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Pre-donation assessment of platelets count among Sudanese blood donors attending central blood bank, Gezira state, Wad Madani, Sudan, 2023

Asad Adam Abbas [1,*] and Soad Fadal Allah Ali [2]

[1] *Department of pathology, Faculty of Medicine, BNNICD, University of Gezira, Wad Madani, Sudan.*
[2] *Department of pathology, Faculty of Medicine, University of Gezira, Wad Madani, Sudan.*

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Abstract

Objective: To estimate platelets count among Sudanese platelets donors attending the central Blood Bank in Wad Madani.

Material and Methods: Venous blood samples were taken from 200 apparently healthy males donors and the platelets count were measured using an automated cell counter (Sysmex KN21).

Results: The study revealed that significant number of donors with abnormal Platelets count specially thrombocytopenia, reactive thrombocytosis occurred in 4 donors with platelet count more than 450 × 10^9 /L.

Conclusion: Complete blood count is very important using automated blood cell counter and must be performed as routine test before donation.

Keywords: Platelets; Thrombocytopenia, Thrombocytosis; Donors.

1. Introduction

1.1. Platelets

Are small, irregularly-shaped anuclear cells (i.e. cells that do not have a nucleus containing DNA), 2-4 μm in diameter, which are derived from fragmentation of precursor megakaryocytes. The average lifespan of a platelet is between 8 and 12 days.

Platelets play a fundamental role in hemostasis and are a natural source of growth factors. Platelets release a multitude of growth factors including platelet-derived growth factor (PDGF), a potent chemotactic agent, and transforming growth factor-β, which stimulates the deposition of extracellular matrix. Both of these growth factors have been shown to play a significant role in the repair and regeneration of connective tissues. Other healing-associated growth factors produced by platelets include basic fibroblast growth factor, insulin-like growth factor-1 (IGF-1), platelet-derived epidermal growth factor, and vascular endothelial growth factor. Local application of these factors in increased concentrations through PRP (platelet-rich plasma) has been used as an adjunct to wound healing for several decades.[1] Platelets are produced in blood cell formation (thrombopoiesis) in bone marrow, by budding off from megakaryocytes. The physiological range for platelet count is 150-400 x 10^9 per litre. Around 1 x 10^{11} platelets are produced each day by an average healthy adult.

* Corresponding author: Asad Adam Abbas

The lifespan of circulating platelets is 7 to 10 days. This process is regulated by thrombopoietin, a hormone usually produced by the liver and kidneys. Each megakaryocyte produces between 5,000 and 10,000 platelets. Old platelets are destroyed by phagocytosis in the spleen and by Kupffer cells in the liver.[2]

Maturation normally takes days to occur. Platelet production is controlled by:

- Negative feedback: the hormone thrombopoietin (TPO) increases platelet numbers. It is internalized by circulating platelets, so the fewer the platelets the less internalization and more free TPO.
- Interleukin-3 (IL-3) and granulocyte-macrophage colony-stimulating factor (GM-CSF)—in combination, these stimulate CFU-megakaryocytes (CFU-MK). [2]

1.2. Platelet functions

The main functions of platelets are adhesion, release reaction, aggregation, procoagulation and tissue repair. [2]

1.3. Thrombus formation

The function of platelets is the maintenance of haemostasis. This is achieved primarily by the formation of thrombi, when damage to the endothelium of blood vessels occurs. On the converse, thrombus formation must be inhibited at times when there is no damage to the endothelium. [2]

The inner surface of blood vessels is lined with a thin layer of endothelial cells that, in normal hemostasis, acts to inhibit platelet activation by producing endothelial-ADPase, noradrenaline, and PGI2. Endothelial-ADPase clears away ADP, a platelet activator, from platelet surface receptors. [2]

Endothelial cells produce a protein called von Willebrand factor, a cell adhesion ligand, which helps endothelial cells adhere to collagen in the basement membrane. Under physiological conditions, collagen does not pass into the bloodstream. However vWF is secreted constitutively into the plasma by the endothelial cells that produce it, or otherwise is stored within the endothelial cells or in platelets. When endothelial damage occurs, platelets come into contact with exposed collagen and vWF, causing a reduction in secretion of endothelium platelet inhibitors. [2] The inner surface of blood vessels is lined with a thin layer of endothelial cells. Under this is a layer of collagen. When the endothelial layer is injured, the collagen is exposed. Then the platelets adhere to collagen and become activated. They are also activated by thrombin (primarily through PAR-1) and ADP receptors (P2Y1 and P2Y12) expressed on platelets. They can also be activated by a negatively-charged surface, such as glass. [2] Platelet activation further results in the scramblase-mediated transport of negatively-charged phospholipids to the platelet surface. These phospholipids provide a catalytic surface (with the charge provided by phosphatidylserine and phosphatidylethanolamine) for the tenase and prothrombinase complexes. [2]

