

Review Article

Pre-analytical Phase in Hemostasis: The Main Anomalies and Means to Correct Them

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Abstract: Pre-analytical errors still represent nearly 70% of all errors occurring in the laboratory, constituting a danger, a waste of time and an additional cost to the patient. The control of the different components of the pre-analytical step is important for the validity of the hemostasis exploration tests. The purpose of our work is to identify the main anomalies of the pre-analytical phase in hemostasis and to propose the means to correct them. We conducted a prospective and descriptive study on the pre-analytical phase of hemostasis. It was in the form of a survey, identifying the main errors related to this phase. It was performed at the Hematology laboratory of the Avicenna Military Hospital of Marrakech and spread over a period of 4 weeks. Our investigation took place at the hemostasis room, which received the collection tubes from the various hospital departments and the blood drawing room (for non-hospitalized patients). The hemostasis room received 400 prescription cards and their corresponding tubes. The parameters related to the prescription file: full name and gender of the patients, were mentioned on all the cards received and they were in conformity with those marked on the corresponding tube. The age of the patients and their clinical and therapeutic informations were mentioned in 73% and 13% of the exam requests, respectively. For the pre-analytical hemostasis parameters related to the blood collection: 63% of samples were taken at the laboratory's blood drawing room, while 37% came from the various hospital departments. Time of the realization of the samples was not mentioned on the cards or on the labels of the tubes. The anticoagulant used for all samples was sodium citrate at a concentration of 3.8%. The filling of the tubes was noncompliant in 22.25%. Registration and triage of the tubes systems were manual. Centrifugation was carried out at a rotation speed of 5000 G for 5 minutes and at a temperature set at 22°C. Hemolyzed samples accounted for 3% of the tubes.

Keywords: Hemostasis, Pre-analytical Phase, Sample

1. Introduction

The management of a sample in biology has three phases linked in time: the pre-analytical, analytical and post-analytical phases. Pre-analytical errors still account for nearly 70% of all errors occurring in the laboratory, posing a danger, a waste of time and an additional cost to the patient [1-3].

Mastery of the various components of the pre-analytical stage is a subject that is still relevant today. It occupies an

important place in the validity of the tests of exploration of the haemostasis. It determines the reliability of results, and it is an important part of quality assurance. This phase includes: a just prescription, information that allows the laboratory to understand and integrate the peculiarities of the patient and its treatment, the optimization of the quality of the sample, the conditions of its transport to the laboratory and its reception, and finally, the preparation of the sample before analysis, with centrifugation, storage, freezing and thawing [4].

The objective of our work is to identify the main anomalies of the pre-analytical phase in hemostasis and propose ways to correct them from the new recommendations improving the daily practices of prescribers, samplers, technicians and biologists.

2. Material and Method

We conducted a prospective, descriptive, analytical and quantitative study on the pre-analytical phase of hemostasis. It was in the form of a survey, identifying the main errors related to this phase. It was performed at the Hematology laboratory of the Avicenne Hospital of Marrakech and spread over a period of 4 weeks from 11/09/2018 to 08/10/2018. Our investigation took place at the hemostasis room, which received the collection tubes from the various hospital departments and the blood drawing room (for non-hospitalized patients). Included in our study were all patients who had a biostatic haemostasis test: for a preoperative assessment, a follow-up report on a known pathology, or a specialized haemostasis report. The information was collected from the prescription vouchers and using a far return. Data entry and analysis were performed with EXCEL software and a descriptive method using simple variables. The total number of tubes examined and the corresponding prescription cards during the study period were 400. The various parameters were measured using calibrated and controlled automatons.

3. Results

During the study period, we received 400 samples for hemostasis assessment. On all prescription cards and their corresponding tubes, were mentioned the first name, family name, and sex of the patient. No errors were reported at this level. However, age and clinical and therapeutic information were not always mentioned. In fact, they were found respectively in 73% and 13% of the cards.

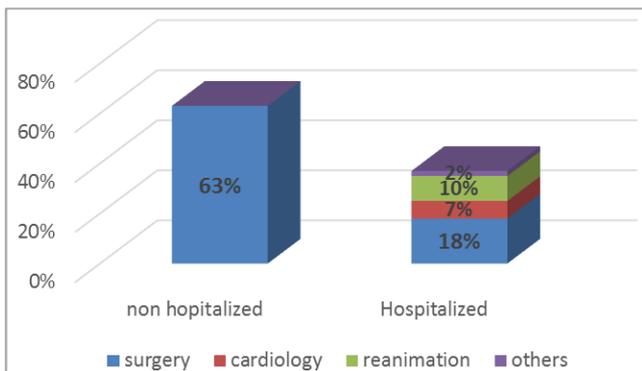


Figure 1. Distribution of balance sheets according to requesting services.

