EVALUATION OF THE PERFORMANCE OF COPPER SULPHATE AND HAEMACUE METHODS FOR HAEMOGLOBIN ESTIMATION AMONG BLOOD DONORS IN DAR ES SALAAM, TANZANIA

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By

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A Dissertation Submitted in (Partial) Fulfillment of the Requirements for the Degree Master of Science (Epidemiology and Laboratory Management)

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CERTIFICATION

The undersigned certify that they have read and hereby recommend for acceptance by Muhimbili University of Health and Allied Sciences a dissertation entitled: **''Evaluation of the Performance of Copper Sulphate and HaemaCue Methods for Haemoglobin Estimation Among Blood Donors in Dar es Salaam, Tanzania",** in (partial) fulfillment of the requirements for the degree of Masters of Public Health of Muhimbili University of Health and Allied Sciences.

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Date

DECLARATION AND COPYRIGHT

I, **Goodluck Eliakim Mwanga,** declare that this **dissertation** is my own original work and that it has not been presented and it will not be presented to any other University for the similar or any other degree award.

Signature…….. **Date** ……...

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DEDICATION

I dedicate this dissertation to my beloved sisters Happiness and Glory Eliakim Mwanga and Mr. and Mrs. Eliakim Andrea Mwanga and my entire family for their love, patience, encouragement, prayers and unwavering support during my studies.

ABSTRACT

Background: The Tanzania National Blood Transfusion Service (NBTS) uses Copper Sulphate ($CuSO_4$) method for Hemoglobin (Hb) measurement in blood donors. However, this method may potentially provide many false results; this is because $CuSO_4$ is affected by the abnormal amount of serum protein since it precipitates Hb and other proteins in the blood. Currently, there is paucity of information in Tanzania on the proportion of false deferrals and false eligible blood donors through $CuSO_4$ method when compared with other Hb estimation methods like HemoCue.

Objective: This study aimed to evaluate the performance of $CuSO_4$ and HemoCue methods for Hb estimation among blood donors in Dar es Salaam, Tanzania.

Methodology: This was a cross-sectional study conducted from January to March 2019. It was conducted in Dar es Salaam at three (3) blood donation centers which are Muhimbili National Hospital (MNH), Eastern Zone Blood Transfusion Centre (EZBTC) and Temeke Regional Referral Hospital. A total of 204 blood donors were recruited. For each blood donor, capillary and venous blood was collected. Hb was estimated by $CuSO_4$, HemoCue and automated hematology analyzer (as a gold standard). Data was analyzed by Epi info 7. P-value of < 0.05 was used as a cut-off for significance test, Correlation coefficient was applied in finding the relation between venous blood Hb and capillary blood Hb and kappa agreements measurements for $CuSO_4$ and HemoCue was also established.

Results: The median age of the study participants was 30 [IQR, 20-39] years and 53% of the participants were males. The study revealed that the proportion of false eligible donor was 19.6% and false deferral blood was 2.9% by $CuSO_4$ method. The specificity for HemoCue was 62.5% and 92.9% in finger prick and in venous sample, respectively. In contrast, that of $CuSO_4$ was 28.6% and 35.7% in finger prick and venous samples, respectively. The sensitivity was above 96% for both methods. Difference in mean of 0.53 g/dl was also observed **by** using HemoCue method between venous and capillary Hb levels. In addition, linear regression model indicated a positive correlation between venous Hb from a finger prick ($r=0.913$).

Conclusion: This study found that the performance of $CuSO_4$ in Dar es Salaam region was low compared to HemaCue. NBTS should review, improve and monitor the performance of $CuSO_4$ before opting for another method

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LIST OF ABBREVIATIONS

CDC	Centers for Disease Control and Prevention
$CuSO_4$	Copper Sulphate
DIID	Donation Induced Iron-Deficiency anemia
ERB	Ethical Review Board
EZNBTC	Eastern Zone National Blood Transfusion Centre
Hb	Hemoglobin
HGB	Hb concentration
MUHAS	Muhimbili University of Health and Allied Sciences
NBTS	National Blood Transfusion Services
NPV	Negative Predictive Value
PPV	Positive Predictive Value
TRRH	Temeke Regional Referral Hospital
USA	United States of America
WHO	World Health Organization

DEFINITION OF OPERATIONAL TERMS

Sensitivity–It is the percentage of donors with hemoglobin (Hb) value below the cutoff of 12.5 g/dl (failed) identified by the test out of all venous Hb values below the cutoff. It denotes the ability of the test to prevent donation from anemic donors.