1.4. Shape changes of platelets

Activated platelets change their shape to become more spherical, and pseudopods form on their surface. Thus they assume a stellate [star-like] shape. [2]

1.5. Granule secretion

Platelets contain alpha and dense granules. Activated platelets excrete the contents of these granules into their canalicular systems and into surrounding blood. There are two types of granules:

- dense granules (containing ADP or ATP, calcium, and serotonin)
- α-granules (containing platelet factor 4, transforming growth factor-β1, platelet-derived growth factor, fibronectin, B-thromboglobulin, vWF, fibrinogen, and coagulation factors V and XIII).[2]

1.6. Thromboxane A2 synthesis

Platelet activation initiates the arachidonic acid pathway to produce TXA_2. TXA_2 is involved in activating other platelets. [2]

1.7. Adhesion and aggregation

Platelets aggregate, or clump together, using fibrinogen and vWF as a connecting agent. The most abundant platelet aggregation receptor is glycoprotein (GP) IIb/IIIa; this is a calcium-dependent receptor for fibrinogen, fibronectin, vitronectin, thrombospondin, and von Willebrand factor (vWF). Other receptors include GPIb-V-IX complex (vWF) and GPVI (collagen). Activated platelets will adhere, via glycoprotein (GP) Ia, to the collagen that is exposed by endothelial damage. Aggregation and adhesion act together to form the platelet plug. Myosin and actin filaments in platelets are stimulated to contract during aggregation, further reinforcing the plug. Platelet aggregation is stimulated by ADP, thromboxane, and α2 receptor-activation, but inhibited by other inflammatory products like PGI2 and PGD2. Platelet aggregation is enhanced by exogenous administration of anabolic steroids. [3]

1.8. Platelet transfusion

Is a lifesaving procedure that is carried out to prevent bleeding or stop ongoing bleeding in patients with low platelet counts or functional platelet disorders. Platelets play an integral role in hemostasis through their response to vascular injury. The relevance of platelet component therapy was better understood in the 1950s and 1960s when severe and fatal hemorrhagic complications of chemotherapy in leukemia were studied. There are minimum thresholds at which platelets are transfused in these patients, as not all low levels of platelet warrants a transfusion. Platelet is a scarce resource as processing, preparing, and transfusing them requires a great deal of precision and effort to maintain a certain quality. This activity highlights the use of platelet transfusion by the interprofessional team.

Platelet concentrates (PC) are widely used to support patients with severe thrombocytopenia. These could be patients with hematologic malignancy, bone marrow failure, or other immune and non-immune causes of platelet destruction, though rare cases could warrant transfusion with normal platelet counts. Platelet is a scarce resource, partly because of their short shelf life of 5 days; it is classified in the World Health Organization's (WHO) list of "Essential Medicine.

1.9. Platelet concentrates

These are harvested by cell separators or from individual donor units of blood. PC can be prepared from whole blood or by apheresis. 6 whole blood unit-derived platelets equal one apheresis platelet. The shelf life of a PC is five days, within which it must be used. The normal dose of platelet transfused is calculated as 10 to 15 ml/kg of the patient. They are stored at room temperature. Platelet transfusion is used in patients who are thrombocytopenic or have disordered platelet function and who are actively bleeding (therapeutic use) or are at serious risk of bleeding (prophylactic use). [4]

For prophylaxis, the platelet count should be kept above $5 – 10 \times 10^9$ /L unless there are additional risk factors such as sepsis, drug use or coagulation disorders for which the threshold should be higher. For invasive procedures (e.g. liver biopsy or lumbar puncture) the platelet count should be raised to above 50×10^9 /L. For brain or eye surgery the count should be > 100×10^9 /L. Therapeutic use is indicated in bleeding associated with platelet disorders. In massive haemorrhage the count should be kept above 50×10^9 /L.

Platelet transfusions should be avoided in autoimmune thrombocytopenic purpura unless there is serious haemorrhage. They are contraindicated in heparin - induced thrombocytopenia, thrombotic thrombocytopenic purpura and haemolytic uraemic syndrome. [4]

Refractoriness to platelet transfusions is defined by a poor platelet increment post – transfusion (< 7.5×10^9 /L at 1 hour or < 4.5×10^9 /L at 24 hours). The causes are either immunological (mostly HLA alloimmunization) or non – immunological (sepsis, hypersplenism, DIC, drugs). Platelets express HLA class I (but not class II) antigens and HLA - matched or cross - match - compatible platelets are needed for patients with HLA antibodies. Platelet transfusions are likely to be reduced with the introduction of direct stimulators of platelet production (e.g. romiplostim or amino acid group). [4]

1.10. Indications

Normal platelet count in humans ranges from 150,000 to 450,000 cells/microliter. Platelet transfusion is mainly indicated to treat or prevent bleeding in patients with thrombocytopenia or platelet function disorder.

