ECAT Newsletter

Case Report

Effect of high haematocrit levels on coagulation testing

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Case description

A 42-year-old man presented at the emergency department in April 2022 with rectal blood loss. The patient had a history of cardiac problems. He was diagnosed in 1979 with congenital tricuspid atresia. The patient had an extensive surgical history of treatment to ensure adequate pulmonary and systemic blood flow: bidirectional cavopulmonary anastomosis (bidirectional Glenn connection), systemic-to-pulmonary shunts (Blalock-Taussig shunt), central shunts (Waterston shunt), Rashkind septostomy and a Fontan procedure [1]. As patients with Fontan circulation are at risk of systemic and pulmonary thromboembolic events, anticoagulation is required. He was therefore treated once daily with warfarin 5 mg and acetylsalicylic acid 80 mg [2].

Secondarily to the cyanotic chronic heart disease, the patient developed polycythaemia. Polycythaemia is defined as haematocrit or haemoglobin above the upper limit of normal. In men an increased haemoglobin > 16.5 g/dL or increased haematocrit > 49% are considered to constitute polycythaemia [3].

At the time of presentation, the laboratory results of the patient showed an extremely increased international normalised ratio (INR) of > 9.1 (reference interval (RI): 0.9-1.1) and prolonged prothrombin time (PT) of > 120 sec (< 7.2% (RI: 70-120%)). Haematocrit was 74.7% (RI: 39.9-51%) and haemoglobin 22.0 g/dL (RI: 13.5-17 g/dL). To prevent bleeding, warfarin was stopped and clinical investigations were performed, but did not reveal any bleeding focus. Anticoagulation with vitamin K-antagonists (VKA) within the therapeutic INR range of 2-3 was of the utmost importance, since the patient had had several ischaemic strokes and transient ischaemic attacks in the past. Warfarin and other VKA have a narrow therapeutic range and the anticoagulant effect is influenced by many factors including drug interactions, dietary factors and genetic polymorphisms [4,5]. An explanation for the supra-therapeutic INR was not found, as the patient did not take any drugs that interact with VKA, nor could any food interactions be found, so the laboratory was contacted to explore analytical interferences.

At the time the blood was sampled for INR testing, the haematocrit was 74.7%. Blood sampling for coagulation testing in tubes containing liquid trisodium citrate solution is standardised at a ratio of 1:9 (anticoagulant: blood). A lower blood volume, or blood with increased haematocrit alters the plasma to citrate ratio since the volume of plasma is lower in the total volume of blood, resulting in a relative excess of citrate. This may impair the recalcification process during the test procedure of coagulation assays. A haematocrit value of 55% is often considered as the threshold above which PT and activated partial thromboplastin time (aPTT) can be falsely prolonged [6].

A formula was set by the Clinical and Laboratory Standards Institute (CLSI) for patients with a haematocrit > 55% to adjust the volume of citrate in the collection tube [7]. Later on, an alternative formula was proposed by the CLSI:

$$C = (1.85 \times 10\text{-}3) \times (100 - HCT) \times (V\ Blood)$$

(Abbreviations: C= volume of citrate in mL to be added to a volume of blood, HCT = patient's haematocrit in %, and V Blood = the volume of blood in mL) [8].

In our case, 2 mL of venous blood was drawn in a 2.7 mL BD Vacutainer EST tube without additives (BD Vacuatainer, New Jersey, US). Next, the correct volume of citrate (92.5 µL 109 mmol/L citrate), calculated according to the CLSI formula, was added with a sterile pipette (1.85x0.001x25x2 mL).

The re-analysed PT from the citrate-adjusted sample resulted in an INR value of 3.7 (PT 49.1 sec, 18.3%). This INR value was much more plausible.

Conclusion

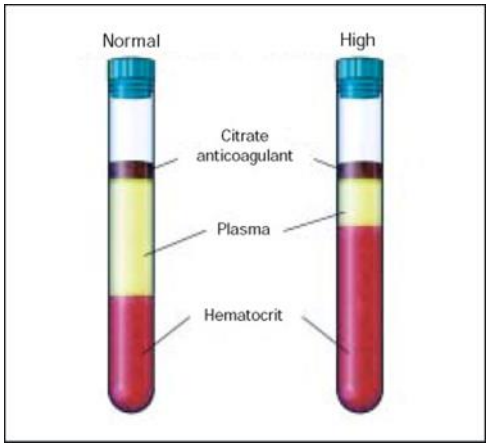
We described a patient with a complex cardiac history and secondary polycythaemia, who had a supra-therapeutic INR of 9.1 with rectal blood present per anum, without other relevant bleeding tendency. At the emergency department an anoscopy was able to detect the presence of haemorrhoids, which might possibly be considered the cause of the rectal blood loss. The unexpectedly high INR value was further explored.