The majority of the tubes received at the laboratory came from non-hospitalized patients (63%), whose samples were taken at the laboratory's blood drawing room. Once completed, the tubes were immediately sent to the hemostasis room and treated without delay. The rest of the tubes were received from the various hospital departments (figure 1). However, the delivery

time to the laboratory was unknown because of the unavailability of information concerning the time of blood collection.

The anticoagulant used was sodium citrate at a concentration of 3.8%. The tubes used were CE marked (European conformity) and the expiry date was respected in all cases. The order of filling of the tubes was as follows: If the haemostasis assessment was requested alone, the citrated tube was taken alone without prior use of a purge tube. If the haemostasis assessment was requested with a series of examinations, citrate tube sampling was performed in 2nd position after no additive tube.

The filling of the tubes varied according to whether the sample was taken at the blood drawing room or in the hospital departments (Table 1).

Table 1. Variation of filling of tubes.

Filling of tubes	Internal blood collection tubes (n=149)	External sampling tubes (n=251)
≥90%	30%	41%
80%	42%	40%
<80%	28%	19%

In all cases, no collection tube received at the hemostasis room was coagulated.

At the hemostasis room, the staff proceeded to the saving, sorting and centrifugation of the tubes, before proceeding to the analytical step. The saving system used was carried out in two stages: The first was manual: The technician proceeded to the identification by giving a number with a marker, to the tube and the corresponding prescription card in the order of arrival of the tubes. The second was computerized: the technician entered the information of each patient on the computer linked to the hemostasis machine, in the order during the manual step.

The triage of the blood collection tubes received was done manually by the technicians. The tubes were sorted according to the examinations requested on the prescription form, in order to be treated later. The check-ups requested for the exploration of haemostasis included orientation assessments, and specialized assessments when necessary (Table 2). The time between receiving the tubes and centrifuging them was 10 to 20 minutes. The centrifuge was programmed to a standard speed of 5000 G and a temperature of 22°C for all sample tubes. The centrifugation time was 5 minutes.

Table 2. Sorting the tubes of samples received according to the balance sheets requested.

Comprise	Number	Percentage
PT	387	97%
INR	121	30%
APTT	258	64%
Fibrinogen assays	15	4%
lupus anticoagulant	3	1%
Protein C et S	3	1%
Antithrombin	3	1%
Factor assays (VIII)	1	0.5%
D-dimer	1	0.5%

After centrifugation, 3% of the samples were hemolyzed. They accounted for 7% of internal samples, and 1% of external ones.

4. Discussion

Any biological analysis can only be done after a correct and accurate identification of the patient. The first and family name and an identifying number (national identity card, entry number for hospitalized patients...) marked on the prescription card, must be identical to those marked on the corresponding tube label. Any tube that does not correspond to its prescription card, must be rejected with communication of the reason for the rejection to the source hospital department, or the blood drawing room.

In our study series, no identification error was objectified. The first and family name marked on the label of each tube were in conformity with those mentioned on the corresponding prescription form. Several studies have evaluated the number of errors concerning this parameter, the percentages of error differ from one study to another, while being relatively low.

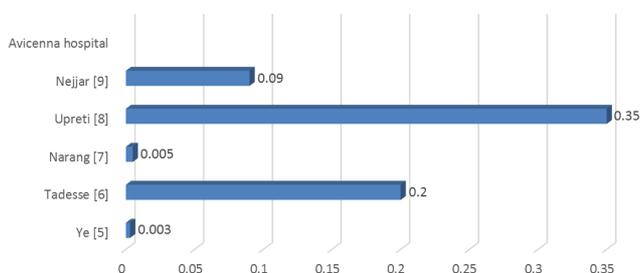


Figure 2. Percentages of identification error from other teams.

The sex of the patients is one of the important factors of physiological variations of the parameters of hemostasis. Its mention on the prescription card is essential. It is obligatory to take it into consideration when interpreting the reports exploring haemostasis [4]. A study carried out in 2018 [6], showed the absence of the mention sex of the patient from the prescription card in 10.3% of the total of the prescription cards received in the laboratory. In our survey, each patient's sex was marked on the corresponding prescription form.

