

DIFFERENCES IN HEMOGLOBIN LEVELS WITH SIT AND SITTING BLOOD DRAWINGS TECHNIQUES

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ABSTRACT

Introduction – Blood is one of the most important organs in human body. Blood samples are usually obtained by drawing blood in a vein known as phlebotomy. Blood specimen collection generally can be done with various postures including lying on a bed or sitting down. Patient should sit comfortably in a suitable chair or lying down when taking blood because posture can cause significant changes in blood cell components such as hemoglobin.

Purpose – This research aims to determine whether there's a difference between hemoglobin levels with sitting and lying blood sampling techniques

Methodology/Approach – The type of research used is comparative cross-sectional, conducted at the Hematology Laboratory IKesT Muhammadiyah, Palembang. Samples were taken from 30 respondents who entered the inclusion criteria. The research results were processed using Statistical Product and Service Solution (SPSS) program. Research data on differences in hemoglobin levels with the sitting and lying blood sampling technique were analyzed using the Dependent T test.

Findings – The average of hemoglobin level in sitting position obtained was 12,7mg/dL, while in the lying position was 12,3mg/dL. The result of the dependent-t test, it was known the signature value was $P=0,000$. The P value obtained is $<0,05$, so it can be concluded that there's a significant difference between sitting and lying blood sampling techniques.

Originality/ Value/ Implication – Difference between this research and other is that using a comparative cross-sectional method where the data processed using SPSS program. The component researched were also different, namely the hemoglobin levels.

Keywords: hemoglobin, sitting position, lying position

INTRODUCTION

Laboratory is a place equipped with various tools, equipment, and chemicals (reagents), to carry out experimental work, research activities, and inspection procedures. Medical Laboratory is one part of the laboratory which is equipped with various biomedical instruments, equipment, materials and reagents (chemicals) to carry out various laboratory examination activities using biological specimens (whole blood, serum, plasma, urine, feces, etc.) (Mardiana, 2018).

Blood is one of the most important organs in the body for humans. Blood contains various components, both liquid components in the form of blood plasma and solid components in the form of blood cells. Hematology is a medical science that studies blood and blood-forming tissues (Firani, 2018). Blood is a special body fluid that

delivers necessary substances to body cells, such as nutrients and oxygen and transports waste products from the same cells (Singhal, 2013).

One of the elements in the human body that plays a role in the body's working mechanism is blood where all organs of the body are connected by blood through blood vessels. Therefore, blood can be a reflection of the state of the body, both in good health and in illness. Blood is still the most reliable source of medical diagnosis. This is because there is a lot of important information contained in the blood (Anamisa, 2015).

The main function of blood especially red blood cells called erythrocytes is to transport hemoglobin, and so carry oxygen from the lungs to the tissues. In addition to transporting hemoglobin, blood also has another function, namely hemoglobin in human blood is a kitchen acid (as in most proteins), so hemoglobin is responsible for most of the transport power around the blood (Anamisa, 2015).

Blood consists of two components, namely a liquid component called plasma and a solid component, namely blood cells. Cells are three types of blood, namely erythrocytes, leukocytes and platelets. Erythrocytes have a very important function in the human body. The most important function of erythrocytes is O₂ and transports oxygen. CO₂ between the lungs and tissues. An erythrocyte protein, namely hemoglobin (Hb) plays an important role both in the transport process (Gunadi et al, 2016).

In the blood there are blood cells and a fluid called blood plasma which contains various nutrients and other substances. About 55% of blood is a component of plasma, the remaining 45% is a component of red blood cells. Most blood cell components are red blood cells or erythrocytes, which is 41%. The volume of blood cells to the total volume of blood is called hematocrit (Hct). More than 99% of the hematocrit is formed by crosses.

Red blood cells are the main part of blood cells, because they are the most numerous compared to other blood cells. In the fetus (fetus) red blood cells are formed in the liver and spleen. After the baby is born, red blood cells are formed in the bone marrow. In adults, every milliliter of blood contains approximately five million red blood cells, the number of red blood cells becomes more when the person lives in the highlands (mountains). This is because oxygen in the highlands is low, so the body must make more red blood cells to bind more oxygen. This situation is an adaptation of the body to the environment (Abdullah, 2007). Erythrocytes are unique among mammalian cells. They do not contain a nucleus or subcellular metabolic structure, but they persist for 3 to 7 months. During their lifetime, erythrocytes travel thousands of miles through tubes of various sizes while delivering oxygen to the

tissues. The survival of almost all other cells in the body depends on the proper functioning of erythrocytes. They travel as individual cells, containing a concentrated solution of hemoglobin surrounded by a membrane. Despite their apparent simplicity, they have limited (but necessary) metabolic capabilities. They are exposed to high oxygen content and come into contact with various endogenous and exogenous chemicals. Their ability to survive depends on the ability of the membrane to remain intact and flexible (Smith, 1987).