The recommendations to adjust the volume of citrate anticoagulant in blood collection tubes in patients with high haematocrit values are based on several studies that evaluated the effect of sample volume and citrate volume on routine coagulation tests. The current guidelines that recommend adjusting the citrate volume for blood samples with a high haematocrit level are mainly based on indirect studies. A study performed in 2006 compared PT and aPTT results from patient samples with a high haematocrit level with and without citrate adjustment. A statistically and clinically significant difference was found for aPTT and PT results between citrate-adjusted and non-adjusted specimens. Furthermore, they observed an exponential increase in aPTT and PT clotting times if the haematocrit value increased from 55% to 72% [6].

Collection tubes with 3.2% or 3.8% sodium citrate concentrations are available. Literature demonstrates that at a higher citrate concentration (3.8%) the impact of a correctly filled tube, and therefore high haematocrit, is also higher than if a collection tube with 3.2% citrate had been used [9].

As clinicians are not always aware of pre-analytical variables that may influence test results, and a high haematocrit is one of the less frequently considered variables in routine testing, it is very important to pay due attention in the lab to this pre-analytical variable. The case we described is an example of the influence of high haematocrit levels on coagulation testing with unnecessary consequences such as extended hospitalisation and needless examinations. This case report provides a practical example of how to use the CLSI formula and how to carry out the citrate adjustment [8]. It is the responsibility of the laboratory to minimise the risk associated with errors in what is called the total testing process or TTP. This concept classifies diagnostic testing pitfalls into three phases: the pre-analytical phase, the analytical phase, and the post-analytical phase [10]. To prevent post-analytical issues, which are generally related to reporting and interpretation, authorised personnel need to systematically assess and validate the results of laboratory testing on the basis of clinical information and previous test results [11]. In the case under discussion the consequence of post-analytical misinterpretation could have been significant as the patient was being monitored for anticoagulant therapy. This may lead to an incorrect dosage of anticoagulant therapy, which is associated with the risk of thrombosis [12].

Figure 1. Effect of haematocrit on relative volume of citrate anticoagulant in blood collection tube [13].



References

1. Bedair, R.; Iriart, X. EDUCATIONAL SERIES IN CONGENITAL HEART DISEASE: Tetralogy of Fallot: diagnosis to long-term follow-up, Echo Research and Practice. 2019, 6, R9-R23.
2. Viswanathan, S. Thromboembolism and anticoagulation after Fontan surgery, Annals of Pediatric Cardiology. 2016, 9, 236.
3. Swerdlow, S. H.; Campo, E.; Harris, N. L., et al. WHO classification of tumours of haematopoietic and lymphoid tissues: International agency for research on cancer Lyon, 2008.
4. Holbrook, A. M.; Pereira, J. A.; Labiris, R., et al. Systematic overview of warfarin and its drug and food interactions, Archives of internal medicine. 2005, 165, 1095-1106.
5. Yang, L.; Ge, W.; Yu, F.; Zhu, H. Impact of VKORC1 gene polymorphism on interindividual and interethnic warfarin dosage requirement—a systematic review and metaanalysis, Thrombosis research. 2010, 125, e159-e166

6. Marlar, R. A.; Potts, R. M.; Marlar, A. A. Effect on routine and special coagulation testing values of citrate anticoagulant adjustment in patients with high hematocrit values, American journal of clinical pathology. 2006, 126, 400-405.
7. Clinical and Laboratory Standards Institute. Procedures for collection of diagnostic blood specimens by venipuncture; approved guideline. CLSI document GP41-A6:2007, 6th edn. Wayne, PA: Clinical and Laboratory Standards Institute; 2007.
8. Clinical Laboratory Standards Institute, Adcock DM, Hoefner DM, Kottke- Marchant K, MArlar RA, Szamosi DI, Warunek DJ. Clinical Laboratory Standards Institute Collection, transport and processing of Blood Specimens for testing plasma-based coagulation assays and mo-lecular haemostasis assays: Approval guidelines – Fifth edition, Wayne PA: CLSI; H21-A5, 2008.
9. Kitchen, S.; Adcock, D. M.; Dauer, R., et al. International Council for Standardisation in Haematology (ICSH) recommendations for collection of blood samples for coagulation testing, International Journal of Laboratory Hematology. 2021, 43, 571-580.
10. De la Salle, B. Pre-and postanalytical errors in haematology, International journal of laboratory hematology. 2019, 41, 170-176.
11. Lenicek Krleza, J.; Honovic, L.; Vlasic Tanaskovic, J.; Podolar, S.; Rimac, V.; Jokic, A. Post-analytical laboratory work: national recommendations from the Working Group for Post-analytics on behalf of the Croatian Society of Medical Biochemistry and Laboratory Medicine, Biochemia Medica. 2019, 29, 228-261.
12. Favaloro, E. J.; Lippi, G. Post-analytical issues in hemostasis and thrombosis testing, Hemostasis and Thrombosis. 2017, 545-559.
13. Favaloro, E. J.; Funk, D. M.; Lippi, G. Pre-analytical variables in coagulation testing associated with diagnostic errors in hemostasis, Laboratory Medicine. 2012, 43, 1-10.