1.11. Platelet transfusion Threshold in Bleeding Patients

- <50,000 cells/microliter in severe bleeding, including disseminated intravascular coagulation (DIC)
- <30,000 cells/microliter when bleeding, not life-threatening, or considered not severe
- <100,000 cells/microliter for bleeding in multiple trauma patients or patients with intracranial bleeding. [5]

1.12. Prophylactic Transfusion Threshold:

Prophylactic platelet transfusion is indicated below a specific threshold and is indicated before specific procedures or to prevent spontaneous bleeding. These include the following:

- To prevent spontaneous bleeding - transfuse at less than 10,000 cells/microliter; some recommend less than 5,000 cells/microliter
- Before neurosurgery or ocular surgery - less than 100,000 cells/microliter
- Before major surgery - less than 50,000 cells/microliter
- In DIC - less than 50,000 cells/microliter
- Before central line placement - less than 20,000 cells/microliter
- Before epidural anesthesia - less than 80,000 cells/microliter
- Before bronchoalveolar lavage (BAL) – 20,000 to 30,000 cells/microliter
- Before endoscopic procedures - less than 50,000 cells/microliter for therapeutic procedures; <20,000 cells/microliter for low-risk diagnostic procedures
- Vaginal delivery platelet transfusion is considered at less than 30,000 cells/microliter, and when traumatic delivery, then less than 50,000 cells/microliter. [6]
- Before lumbar puncture - less than 20,000 cells/microliter in patients with hematologic malignancies and 40,000 to 50,000 cells/microliter in patients without hematologic malignancies
- Platelet transfusion is not routinely indicated prior to bone marrow biopsy, peripheral or central catheter insertion, traction removal of tunneled central venous catheters, and cataract removal. [5]

1.13. Platelet Transfusion in Specific Settings

- Idiopathic thrombocytopenic purpura (ITP) - transfusion is avoided unless severe bleeding is present.
- Malignancy and chemotherapy - In most cancers, platelet transfusion thresholds are as indicated above, except in acute promyelocytic leukemia, in which there is increased bleeding risk. Hence transfusion is indicated at counts less than 30,000 cells/microliter. chemotherapy is carried out at counts less than 20,000 cells/microliter. [7]
- Cardiac surgery - Patients undergoing cardiac surgery get exposed to a blood-pumping circuit, which activates platelets that get destroyed once back in circulation; hence even at normal counts, platelet transfusion is indicated during cardiac surgery.
- Inherited and acquired platelet disorders like Glanzmann thrombasthenia, Bernard-Soulier syndrome, and other congenital platelet defects, acquired platelet disorders like patients with uremia or drug-induced platelet dysfunction. In these situations, platelet transfusion is indicated only when bleeding Is present. [5]
- For pediatric patients, transfusion indications are otherwise similar in older infants and children compared to adults, as demonstrated by the PLADO study, except in the following situation. We transfuse platelets when the platelet count is: [8]
    - <30,000 cells/microliter in neonates without any bleeding or symptom and failure to produce platelet.
    - <50,000 cells/microliter in an infant with active bleeding or undergoing an invasive procedure. For the same situation in a premature infant, we transfuse at less than 100,000 cells/microliter. [9]
    - Patient undergoing extracorporeal membrane oxygenation (ECMO) and platelets <100,000 cells/microliter.

1.14. Contraindications

The only agreed upon contraindication to platelet transfusion is thrombotic thrombocytopenic purpura (TTP) due to the increased risk of thrombosis, although studies on outcomes and mortality have shown mixed results. [10] Platelet transfusion is reserved for life-threatening bleeding only.

Heparin-induced thrombocytopenia (HIT) is another condition where platelet transfusion may increase the risk of thrombosis, but recent studies have shown no risk association. [11] In HIT, transfusion is reserved only for pre-procedure or surgery, and in severe bleeding, prophylactic transfusion, however, is not indicated. [5]

Objectives

General objective

To Assess the Platelet Count Among Sudanese Blood Donors Attending the Central Blood Bank.

Specific objectives

- To measure Platelet count for donors using automated machine (Sysmex).
- To detected any abnormalities of the in apparently healthy Platelet donors (thrombocytopenia & thrombocytosis).

2. Material and methods

2.1. Study area

The study was carried out in the Wad Madani Central blood bank. Wad Madani is the capital of Gezira state, it is considered one of the largest states in Sudan with an area of 35.304 km and population of 4 million. The Central Blood Bank provide blood donation services to 4 governmental hospitals and other private hospitals in Wad Madani. About 1600 to 1700 donors attend the central blood bank monthly. Different types of blood components (whole blood, packed red cells, platelets, fresh frozen plasma) are prepared from whole blood using large refrigerated centrifuges. All donors are selected according to the accepted criteria for donation including age , weight ,physical and medical examination and screening for viral infections (hepatitis B, C and HIV) and the test for syphilis . Haemoglobin level assessment is performed by copper sulphate method and donors are reported as fit for donation if a drop of blood sinks in a copper sulphate solution, of a certain specific gravity.

