

The most frequent causes of hemolysis involve preanalytic errors that are preventable.

ERROR	EFFECT	PREVENTION
SPECIMEN COLLECTION		
Extended tourniquet application	Leaving the tourniquet on for over one minute causes hemoconcentration and potential for cell rupture.	If vein location takes longer than one minute, the tourniquet should be released for a minimum of two minutes before reapplying.
Alcohol used for cleansing site still wet when puncture performed	Residual alcohol can bring about increased pain and discomfort to the patient and hemolysis of cells exposed to the alcohol.	Allow alcohol to dry for 15-30 seconds.
Traumatic draw	Vein collapse, trauma to the vein or probing for vein location can cause cells to rupture.	Choose appropriate equipment for vein condition and avoid probing for the vein. If the vein cannot be located by moving the needle slightly up or down, then the needle should be removed and a 2nd venipuncture attempted.
Short draw	Inappropriate ratio of blood to additive can cause cells to rupture.	Fill tubes to the fill mark or range provided on the label of all Greiner Bio-One VACUETTE® tubes. Ensuring that the tube is properly seated, appropriately anchoring the tube during the draw, and waiting for blood flow to cease prior to tube removal will help ensure proper fill.
Vigorous mixing of tube	Mechanical trauma to the red cells causes rupture.	Tubes with additives should be mixed using the appropriate number of complete, gentle inversions as stipulated in the VACUETTE® Instructions for Use.
Draw from a vascular access device (VAD)	Most of these devices are meant for infusion and can cause trauma to cells as blood is withdrawn.	Avoid drawing from VADs if another site is available. If not, approximately 5cc of fluid should be withdrawn and discarded prior to drawing the sample. If a syringe is used, blood should be transferred to tubes using an appropriate transfer device.
Inappropriate needle gauge	Choosing a needle gauge that is too small can result in forces that can rupture cells. If too large, the needle may injure the vein and result in a traumatic draw.	Select the appropriate gauge for the vein size and condition.
Inappropriate tube draw volume	Too much vacuum applied to a small or fragile vein can result in a traumatic draw and hemolysis of cells.	Select the minimum draw volume acceptable for the test(s) ordered.

TROUBLESHOOTING HEMOLYSIS

ERROR	EFFECT	PREVENTION
Excessive pulling force with a syringe	Pulling back too rapidly on the syringe plunger creates excessive force on cells being pulled into the barrel causing rupture.	After unseating the plunger to expel all air in the syringe barrel, proceed with venipuncture and pull back slowly and steadily once the vein has been accessed. Transfer the sample to tubes using an appropriate transfer device.
SPECIMEN TRANSPORT		
Excessive temperature fluctuation	Extreme temperature changes can cause cells to rupture. For example, sitting in a hot or cold car, depending on the season and geographical area, during transport.	Blood samples should be maintained at the appropriate temperature for the test(s) ordered. Extreme fluctuation during transport (couriers, shipping) can be prevented by using insulated containers to store samples.
Pneumatic tube systems	Sudden starts and stops in containers moved by vacuum or compressed air can put excessive force on cells causing them to rupture.	Pneumatic tube systems should be validated for transport of blood samples prior to use to ensure there is no adverse effect to the sample.
Time delay	The longer plasma/serum is in contact with the cells, the more likely analyte levels are altered and the greater the chance of intracellular changes and hemolysis.	Samples should be centrifuged within two hours of collection when appropriate for the test(s) ordered.
SPECIMEN PROCESSING		
Excessive centrifugation	Centrifuging for too long or with too much centrifugal force will cause red cells to rupture.	Samples should be centrifuged according to manufacturer's recommendations. Centrifuging with alternative settings must be validated by the site prior to implementation.
Excessive temperature fluctuation		Samples should be centrifuged under the conditions stipulated in the VACUETTE® Instructions for Use.
Rimming the tube	Mechanical disruption of cell membranes with wooden applicator sticks used to remove microclots or fibrin results in hemolysis.	
Time delay	Significant delays in processing can cause alteration in analyte levels and increase the chances of intracellular changes and hemolysis.	Processing of samples should be carried out in a timely fashion to avoid compromising test results.
STORAGE		
Plasma/serum contact with cells	Red cells are more likely to rupture as they age as with extended storage.	Length of storage should not exceed test manufacturer's recommendations.
Freezing samples for long term storage	Red cells should never be frozen since formation of ice crystals within the cells will cause them to rupture.	Separate plasma or serum from cells prior to freezing.