Specificity–It is the percentage of donors recruited for blood donation (passed) by the test out of all with venous Hb above the cutoff value.

Positive Predictive Value (PPV) - It is the probability that a below-cutoff value on the test method to be actually lower than a cutoff value by the reference method.

Negative Predictive Value (NPV) - It is the probability of an above-cutoff value to be actually so by the reference method, important for donor safety.

Cohen's kappa coefficient (κ) is a statistic which measures inter-rater agreement for qualitative (categorical) items.

Deferral donor means that an individual is not eligible to donate based on the criteria used to protect the health and safety of both the donors and transfusion recipient.

A regular blood donor is the one who has donated at least three times, the last donation being within the previous year, and continues to donate regularly at least once per year.

Eligible donor means that an individual is qualified to donate based on the criteria used to protect the health and safety of both the donors and transfusion recipient.

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background

Screening of donated blood before transfusion is a safety and quality procedure which protects blood transfusion recipients from acquiring infections that can be transmitted parenterally**.** Hemoglobin (Hb) screening is done as one of the pre-donation screening tests for blood donor selection along with observation of weight and blood pressure (1,2).

Hemoglobin is a protein found in red blood cells that carries oxygen in the body. Eligible blood donor should have Hb concentration of more than 12.5 g/dL for both men and women because, it is expected that the donor's Hb will decrease approximately by 0.7 to 1.5 g/dL after whole blood donation (3,4).

Hemoglobin examination is done to estimate the amount of iron present in the blood because there is no simple, rapid and direct bedside methods for determining the iron status (5). About 70% of human body iron is found in hemoglobin protein. A unit of transfused blood contains approximately 200 to 250 mg of iron (6,7) which is equal to 4-8% of total body iron (8). After the whole blood donation the body Hb level remains the same up to 2 weeks then the body starts to recover its previous Hb level from 3 to 4 weeks after phlebotomy (3) while lost fluid will be replaced within about 36 hours (9).

When blood donation takes place without accurately observing the threshold set of 12.5 g/dl of Hb, it may cause donation induced iron-deficiency (DIID) anemia (5) due to negative iron balance which has several complications to blood donors such as increased thirst , fatigue, shortness of breath, lower leg cramps , weak and rapid pulse , chest pain, faintness and heart-related symptoms like abnormal heart rhythms, heart murmur, enlarged heart, neurological malfunction and heart failure (10,11). Women of reproductive age have high risks to acquire anemia during blood donation (7,12) since iron stores are in average of 250 mg in women while 1000 mg in men (13,14). In developed countries women have the average amount of stored iron (ferritin and hemosiderin) of about 300 mg (15) while in developing countries most

women have depleted iron stores and will inevitably be precipitated into negative iron balance by blood donation when more than 200 mg of iron is lost through blood donation (5).

Substandard performance of Hb estimation methods in blood donors does not only increase the risk of developing anemia but also it may act as a source of losing current and future blood donors (16). When eligible donors **are** deferred, even with temporary deferral, the chance of them to return for more donation is 29 less compared to non-deferral donors, the difference continues to be magnified with time (17,18). The risk of losing blood donors is amplified more in first-time donors (16,18) wherein some of the studies they observed that none of the first time deferred donor return for blood donation (18,19).

Even though there are numerous methods of measuring Hb, there is no conformity on the ideal method to be used in Hb screening of the blood donors. Many countries used Copper Sulphate (**$CuSO_4$**) as the method for Hb estimation for many years before the 1950s (20) while HemoCue is the common method for Hb measurement for the general population rather than blood donors.

Copper Sulphate is a qualitative method based on the estimation of the specific gravity of blood when assuming that the donor has normal protein levels. The specific gravity of 1.053 corresponds to Hb level of 12.5 g/ dl. This method is known to be easy to perform, quick and cheap. But it has several weaknesses such as the blood drop into the solution gradually changes its specific gravity, as evaporation of the solution occurs, which can be a problem in areas with a hot climate. Waste disposal is an issue both because the solution used is a biohazard, and in some countries, also regarded an environmental toxin. Moreover, there is a lack of a generally accepted quality control for the method and the fact that it cannot quantify the exact amount of Hb therefore, it is not feasible to detect abnormal low or high Hb level (17).