2.2. Study population

Apparently healthy male Platelet donors attending the Central Blood Bank (200 donors).

2.3. Selection criteria

Donors were selected according to the accepted criteria for donation.

- Age between 18- 60 years.
- Weight : 50 Kg (110 pounds) and more.
- Haemoglobin : 12.5 g/dl.- 17.5 g/dl

Donors were selected with clinical examination (abdominal, cardiopulmonary), pulse and blood pressure were measured, and VDRL, hepatitis B, C and HIV were screened. [12]

2.4. Exclusion criteria

- All donors should be clinically in a good health, subject with any disease symptoms and signs should be excluded.
- Any person taking medications.

2.5. Study design

Descriptive, prospective cross sectional study was conducted in Wad Madani central blood bank, during the period from March 2023 to April 2023.

2.6. Sample collection

A total of 200 apparently healthy adult male donors were screened for Platelet count. This analysis was conducted at the Wad Madani central blood bank. Venous blood samples were taken from an antecubital vein by a 5ml syringe. The site of collection was cleaned using 70% alcohol and left to dry. An elastic tourniquet was applied if needed to the arm for a period not exceeding one minute to avoid haemoconcentration. 2.5 ml of blood was taken into a container with 0.05ml (K2 EDTA) as an anticoagulant with a concentration of 1.5- 2.2 mg/ml and then the sample gently mixed. The blood samples were tested within 2 hours of sample collection using an automated blood cell counters (Sysmex KN21 analyzer) with a flow cytometry using a laser light to perform Platelet counts (PLTs). It is calibrated by a standardized commercially prepared calibrators.

2.7. Data analysis

All data collection from practical and questionnaires survey was entered in Microsoft office excel. Then the result analyzed by Statistical Package for Social Sciences (SPSS) program version 20, across tab correlation was done.

Statistical analysis: The results were analyzed using statistical software package of social sciences (SPSS) version 17 and descriptive data were expressed as means.

2.8. Ethical clearance

Ethical clearance was obtained from the University of Gezira ethical committee and blood bank authority. Verbal informed consent was obtained from all donors.

3. Results

The study included a total of 200 apparently healthy Sudanese male platelet donors. The mean platelet level was found to be 207.1 +/- 63.41 standard deviation with maximum value 631 and minimum value 17, with 33 cases ranged from 17 to 149, 163 cases ranged from 150 to 449 and 4 cases more than 450.

Table 1 Mean, minimum, maximum values and standard deviation (SD) for the platelets values in 200 apparently healthy Sudanese male donors

	Number of sample	Minimum value	Maximum Value	Mean value	Standard Deviation
PLTs	200	17 x 10^9 /L	631 x 10^9 /L	207.1	63.41

4. Discussion

Platelets are a blood component that still does not have a substitute as their functions are complex. Platelet transfusion is a lifesaving procedure that is carried out to prevent bleeding or stop ongoing bleeding in patients with low platelet counts or functional platelet disorders. The mean platelet level was found to be 207.1 +/- 63.41 standard deviation with maximum value 631 and minimum value 17, with 33 cases ranged from 17 to 149, 163 cases ranged from 150 to 449 and 4 cases more than 450. Isolated thrombocytopenia from mild, moderate to severe was observed in 33 cases (16.5%) may be due to asymptomatic parasitism (e.g. Malaria).

Reactive thrombocytosis occurred in 4 donors with platelet count more than 450 (2% of cases) accompanied by low MCV and low MCH (suggestive of iron deficiency) and leucocytosis which the one of causes of thrombocytosis.

5. Conclusion

The study revealed that significant number of abnormal Platelets count specially thrombocytopenia. Complete blood count is very crucial and must be performed as routine test before donation.

Abnormal Platelet counts (i.e. less than 150 x 10^9/l or more than 450 x 10^9/l may be due to an underlying disease process. Low platelet may be due to asymptomatic diseases (e.g. Malaria, hypersplenism). High platelet counts can also be associated with iron deficiency. Transfusion services should ensure that, where abnormal platelet counts are identified as part of routine donation testing, apheresis technology should be used instead of the old technique (whole blood preparation).

Compliance with ethical standards

Acknowledgments

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Disclosure of conflict of interest

The authors do have not any conflicts of interest in this case report and any financial resources.

Statement of ethical approval

Ethical approval was obtained from the University of Gezira ethical committee and blood bank authority.

Statement of informed consent

Informed consent and verbal permission were obtained from the donors before the submission of this article.

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