Leukocytes (white blood cells [WBC]) are nucleated and translucent except for colored cells that protect the body from foreign invaders and abnormal cells and can therefore leave the blood and migrate to tissues where they carry out various immune-related activities, while other blood cells do not leave the blood vessels. Morphologically, this nucleus may be in one section or several lobes or segments. Young cells have horseshoe-shaped nuclei that become multilobed as these cells age. Based on the presence or absence of large cytoplasmic organelles called granules (or vesicles), leukocytes are classified into granulocytes (i.e. neutrophils, eosinophils, and basophils) and agranulocytes (i.e. lymphocytes and monocytes). Most of them have a short lifespan (usually a few days), but monocytes can survive for several months and some lymphocytes can live for years. Granulocytes have a lobulated nucleus and visible cytoplasmic granules, whereas agranulocytes have a non-lobed nucleus and contain only fine granules that are rarely visible under an optical microscope. Of these, granulocytes, which are also called polymorphonuclear leukocytes (PMNs), and especially neutrophils which are the most abundant type of PMN, are the most abundant nucleated cells in the blood, constituting about 60-70% of all circulating leukocytes (Chmielewski, 2018). Like erythrocytes, leukocytes are produced constitutively throughout adult life from hematopoietic stem cells in the red bone marrow; they are released into the circulation where they do their job, and are then removed from the blood by the liver and spleen. Unlike erythrocytes, these large, nucleated, translucent cells fulfill an important protective function. They are often capable of extravasation (diapedesis) and phagocytosis and are highly specialized to defend the body against various pathogens, such as bacteria, viruses, fungi, parasitic protozoa, worms, and other harmful factors such as tumor cells or foreign substances. In general, leukocytes are motile and very flexible. Most of these cells are found in body tissues, not in the bloodstream. Several specific molecules released by damaged, abnormal, and dead cells, or by foreign invaders, attract leukocytes by chemotaxis to sites of injury, infection, and inflammation (Chmielewski, 2018).

Platelets are small anucleated cells originally derived from the hematopoietic lineage through megakaryocytes. The production of platelets from megakaryocytes is a systematic and orderly process thought to occur in the bone marrow or, as has been recently demonstrated, in the lungs. Due to most of the extreme shearing forces, the platelets are exposed within the vessels as well as the limitations imposed on the platelets due to the absence of a nucleus; Platelet lifespan is limited to 5 to 7 days after formation and separation from megakaryocytes. While some laboratories have recently demonstrated that it is possible for platelets to split into several smaller functional platelets under certain experimental conditions using the

transcription machinery within the platelets, this process is rarely observed outside controlled conditions in the laboratory, and is important. In the normal physiology of blood vessels remains unclear. During their normal life cycle, platelets decrease in size so that young platelets are much larger than older platelets. At the end of their life in the vessel or after full activation of platelets and incorporation into clot-forming in the vessel, they are removed from the vessel by neutrophils and macrophages and transported to the spleen to be excreted from the body (Holinstat, 2017).

Platelets were discovered by Giulio Bizzozzero in 1882, but for decades the dynamic and multifunctional nature of platelets has remained an area of interest only for biologists. Anucleate, discoid platelets are the smallest blood particles that reveal their dynamics through their morphology. Primarily they are related to hemostasis, which initiates blood clotting. Although very dynamic, they usually prefer to remain in an inactive state and are activated only if a blood vessel is damaged. But hemostasis or blood clotting is not the only function of platelets; rather it is used in several multifunctional attributes that monitor body homeostasis. Its high sensitivity to various disease states has finally established it as one of the most accessible markers. While maintaining interactions with leukocytes and endothelial cells, it restores its behavior as an important inflammatory marker (Ghoshal, 2014).