The $CuSO_4$ method can false deferred up to 80% of deferred donors which can be recovered if another method of Hb is applied (21). Even with strict quality control, it observed that the $CuSO_4$ method may still fail to identify healthy donors and/or passes those with abnormal proteins and leukocytosis (22). Due to this, blood transfusion centers in other countries such as India have started to shift towards other methods of Hb estimation (17).

HemoCue is one of point-of-care test which has suitably replaced traditional laboratory setup in the modern health care arena. HemoCue provides easy and convenient Hb estimation based on the spectrophotometric reading. There are several advantages of this method, such as it is simple, portable, provides rapid (immediate result), it is battery operated, non-toxic and easy to use in poor settings where skills and resources are limited while the disadvantage of it, is that it uses disposable cuvettes which makes it expensive.

In response to that, this study aimed to evaluate and compare the performance for CuSO4 and HemoCue for blood donors screening; in order to find the best Hb screening method which can balance optimal safety for both donors and recipients while at the same time ensuring an adequate supply of blood and blood products in blood banks services.

1.2 Problem Statement

In order to safeguard the health of potential donors and ensure an adequate quality of blood for recipient's pre-donation hemoglobin screening is mandatory. Hence methods used for hemoglobin screening should have the ability to accurately detect the threshold set (12.5 g/dl). Copper Sulphate ($CuSO_4$) is the common method used in Hb estimation for blood donors. However, this method is affected by the abnormal amount of serum protein since $CuSO_4$ precipitates not only hemoglobin but also other proteins in blood such as white blood cells (23). Therefore, $CuSO_4$ may potentially provide false results by giving the results above or below 12.5g/dl thus will lead to wrong decisions being made regarding donor selection by causing either unnecessary anemia (DIID) to the donor after donation of blood or deferral of an eligible donor.

Since the establishment of Tanzania National Blood Transfusion Service (NBTS) in 2004, $CuSO_4$ was adopted as a method for Hb estimation for blood donors. However, the proportion of people who are false eligible and deferred blood donors is not known. This study intends to respond to the information gap of the exactness of this method towards screening of blood donors Hb by comparing with HemoCue as the common method used for Hb measurement.

1.3 Conceptual Framework

This is a laboratory investigations study; the conceptual framework is not applicable to this study. Please refer to MUHAS general regulations and guidelines for postgraduate programmes page number 158. The diagram below (Figure 1) is a simple structure used to illustrate research thesis.