The interpretation of hemostasis assessments is also closely related to age groups. Mention of the age or date of birth of the patient on the prescription form is essential for correct interpretation [10-12]. In the series of Tadesse et al [6], 11.5% of cards did not mention the age of the patient. In our series, age was not mentioned on 27% of the total number of cards received.

Pathological variations or some drugs may alter the results of examinations exploring hemostasis. Therefore, the mention of clinical and therapeutic information on patient prescription cards is essential. This allows the biologist to correctly interpret the results obtained, and also to act by carrying out, if necessary, additional examinations that can provide more details for the diagnosis [1, 4, 13-16]. In the series of Tadesse et al. [6], clinical and therapeutic information was on prescription cards in 70.1% of cases. In our series, they were mentioned only in 13% of cases and absent in 87% of all prescriptions received.

According to The Clinical and Laboratory Standards Institute (CLSI), the French Study Group on Hemostasis and

Thrombosis (GFHT) and European Concerted Action on Thrombosis (ECAT), specimens kept at room temperature (15°C-25°C) for routine haemostasis tests or determination of coagulation factors should be analyzed within 4 hours of collection, except for the quick time (TQ), which has a stability of up to 24 hours, and tests to monitor treatment with unfractionated heparin, for which the delay should not exceed 2 hours [17, 18]. Several studies have demonstrated longer stability for many hemostasis parameters. This can be interesting, for example, when additional coagulation tests are requested or when laboratories must subcontract coagulation tests to a laboratory remote from the sampling site [2, 17-19]. However, further studies are needed to confirm the validity of this eventuality. A cross-sectional study carried out in 2018 in China by Ye et al's team [5], including 1586 laboratories, during a period of one month, showed a rate of 0.001% of time-lapse between the blood collection and the receiving the tubes at the treatment room. This very low rate has been explained by the attention given to samples and their reception over the last 5 years, which has generally minimized the rejections due to pre-analytical errors. In our study, the delay between the collection blood and the arrival of the tubes in the haemostasis room depended on the origin of the tubes: the tubes from the laboratory's blood drawing were immediately sent to the treatment room; the delivery delay of tubes from hospital services was unknown. In the latter case, it is a major problem, which the consequences may be detrimental to the patients, having samples received after exceeding the acceptable time limits (erroneous results). The staff responsible for sampling within the services must be aware and informed of the importance of the time limit for the analysis of haemostasis samples.

According to the GFHT (May 2017), it is recommended to use plastic tubes (polyethylene terephthalate). The expiry date of the tubes must absolutely be respected. It is also important that the quality of the tubes is documented and recognized by CE marking [20]. In our survey all tubes used were in accordance with the latest GFHT recommendations.

The anticoagulant used in was sodium citrate at 3.8% concentration. According to the French (GFHT) and American (CLSI) recommendations, using this concentration is acceptable. However the 3.2% concentration is more recommended [21, 22].

The order in which the tubes are taken is also important, particularly in hemostasis. According to the recommendations of the GFHT and CLSI, the haemostasis sample must be taken in 2nd position after a no additive tube, blood cultures, or a "purge" tube in the case where the haemostasis assessment is requested alone [4, 23]. The order of blood collection of the 149 tubes from the hospital departments was unknown. This makes the evaluation of this parameter impossible and can be an unknown source of error compromising the validity of the results. For the 251 tubes taken from the blood drawing room of the laboratory, the order depended on the analyzes requested: if the haemostasis assessment is requested with a series of examinations, the citrated tube is placed in the 2nd position according to the recommendations; if it is requested

alone, the haemostasis sample was taken without prior use of a "purge" tube. However, this last course remains acceptable according to the GFHT only for routine haemostasis tests and with a non-traumatic clear venipuncture.

The filling of the tubes, whatever its origin, is evaluated

according to the latest recommendations of the GFHT: Recommended filling: to the mark noted on the tube, or more than 90%; Acceptable filling: tube filled to more than 80%; Under-filling: tube filled to less than 80%.



Figure 3. Recommendations for a valid prescription and a valid blood collection.

The under-filling tubes accounted for 22.25% of all tubes received in the hemostasis room. The rate of under-filling tubes from services (28%) was higher than that of tubes received from the blood drawing room (19%). Our rates are high by comparing them to the rates of similar studies [5-7, 9-25].

In contrast, no samples received at the hemostasis room were coagulated. Some teams [5-25] detected the presence of coagulated samples among the sampling tubes. However, the percentages remain below 1%.