Blood samples are usually obtained by taking blood in a vein, also known as phlebotomy. According to the WHO, phlebotomy - drawing blood - has been practiced for centuries and is still one of the most common invasive procedures in healthcare. Each step in the phlebotomy affects the quality of the specimen and is therefore important to prevent laboratory errors, patient injury, and even death.

The most common methods today for blood collection are: venipuncture - drawing a venous blood sample using a needle and skin prick - piercing the skin (usually in the finger or heel) for a small amount of capillary blood (skin prick also referred to as a skin stick, capillary pull, stick finger, and heel stick.) (Thimesch, 2018).

The three areas most frequently used in selecting veins are the median cubital, cephalic, and basilic veins. The median cubital vein is located in the middle of the antecubital area. This vein is the largest of the three main veins and is more deeply embedded in the arm. This vein is located near the tendon, usually the first choice when performing venipuncture. Outside the thumb of the antecubital area. This vein is more prominent in obese patients. It also tends to be the smallest of the three main veins, but is still accessible. The basilic vein is this vein located on the little side of the arm. This vein is the third choice when performing this procedure. venipuncture. Caution should be exercised when using the basilic vein because it is located above, or close to, the artery (Thimesch, 2018).

The venipuncture procedure requires knowledge and skills to perform. Each phlebotomist generally establishes a comfortable routine for him (Medtexas, 2007). In general, ethylenediaminetetraacetic acid (EDTA) specimen collection by venipuncture can be performed with the patient in a variety of postures including lying in bed, after walking through outpatient services and sitting right before the test or sitting for long periods of time (Oliveira, 2017).

Clinical and Laboratory Standards Institute (CLSI) document H03-A6, renamed standard GP 41-A6, currently recommends that blood specimens should be collected from patients sitting comfortably in an appropriate chair or reclining, but does not provide specifications regarding supine or positioning. standing and permanent time in a certain position. Posture can affect the concentration of some blood constituents because of the decrease in plasma volume that occurs with the change from lying to standing, it is conventionally assumed that staying supine for long periods of time can be associated with consistent hemodilution. On the other hand, standing posture can be a cause of blood concentration due to the influence of gravity and hydrostatic pressure which causes ultrafiltration of plasma and small molecules in the interstitial space (Oliveira, 2017). The change from the supine to sitting position causes a clinically significant increase in hemoglobin, hematocrit and red blood cell count (Oliveira, 2017).

Based on the background above, the problem can be formulated as follows:

Is there a difference in hemoglobin levels with sitting and lying down blood collection techniques?

LITERATURE REVIEW

Hemoglobin is a non-protein tetrameric erythrocyte binding molecule, which is a compound called porphyrin heme iron. Hemoglobin has two important transport functions in the human body, namely the delivery of oxygen from respiratory organs to peripheral tissues and transport of carbon dioxide and various protons from peripheral tissues to respiratory organs and then excreted to the outside (Kosasi, 2014). Hemoglobin is a protein in red blood cells that functions to carry oxygen from the lungs to the rest of the body. Hemoglobin can be increased or decreased. A decrease in the level of hemoglobin in the blood is called anemia. Anemia is caused by many factors including bleeding, low nutrition, low iron levels, folic acid, vitamin B12, while an increase in hemoglobin levels in the blood is called polycythemia. Symptoms that occur when high hemoglobin is almost not found, are only known during hemoglobin examination (Tutik and Ningsih, 2019).

Hemoglobin examination is one of the routine blood tests that is most often carried out by every laboratory. The examination of hemoglobin levels can be determined by several methods, namely the Sahli method, the cyanmethemoglobin method manually and automatically.

The most hemoglobin examination method is the Sahli method, in the Sahli method, hemoglobin is hydrolyzed with HCL into brown acid hematin, the color formed is compared to the standard color. The color change of hematin acid is carried out by diluting it, so that the color is the same as the standard color. This method is not good because not all hemoglobin can be converted into acid hematin such as carboxyhemoglobin, methemoglobin and sulfhemoglobin. The results of the examination are influenced by subjectivity factors, the standard color fades, irradiation, the error factor reaches 5% -10%.

Another method that is widely used in clinical laboratories is the cyanmethemoglobin method, for clinical purposes of measuring hemoglobin levels, the cyanmethemoglobin method is easy to perform and the results are more accurate than the Sahli method.