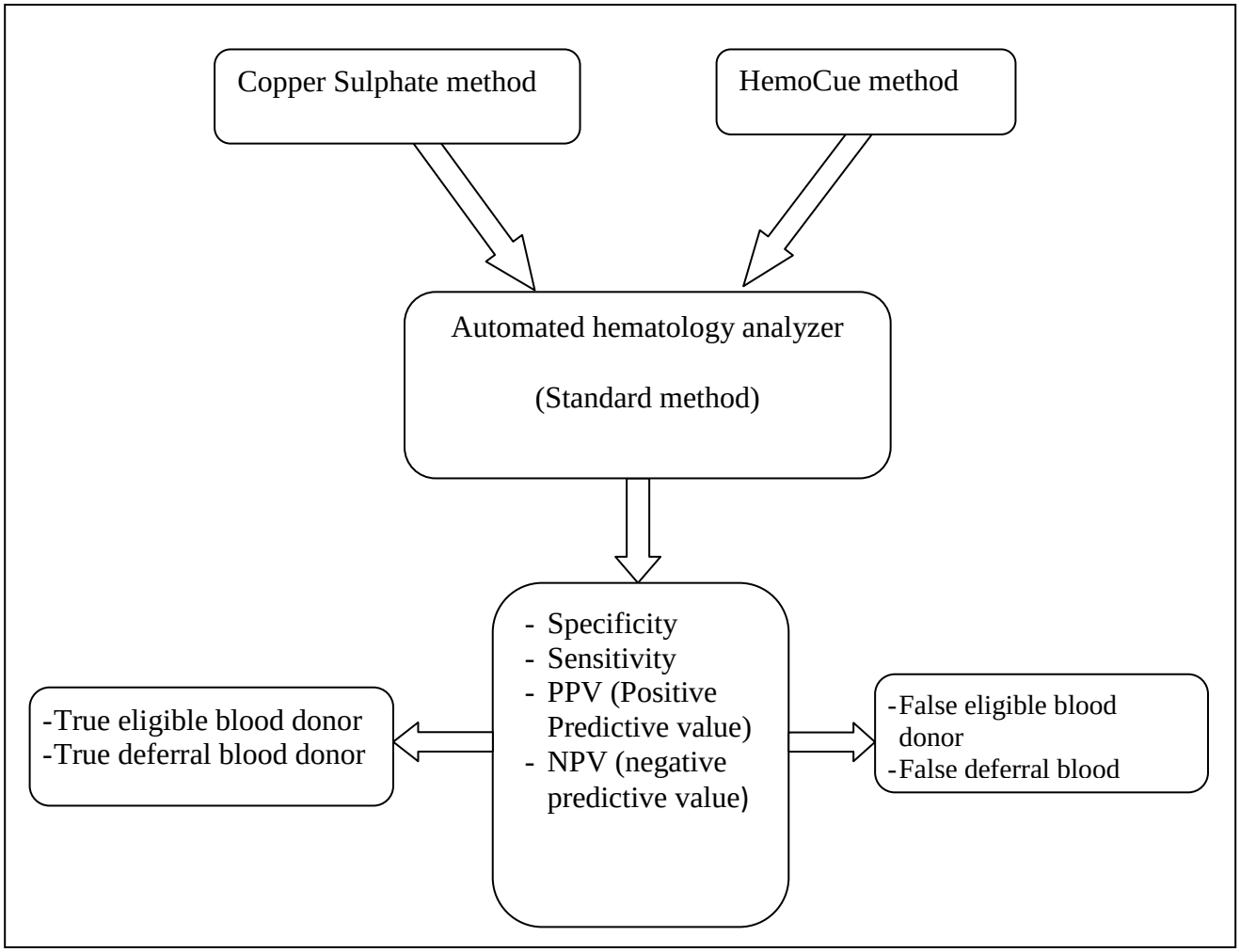


Figure 1: Simple structure for the research concept

The study looked at the performance of $CuSO_4$ and HemoCue 301 by focusing the sensitivity, specificity, positive and negative predictive value when automated Cell-dyne 3700 analyzer (Hematology analyzer) was used as the reference method. The performance of the methods will reflect true and false eligible (pass) and deferral donors.

1.4 Rationale of the study

The results obtained from this study will assist National Blood Transfusion Services (NBTS) and others who are engaged in blood donation activities to opt for the best methods of Hb estimation with minimal discomfort to donors, quick, as safe as possible, applicable in a mobile setting, accurate enough to avoid accepting anemic individuals for donation, and not unnecessarily reject eligible donors. Also, it will assist in providing best estimates of venous Hb of blood donor by using fingers capillary Hb, hence contributing towards improvement of quality of blood transfusion services in Tanzania.

1.5 Research Questions

1.5.1 Main research question

What is the performance of $CuSO_4$ and HemoCue methods for hemoglobin estimation among blood donors in Dar es Salaam, Tanzania?

1.5.2 Specific research questions

1) What is the proportion of false eligible and deferred blood donors by using $CuSO_4$ as the method for hemoglobin estimation in Dar es Salaam region?
2) What is sensitivity, specificity, positive and negative predictive value of $CuSO_4$ and HaemaCue methods used in hemoglobin estimation used in Dar es Salaam?
3) What is correlation between blood hemoglobin for venous and finger prick blood sample?

1.6 Objectives

1.6.1 Broad objective

- To evaluate the performance of $CuSO_4$ and HemoCue methods for hemoglobin estimation among blood donors in Dar es Salaam, Tanzania.

1.6.2 Specific objectives

1) To determine the proportion of false eligible and deferred blood donors by using **$CuSO_4$** as the methods for hemoglobin estimation in Dar es Salaam donation centers.

2) To determine sensitivity, specificity, positive and negative predictive value of $CuSO_4$ and HemoCue methods used in hemoglobin estimation by using cell dyne full blood picture analyzer as reference method.

3) To determine correlation between hemoglobin from venous and finger prick blood samples.