The recording and sorting systems during our investigation were mainly manual. The latest recommendations favor the use of automatic systems allowing: Computer registration of patients (identifiers, results of previous assessments, consultations, hospitalization report...); the request for biological examinations without the use of prescription cards; automated labeling (barcodes).

This automation significantly reduces patient identification error rates. For example, the Killeen et al team reported that the introduction of automatic systems in an emergency department reduced identification error rates from 2.56 to 0.49 per 1000 samples [26, 27].

The delay between receiving the tubes and centrifugation was between 10 and 20 minutes for all the tubes received at the hemostasis room. This delay was in line with the recommendations of the GFHT. The temperature of the centrifuge was set at 22°C for all sample tubes. This is in accordance with the recommendations of the GFHT which specified that the temperature of the centrifuges must be between 15°C and 25°C.

The rotation speed and the centrifugation time were programmed at 5000G for 5 minutes for all the sample tubes received. According to the GFHT, it is a rapid centrifugation, having been validated and can be used just for carrying out some tests: prothrombin time (PT), activated partial thromboplastin time (APTT), Fibrinogen assays, D-dimer assays. However, the laboratory has received 10 requests for tests for: Proteins C and S, factor VIII, antithrombin and circulating anticoagulant antibodies. For the latter, a double centrifugation had to be carried out instead of rapid centrifugation according to the GFHT.

The rate of haemolyzed samples was 3% of all tubes received. Similar studies have also reported hemolyzed sample rates of less than 1% [5, 8-24].

In order to improve the quality of haemostasis samples, and to minimize the errors related to the pre-analytical phase, we propose to remind prescribers and samplers the basic conditions for a valid prescription and a haemostasis compliant sample, using a poster (figure 3), which would be displayed at the sampling room and the various services.

In addition, the standardization of acceptance criteria for sampling would make it possible to achieve the desired reliability of results. This is the reason why a proper management of non-compliant samples by the staff of the hemostasis room is required. Any sample that does not comply with the recommendations related to the pre-analytical phase must be rejected, with notification of the cause of rejection to

the source department or the laboratory's sample room.

5. Conclusion

The pre-analytical phase remains the sensitive point of the hemostasis analysis process and its parameters are as important as each other. This is a difficult phase to master because of the large number of stakeholders involved and the diversity of the parameters that make it up. Mastery and efforts to standardize pre-analytical conditions are essential to ensure the quality of hemostasis exploration.

Our study shows that the general progress of the pre-analytical phase in the hematology laboratory of Avicenne Hospital in Marrakech, respects most of the recommendations. However, despite the efforts made by the various stakeholders involved in this process, certain parameters still need to be reinforced and taken into account in order to obtain the desired reliability of the results.

References

- [1] Ellouze R, Guermazi S. Importance of preanalytical step in hemostasis. *Annales de Biologie Clinique*, Vol 71, n°4, p401-407, 2013.
- [2] Mauge L, Alhenc-Gelas M. Stability of coagulation parameters review of available data. *Annales de Biologie Clinique*, n°2, p 141-145, 2014.
- [3] Plebani M et al. Quality indicators to detect pre-analytical errors in laboratory testing. *Clinica Chimica Acta*, vol 432, p. 44-48, Mai 2014.
- [4] Gris JC. Etapes préanalytiques en hémostasie. *EMC – Biologie médicale*, vol 6, n°3, p. 1-7, Jan 2011.
- [5] Ye Y et al. Haematology specimen acceptability: a national survey in Chinese laboratories. *Biochemia Medica*, vol 28, n°3, Oct 2018.
- [6] Tadesse H et al. Errors in the Hematology at St. Paul's Hospital Millennium Medical College, Addis Ababa, Ethiopia. *BMC Research Notes*, vol 11, n°1, Dec 2018.
- [7] Narang V et al. Preanalytical Errors in Hematology Laboratory - an Avoidable Incompetence. *Iranian Journal Of Pathology*, Vol 11, n°2, p. 151-154, 2016.
- [8] Upreti S et al. Types and Frequency of Preanalytical Errors in Haematology Lab. *Journal of Clinical and Diagnostic Research*, Vol 7, n°11, p. 2491-2493, Nov 2013.
- [9] Nejjar N. La phase préanalytique en hémostasie: données de la littérature et enquête au laboratoire d'hématologie à l'HMIMV de Rabat. Thèse Doctorat Pharmacie, université Mohamed V faculté de médecine te de pharmacie-Rabat, n°101, 2010.
- [10] Hézard N. Hémostasie pédiatrique. *Bio Tribune Magazine*, Vol 18, n°1, p. 40-43, 2006.
- [11] Hurtaud-Roux MF, Vincenot A, Lasne D. L'hémostasie en pédiatrie, ses particularités, les principales pathologies hémorragiques et leur gestion. *Anesthésie & Réanimation*, vol 4, n°4, p. 290-299, 2018.