The cyanmethemoglobin method is a reference method for hemoglobin estimation, all types of hemoglobin can be measured except sulfhemoglobin, the error factor is $\pm 2\%$, the cyanmethemoglobin method is still widely used in several hospitals and health centers. The principle of cyanmethemoglobin examination is heme (ferrous) is oxidized by potassium ferricyanide to (ferric) methemoglobin then reacts with cyanide ion methemoglobin to form brown cyanmethemoglobin, absorbance is measured by colorimeter or spectrophotometer at 540 nm.

How to check hemoglobin cyanmethemoglobin levels using a napkin solution with a potassium-iron composition that binds heme (ferrous) with (ferric) methemoglobin, converts methemoglobin cyanide ions into cyanmethemoglobin, KH_4PO_4 regulates the pH of the solution (7.0-7.4) and non-ionic detergent function Erythrocyte lysis, thus High leukocyte counts can cause cloudiness and interfere with spectrophotometer readings. Turbidity can also be caused by hyperlipemia and the presence of globulins. Turbidity due to leukocytosis causes the absorbance measurement to increase significantly and an increase in hemoglobin levels is incorrect (Norsidahwahdah, 2015).

According to the Indonesian Ministry of Health, hemoglobin functions include; Regulate the exchange of oxygen with carbon dioxide in the body's tissues, take oxygen from the lungs and then carry it throughout the body to be used as fuel, carry carbon dioxide from body tissues as a result of metabolism to be disposed of, to determine whether a person lacks blood or not, can be determined by measuring hemoglobin levels. A decrease in hemoglobin levels than usual means there is a blood deficiency called anemia (Arif Syaiful, 2017).

The normal limit value of Hb according to the World Health Organization 2001 is for ages 5-11 years $<11.5 \text{ g/dL}$, age 12-14 years 12.0 g/dL , 15 years for women $> 12.0 \text{ g/dL}$ and men -male $> 13.0 \text{ g/dL}$. (Gunadi, et al. 2016).

Hb levels in the blood can be influenced by several factors, one of which is physical activity. Physical activity by humans will affect the increase or decrease in hemoglobin levels in the blood. Physical activity is divided into light activity, moderate physical activity, and strenuous physical activity. Physical activity can affect Hb levels of moderate to moderate intensity physical activity, heavy. Changes in Hb levels through moderate to heavy physical activity are hypothesized to occur due to changes in plasma volume, changes in pH, and intravascular hemolysis. When doing physical activities such as sports will increase high metabolic activity, where acid is produced in the form of hydrogen and more lactic acid ions, this will cause a decrease in pH. The affinity between oxygen and hemoglobin decreases when the blood pH is low. between oxygen and decreased hemoglobin, hemoglobin will release more oxygen thereby increasing oxygen delivery to muscles. As a form of adaptation of the body to moderate \pm heavy intensity activities, there can be changes in blood plasma volume where plasma volume will decrease and will cause hemoglobin levels in the blood to increase, besides that when doing moderate \pm heavy intensity activities, the body needs more oxygen. , to balance the body's need for oxygen to carry out erythropoiesis which will also make Hb levels increase.

Hemoglobin levels are influenced by several factors such

as age and gender, living in the highlands, smoking, physical activity and nutrition. Physical activity and daily exercise or sports that a person does can affect hemoglobin levels including sitting and lying positions at the time of blood collection. In individuals who routinely do physical exercise, the hemoglobin level will increase slightly, while in people who carry out physical activity with heavy intensity which is carried out continuously as done by construction workers (Gunadi, et al. 2016). Hemoglobin levels are also influenced by various other factors, including age, gender, iron intake (nutritional status), demographic conditions (beach and mountains), lifestyle (alcohol, caffeine), diet and chronic diseases (malaria, hookworm infection, etc.) (Nurdiana. 2015). The blood sampling stage includes three stages, namely pre-analytical, analytical and post-analytic stages.

Pre-analytic is the stage of starting to prepare patients, receiving specimens or samples, giving specimen identities, taking specimens, storing specimens, filling out an examination request form (Ginting, 2019).

At this stage, where it is checked whether the patient's identity, sender's identity, laboratory number, date of examination, and request for examination are complete and clear. As well as whether all examination requests have been marked, before carrying out the examination it is necessary to pay attention to the identification and recording of correct patient data.