CHAPTER TWO

2.0 LITERATURE REVIEW

Globally one of the major reasons for blood donor deferral is low haemoglobin. This is seen in both developed and developing countries, for instance in United States of America (USA) each day 10% among 50,000 candidates of blood donation are deferred for the reason of low haemoglobin (24,25) while in Sub Saharan Africa countries, the country such as Ivory coast low haemoglobin contribute to 42.5% of deferred donors (7). Even though low Hb theoretically makes the donor only temporarily unable to give blood, deferral usually results in the permanent loss of the donor (15).

Although large number of donors are deferred and lost, the situation in Sub Saharan countries shows that more than 37 countries their blood collection is less than 10 units of blood per 1000 population while 25 of these countries collect less than half of the minimum estimated transfusion blood units in their countries (26,27). In this circumstance, blood transfusion programs must choose the best method for screening Hb which is accurate enough to avoid DIID and sensitive enough to capture every eligible donor as possible.

In order to ensure the blood donors are in good health and prevent blood collection from anemic donors, several methods are employed to measure the Hb level before the blood donation process. Those methods are hemoglobin color scale, Spun microhematocrit, Cyanmethemoglobin (HiCN) method, photometric method (HemoCue Photometer, HemoCue 201, Diaspect TM and Hem Control), noninvasive spectrophotometry (which includes NBM 200, OrSense Co., Petah-Tikya, Israel) and Copper Sulphate. The common methods in Africa countries for Hb estimation in general are Copper Sulphate, HemoCue and automated analyzer.

2.1 Copper Sulphate ($CuSO_4$)

The Copper Sulfate method is performed by letting a drop of blood fall into a $CuSO_4$ solution of known specific gravity (28) of 1.053 for blood donors. The drop of about 20 µl will either sink or float depending on whether it is heavier or lighter than the $CuSO_4$ solution. If the drop falls steadily to the bottom of the container, even if slowly, the specific gravity of the blood sample exceeds the solution. The blood droplet should be dropped into the copper sulfate solution from 1 centimeter above the surface, and the result should be read after 15 seconds. Assuming a normal specific gravity of plasma, the specific gravity of the blood sample is directly proportional to the Hb concentration (17). Each extra g/dl of plasma protein is equivalent to 0.7 g/dl Hb (23).

2.2 HemoCue

HemoCue is **a** battery-operated portable analyser. It consists of disposable micro-cuvettes containing reagent in a dry form and a single purpose designed photometer. Blood **is** drawn into the microcuvette by capillary action and spontaneously **mixes** with the reagents inside the microcuvette. The reaction in the microcuvette is a modified azidemethemoglobin reaction. The cuvette is inserted into the cuvette holder of the HemoCue. Sodium deoxycholate haemolyses erythrocytes and haemoglobin released. Sodium nitrite converts haemoglobin to methaemoglobin which, together with sodium azide, gives azidemethemoglobin. The absorbance is measured at two wavelengths (570 nm and 880 nm) in order to compensate for turbidity in the sample (23,29,30).

2.3 Automated haematology analyser

The automated hematology analyzers are computerized, highly specialized and automated machines that count the number of different kinds of white and red blood cells in a blood sample. It **is** taken as a reference method against which all the other methods **are** tested. The automated blood cell counter is calibrated annually and its quality control **is** done before use with the stabilized control reagents provided.

The Cell-Dyn 3700 system machine is a multiparameter, automated hematology analyzer designed for in vitrodiagnostic use in clinical laboratories in which the Hb concentration (HGB) is measured spectrophotometrically.

The HGB channel is used for the colorimetric determination of hemoglobin. A 1:301 dilution of the sample is made with the diluent and the HGB lyse reagent. The HGB lyse reagent, lyses the diluted red blood cells and converts the hemoglobin that is released to a stable chromogen.

