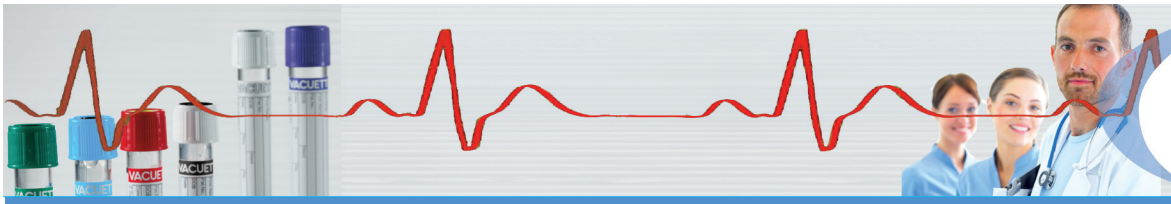

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Coagulation Testing: High Hematocrit-Anticoagulant Adjustment

In 1980, CLSI released H21-A4 guidelines for coagulation testing, which included the recommendation to adjust/correct the amount of citrate in blue-top evacuated blood collection tubes for patients presenting hematocrits greater than 55%. In addition to the CLSI guidelines, the CAP hematology checklist HEM.22830 includes the question, “Are there documented guidelines for the detection and special handling of specimens with elevated hematocrits?”

Principle

The adjustment is to assure the plasma:anticoagulant ratio (not the blood:anticoagulant ratio) stays consistent. A patient with high hematocrit, greater than 55%, will result in less plasma after centrifugation. The plasma fraction will contain an increased concentration of sodium citrate anticoagulant. The increased concentration may result in falsely prolonged test results for Prothrombin Time (PT) and Activated Partial Thromboplastin Time (aPTT).

Formula

1. To calculate the corrected 3.2% sodium citrated whole blood for hematocrit > 55%, adjust the citrate to the proper volume with the following formula:
   $C = (1.85 \times 10^{-3})(100\text{-}HCT)(V_{blood})$
   Where:
   C = volume of sodium citrate required for that volume of blood
   HCT = patient’s hematocrit
   V = volume of blood required in the blood collection tube (example if a 3mL tube is used the blood draw volume is 2.7mL)
   1.85×10^{-3} is a constant (considering the citrate volume, blood volume, and citrate concentration).

2. Example: Patient has a Hct of 60% and the patient’s blood will be drawn into a 3mL VACUETTE® sodium citrate (blue-top) blood collection tube. Adjustment of the sodium citrate volume is calculated as:
   $C = (1.85 \times 10^{-3})(100\text{-}60)(2.7mL)$
   C = 0.20mL (rounded up from 01.998mL)
   Remove: 0.30 - 0.20 = 0.10mL of sodium citrate to be removed (0.30 is the difference between the total tube volume of 3.0mL and the blood drawn into the tube of 2.7mL)

Method

The instructions to prepare an adjusted sodium citrate tube are to be documented in the Laboratory Standard Operating Procedures. Basic information should cover:

- Determination of the patient’s hematocrit level from hematology testing.
- Based on the patient hematocrit, calculation of the amount of citrate to be removed by using the previously discussed formula.

- Withdrawal of the amount of citrate calculated and discard by uncapping the tube and using a sterile pipette (recap when completed). Perform venipuncture using a syringe. Transfer blood from syringe to the adjusted tube using an appropriate transfer device and filling to within the range indicated by the black arrow (see below) to assure the 9:1 ratio. Transfer should be done immediately. Alternatively, procedures may include withdrawal of the amount of citrate calculated and discard by inserting a sterile tuberculin syringe through the rubber stopper. Correctly done, the vacuum will not be affected. Perform venipuncture using the adjusted tube and fill to within the range indicated by the black arrow (see below) to assure the 9:1 ratio.

- Mixing of the specimen by gently inverting the tube four to eight times.
- Centrifugation according to manufacturer's recommendation or laboratory's validated procedure.

Related Clinical Information

The "unproven leap of faith", as it has been referred to, has been the necessity to adjust sodium citrate and whether the adjustments should extend to coagulation assays in addition to PT and aPTT. Early studies, which were incorporated in to CLSI guidelines, were performed on "normal" samples where citrate concentration was adjusted to simulate high hematocrit values and only considered PT and aPTT values. Marlar, et.al. questioned the statistical significance of using "normal" subjects and performed a direct study on actual samples from patients with hematocrits greater than 55%. Included in the study results was the impact of adjustment on coagulation assays for Fibrinogen, Factor VIII, Protein C activity, and Protein C antigen. Marlar's data demonstrated statistically significant clinical differences in the value between citrate-adjusted and non-citrated-adjusted samples for PT and aPTT, supporting the adjustment of citrate for patient with hematocrits greater than 55%. Differences in Fibrinogen, Factor VIII, and Protein C activity were also statistically significant, however, fewer results were clinically significant, while Protein C antigen values were not significantly different.

Though the adjustment of sodium citrate where hematocrit is greater than 55% has been debated, there is evidence that hematocrit values below the normal range (22% or lower) require no adjustment.

References
CLSI Guideline for reference is "Collection, Transport, and Processing of Blood Specimen for Testing Plasma-Based Coagulation Assays and Molecular Hemostasis Assays: Approved Guideline – Fifth Edition," H21-A5 Vol. 28 No. 5.
Midyett, R. "Under the blue top: coags, corrections, and 'crits"; Medical Laboratory Observer (MLO), February 2005, pages 20/22.
Marlar, R., et.al, Effect on routine and Special Coagulation Testing Values of Citrate Anticoagulant Adjustment in Patients with High Hematocrit Values; Am J Clin Pathol, 2006, 126: 400-405.