This stage includes patient preparation, which checks whether the patient's preparation is in accordance with the requirements and makes written instructions for patient preparation for each laboratory examination.

The next stage is the collection and acceptance of patients where it is checked whether the specimen is collected correctly, taking into account the type of specimen.

The third stage is specimen handling, where it is checked whether the processing of the specimen is carried out according to the requirements, whether the handling of the specimen is correct for special examination. And the last stage is the preparation of tools and materials.

The analytical stage is the stage starting from processing specimens, calibrating laboratory equipment, to testing accuracy. This stage includes the preparation of reagents/media by checking whether the reagents/media meet the requirements, whether the expiration period has not been exceeded, whether the dilution method is correct and whether the solvent (aquadest) meets the requirements. The next stage is pipetting reagents and samples by checking whether all laboratory equipment used is clean, meets the requirements, whether the pipettes used have been calibrated and whether the pipette is properly pipetted (Ginting, 2019).

Laboratory activities carried out at the post-analytic stage, namely before the examination results are submitted to the patient, include writing results, interpreting results, reporting results (Siregar et al, 2018).

Blood sampling by venipuncture for various laboratory analyzes is one of the most invasive common procedures, yet at the same time the most underrated procedure in a hospital setting. Although venipuncture is considered a safe procedure and very easy for patients, several studies have shown that venipuncture has inherent risks. That is, even if the staff is trained with the most advanced instruments in the clinical laboratory, an accurate analysis cannot be performed, unless the biologic material is adequately collected. When performing a venipuncture

without adhering to good phlebotomy recommendations, a number of complications can occur, with in vitro hemolysis being very common (Milutinović, 2015). Although the larger and fuller median cubital and cephalic veins of the arm are most commonly used, The wrist and hand can also be used for venipuncture. Corrective techniques are used when there is no return of blood flow. After two failed attempts, the rule for the Phlebotomist is to call another Phlebotomist to perform venipuncture. However, you should try to successfully get a sample before giving up. The proper procedure to do this is called Corrective Technique (Medtexx, 2007).

METHOD

In general, this research has the aim of knowing the difference in hemoglobin levels with the technique of taking blood sitting and lying down and the specific objectives are to measure hemoglobin levels with a sitting blood collection technique, measure hemoglobin levels with a lying blood collection technique and measure differences in hemoglobin levels with a sitting and lying blood sampling technique.

This study was conducted on 60 samples examined at the Hematology Laboratory of IkesT MP in March – April 2021.

This type of research is a quantitative research that emphasizes analysis on numerical data (numeric). This research uses quantitative research method with sampling technique using non-probability sampling technique. Non-probability sampling is a sample collection technique that does not provide equal opportunities for each population or group of people to be selected as samples. Determination of the sample using purposive sampling as many as 30 samples with 2 treatments (Siswanto, 2018).

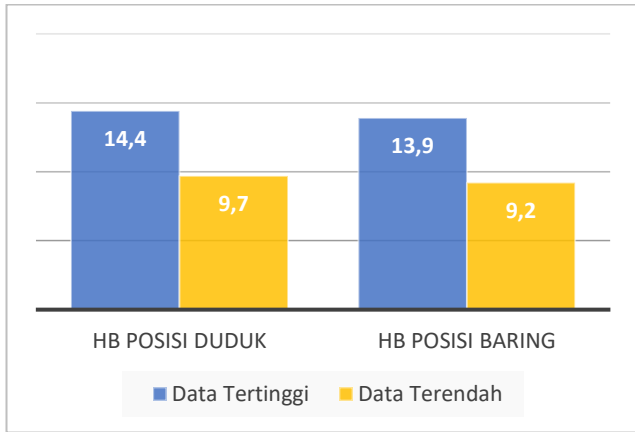
This research design uses One group pretest post test Design (Sugiono, 2013). Where the design of this study was used to find differences, taking venous blood in sitting and lying positions on hemoglobin examination with the automatic hematology analyzer method.

This study uses independent and dependent variables, where the independent variable is the blood sampling technique and the dependent variable is the hemoglobin level. Hypothesis testing is then carried out with the dependent T test or with an alternative non-partial test, namely the Wilcoxon test.