This dilution is used for the HGB measurement. The Cell-Dyn 3700 System uses a cyanide-free reagent. This reagent converts HGB to a hemoglobin- hydroxylamine complex. A filtered light-emitting diode (LED) with a wavelength of 540 nm is the light source. A photo detector measures the light that is transmitted. The HGB is directly proportional to the absorbance of the sample at 540 nm. Five separate HGB readings are made on each sample. The lowest and highest are eliminated and the remaining three are averaged to give the final HGB sample reading. After the hemoglobin readings have been made, the HGB flow cell is rinsed with detergent. The rinse is drained and more detergent is delivered to the flow cell. A zero or blank reading is then obtained on the detergent to provide a reference to which the sample signal is compared. Five separate blank readings are made on each sample. The lowest and highest are eliminated and the remaining three are averaged to give the final HGB reference reading. The HGB result is expressed in grams of hemoglobin per deciliter of whole blood. Up to two decimal places may be displayed for hemoglobin results less than 10 g/dl. (31)

2.4 Proportion of false eligible and deferred blood donors by using Copper Sulphate

There are several studies which have demonstrated inappropriate characteristics of $CuSO_4$ when used as the method for Hb estimation. For example, the study conducted in a regional blood transfusion center of Western India which includes the total of 35,339 voluntary donors, $CuSO_4$ deferred half of the total deferred donors. When digital hemoglobinometer was applied to donors who were deferred by $CuSO_4$ method 37% were found to have Hb above the threshold (32).

In another study **$CuSO_4$** screening test inappropriately passed 19/500 (3.8%) donors, out of these 17 donors had Hb values between 11.0- 12.4 g/dl when tested by Cell Counter, while 07/500 (1.4%) donors were falsely deferred by $CuSO_4$ method (23).
The study done at Northern Ohio (USA) which includes more than 1000 participants, $CuSO_4$ method deferred 10.2% of blood donor while half of the deferred donor passed both micro hematic and cyanmethemoglobin test thresholds (33).

Also the accuracy of the Copper Sulphate investigated under usual blood bank conditions at Pacific Northwest Regional Blood Center (USA) using the cyanmethemoglobin technic as a gold standard. It was demonstrated that 83.9 % of females and 80.4 % of males false deferred by the $CuSO_4$ method while they had Hb above the threshold cut point (21). These findings led to several issues being raised regarding its screening accuracy such as a case publish by Mannarino et al wherein a donor with less than 8 g/dl of Hb passed the $CuSO_4$ screening test while another study in India concluded that 50% of $CuSO_4$ deferral donors can be recovered if another Hb method was used (4).

2.5 Sensitivity, specificity, positive and negative predictive value of CUSO4 and HemoCue methods used in Hb measurement.

Even though there is no consensus of the best method to be used in Hb screening of blood donors, it is important that the method chosen must have high sensitivity, specificity, PPV and NPV enough to avoid DIID but no less important, a method that doesn't defer prospective donors inappropriately. The study done by Gupta et al (2015) when automated Cell Counter is used as gold standard revealed that the performance of $CuSO_4$ to have specificity of 62 %, sensitivity of 98.4% Positive Predictive Value (PPV) of 91.86% and Negative Predictive Value of 81.4% while HemoCue was found to be more efficient with specificity of 84%, sensitivity of 97.7%, PPV of 98.24% and NPV of 97.6% (23).

A study done at Indonesian Red Cross Blood Transfusion Centre, Jakarta, found the **$CuSO_4$** method **to** have different values of specificity and sensitivity according to the level needed in detecting anemia. $CuSO_4$ measured its performance from the range of hemoglobin values of 3 to 17 g/dl (specific gravity range of 1.035–1.062) , its specificity range from 82.7% to 99.3% while sensitivity range from 55.6% to 76.8% when the hemoglobin-cyanide method (HiCN) was used as the gold standard (28).

A study done at Sao Paulo (Brazil) reported that HemoCue had sensitivity of 56% and specificity of 93.5% when ABX Pentra 60 was used as a gold standard method (34).

Performance of $CuSO_4$ in Tanzania is not well known with consideration that Hb measurement values are affected by many factors including age, sex, site from which the sample has been taken and races, (4,5,35) while $CuSO_4$ itself can be affected by high-temperature environment (due to its molecular structure of $CuSO_4$. 5H,O crystals) (17,36) and proteins presents in blood donors (17). This study will not only reveal the performance of $CuSO_4$ but also, it will compare the performance of $CuSO_4$ with HemoCue which is also the common method used in Tanzanian settings to measure Hb in individuals other than blood donors.

2.6 Correlation of haemoglobin from venous and finger prick blood samples

One of the reasons for measuring Hb is to determine the amount of iron present in the body before blood donation so as to prevent DIID. However, there is a difference seen in similar kinds of literature when it comes for selection of the best anatomical site for determination of the Hb in blood donors.