- [12] Lasne D, Hurtaud MF. Particularités de l'hémostase chez le nouveau-né. *Revue Francophone des Laboratoires*, n° 508, p. 72-80, Jan 2019.
- [13] Ozier Y, Cadic A, Dovernge A. Prise en charge des troubles de l'hémostase chez l'insuffisant hépatique. *Transfusion Clinique et Biologique*, vol 20, n°2, p. 249-254, 2013.
- [14] Fattah MA, Shaheen MH, Mahfouz MH. Disturbances of haemostasis in Diabetes Mellitus. *Disease Makers*, vol 19, n°6, P. 251-258, 2004.
- [15] Magnette A et al. Pre-analytical issues in the hemostasis laboratory: guidance for the clinical laboratories. *Thrombosis Journal*, vol 14, n° 1, Dec 2016.
- [16] Blanloeil Y et al. Effets des solutés de remplissage vasculaire sur l'hémostase. *Annales Françaises d'Anesthésie et Réanimation*, vol 21, n°8, p. 648-667, Oct 2002.
- [17] Linskens EA, Devreese KMJ. Pre-analytical stability of coagulation parameters in plasma stored at room temperature. *International Journal of Laboratory Hematology*, vol 40, n°3, p. 292-303, 2018.
- [18] Toulon P et al. Impact of different storage times at room temperature of unspun citrated blood samples on routine coagulation tests results. Results of a bicenter study and review of the literature. *International Journal of Laboratory Hematology*, vol 39, n°5, p. 458-468, Oct 2017.
- [19] Feng L et al. Effects of storage time and temperature on coagulation tests and factors in fresh plasma. *Scientific Reports*, vol 4, n° 1, May 2015.
- [20] Laboratoire de Biologie Médicale, CHU Tours. GFHT recommandations préanalytiques en hémostase, révision octobre 2015 (dernière mise à jour Mai 2017). Disponible sur: https://www.chu-tours.fr/assets/files/pdf/manuel_prelevement/LBM_M_N_DX005_02_Recommandations_pr%C3%A9-analyti.pdf.
- [21] Louati N, Ben Amor I, Daoued G. Effet de l'anticoagulant et de la conservation des échantillons sanguins sur les tests d'exploration de la coagulation. *Journal de l'Information Médicale de Sfax*, n°24, p. 22-23, Oct 2016.
- [22] Samama MM. Le pré-analytique en hémostase. *Bio tribune magazine*, n°10, p XIII, Mai 2004.
- [23] Cornes M et al. Order of blood draw: Opinion paper by the European Federation for Clinical Chemistry and Laboratory Medicine (EFLM) Working Group for the Preanalytical Phase (WG-PRE). *Clinical Chemistry and Laboratory Medicine (CCLM)*, vol 55, n°1, p. 27-31, Jan 2017.
- [24] Shukla DKB et al. Study of pre-analytical and post-analytical errors in hematology laboratory in a tertiary care hospital. *Journal of Medical Science And clinical Research*, vol 4, n°12, p. 14964-14967, Dec 2016.
- [25] Salvagno GL et al. Prevalence and type of pre-analytical problems for inpatients samples in coagulation laboratory. *Journal of Evaluation in Clinical Practice*, Vol 14, n°2, p. 351-353, 2008.
- [26] Lippi G et al. Patient and sample identification. Out of the Maze ?. *Journal of Medical Biochemistry*, vol 36, n°2, p. 107-112, Apr 2017.
- [27] Lippi G et al. Causes, consequences, detection and prevention of identification errors in laboratory diagnostics. *Clinical Chemistry and Laboratory Medicine (CCLM)*, vol 47, n°2, Jan 2009.
- [28] Heyer NJ et al. Effectiveness of practices to reduce blood sample hemolysis in EDs: A laboratory medicine best practices systematic review and meta-analysis. *Clinical Biochemistry*, vol 45, n°13-14, p. 1012-1032, Sep 2012.