RESULT AND DISCUSSION

The results of the examination of the difference in hemoglobin levels with the technique of taking blood sitting and lying down, the results of the examination are as follows:

Figure 1. Graph of Hemoglobin Level Examination Results with Sitting and Lying Blood Drawing Techniques



Based on the results listed in table 1, the highest data for Hb sitting position is 14.4 g/dL, while the lowest data is 9.7 g/dL.

The highest data for Hb in sitting position is 13.9g/dL while the lowest data for b-data is 9.2g/dL.

From the study of differences in hemoglobin levels with the technique of taking blood sitting and lying down using a Hematology Analyzer, it was analyzed by the dependent t test. Prior to the Dependent test, a normality test was performed using Shapiro-Wilk. The test results are as shown in table 1 below:

Table 2. Data Normality Test

No.	Variabel	Sig.	Acceptance limit	Conclusion
1.	Hb lying position	0.005	$p \geq 0.05$	abnormal data
2.	Hb Sitting Position	0.003	$p \geq 0.05$	Abnormal data

Based on Table 5.2 Normality Test Results Data using Shapiro-wilk obtained a value of $p = 0.005$ for the sitting position and $p = 0.003$ for the lying position. So it can be concluded that the data is not normally distributed, which is then followed by a transformation with Log 10 with a p value < 0.005 which means the data is not normally distributed. Then tested the hypothesis with the dependent T test or with an alternative non-paramteri test, namely the Wilcoxon test.

Table 3. Result Wilcoxon test

	Sig.	Acceptance limit	Conclusion
Hb lying position	0,000	$p \geq 0,05$	There are significant changes
Hb Sitting Position			

Based on the results of the Statistical Test in table 3, it was found that Asymp.Sig.(2-tailed) was worth 0.000. Because the value of 0.000 is smaller than < 0.05 , it can be concluded that "Ha is accepted". This means that there is a difference between Hb Levels with Sitting and Lying Blood Collection Techniques, so it can be concluded that "There is a Difference in Hb Hb Levels with Sitting and

Lying Blood Collection Techniques".

Based on research conducted in the Hematology Laboratory of IkesT MP on 30 venous blood samples of IkesT MP students majoring in DIV TLM, level I and level IV students, the Hb level obtained in the sitting position blood collection was an average of 12.7 while in the lying position blood collection the Hb level was obtained with an average of 12.3. By comparing the average of the two different examination results between the examination of Hb levels in a sitting position with a lying position. The average result of sitting Hb levels is higher than the average lying position Hb levels. This shows that the position of the blood draw affects the patient's Hb level.

In general, ethylenediaminetetraacetic acid (EDTA) specimen collection by venipuncture can be performed with the patient in various postures including lying in bed, after walking through an outpatient service and sitting right before the test or sitting for long periods of time (Oliveira, 2017).

Clinical and Laboratory Standards Institute (CLSI) document H03-A6, renamed standard GP 41-A6, currently recommends that blood specimens should be taken with the patient sitting comfortably in an appropriate chair or reclining, but does not provide specifications on the supine or position. standing and permanent time in a certain position. Since posture can affect the concentrations of some blood constituents due to the decrease in plasma volume that occurs with the change from lying to standing, it is conventionally assumed that staying supine for long periods of time can be associated with consistent hemodilution. On the other hand, standing posture can be a cause of blood concentration due to the influence of gravity and hydrostatic pressure which causes ultrafiltration of plasma and small molecules in the interstitial space (Oliveira, 2017). Because the change from the supine to sitting position causes a clinically significant increase in hemoglobin, hematocrit and red blood cell count (Oliveira, 2017).

The results of this study are in line with the opinion of WHO which says that every step in the phlebotomy process affects the quality of the specimen and is therefore important to prevent laboratory errors, patient injury and even death.

Although according to Oliviera, 2017 in general, ethylenediaminetetraacetic acid (EDTA) specimen collection by venipuncture can be done with the patient in various postures including lying in bed, after walking through an outpatient service and sitting right before the test or sitting for long periods of time (Oliveira, , 2017).

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CONCLUSION AND RECOMMENDATION

Conclusion: Based on the results of research that has been done, it can be concluded that the position of taking venous blood lying down and sitting can affect hemoglobin levels.

Suggestion: Laboratory personnel are expected to pay more attention to the position of taking venous blood because it can affect the results of the examination, for further researchers, the results of this study can be used as comparison and reference material for research, and as consideration for further deepening further research, it is hoped that further researchers increase the number of samples so that the results are representative.

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