Studies have reported that, finger capillary sample overestimates the Hb values for instance a study which took place in the Department of Transfusion Medicine at the National Institutes of Health (NIH), Bethesda (USA) which included 150 blood donors revealed that capillary finger-stick HemoCue hemoglobin values were significantly higher than venous HemoCue hemoglobin results (37) .These results obtained in NIH, Bethesda are comparable to results obtained from Sao Paulo (Brazil) when they measure Hb from finger stick and venous blood

samples from 969 unselected potential female donors and revealed that there is overestimation of Hb values by finger stick sample by 5.9 g/l (34).

Other studies proclaim that there is no significant difference between finger stick and venous Hb values. A study done at Irish Blood Transfusion Service (IBTS) involving 36,258 participants with paired capillary and venous samples, established that the capillary hemoglobin was <12.5 g⁄ dl and < 13.5 g⁄ dl respectively, it concludes that capillary hemoglobin levels of 12.0–12.5 g⁄ dl in healthy females or 13.0–13.5 g⁄ dl in healthy males are substantively equivalent to venous hemoglobin levels of 12.5 and 13.5 g⁄ dl for women and men respectively (38).

In Tanzania finger stick capillary is used to determine the Hb value of blood donors, but there is paucity of information about the accuracy of determination of blood donor Hb by using finger stick site. This study, will establish if there is a significant difference between finger prick/stick capillary and venous blood Hb samples. If there is significant difference the association was established by using a linear regression method in order to predict the Hb value of blood donors by using the finger stick capillary samples.

CHAPTER THREE

3.0 METHODOLOGY

3.1 Study design and duration

This study was a cross-sectional study design, conducted from January to March 2019, within 3 blood donation centers located at Dar es Salaam region. In this study, the performance of Hb methods for blood donors was determined by using capillary and venous blood samples obtained from blood donors who presented themselves to selected sites and tested for Hb.

3.2 Study Area

The study was conducted in Dar es Salaam region. Dar es Salaam is among the coastal regions with an elevation of 24m from the sea level, found with Global Positioning System (GPS) coordinates of 6° 48' 8.4708" S and 39° 16' 46.4016" E. It is the largest city in Tanzania with a population of 4.3 million people (39). The city's land area is 538 square miles, making the population density 8,100 people per square mile. The annual population growth rate is the highest rate in the country of about 5.6%. Dar es Salaam is the region which contributes the highest number of all collected blood units, among 26 regions of Tanzania mainland (approximately 51000 units annual, which is 20% of total country wise blood collection) (40), with 9 permanent blood donation centers. Three (3) out of 9 blood center selected because of higher number of donors of blood donated in Dar es Salaam which are Muhimbili National Hospital (MNH), Eastern Zone Blood Transfusion Centre (EZBTC) and Temeke Regional Referral Hospital (40).

3.3 Source Population

The study population was clients who participated in hemoglobin screening before blood donation, from January to March 2019 in selected blood transfusion sites within Dar es Salaam region.

3.4 Sample size of the study

The following formula for calculation of sample size in this study was adopted from Hajian-tilaki 2014 (41):

$$n = \frac{\left[Z_{\frac{\alpha}{2}}\sqrt{P_0(1-P_0)} + Z_{\beta}\sqrt{P_1(1-P_1)}\right]^2}{(P_1 - P_0)^2}$$

Where:

n =Minimum sample size

P_0= Pre determined value of sensitivity or specificity (from previous study reported by Tondon *et al.*, 2009)

P_1 = assumed value of sensitivity (or specificity) under alternative hypothesis.

$Z_{\alpha/2}$ =Standard Z value for 95% confidence interval =1.96

Z_{β} = Standard Z value corresponding to 80% power = 0.84

For both CUSO4 and HemoCue methods, the sample size is calculated using the formula above. The estimates of sensitivity and specificity (P_0) from a previous study done in India (42) are:

CuSO4: Sensitivity = 98.8 % and Specificity = 58.1%

HemoCue: Sensitivity= 99.4% and Specificity = 84.4%

The assumed estimates of sensitivity and specificity under the alternative hypothesis for both methods (P_1) are:

$CuSO_4$: Sensitivity = 96% and Specificity = 68%

HemoCue: Sensitivity= 97% and Specificity = 92%

The table below shows the sample size estimates assuming 80% power, 95% confidence level and the different assumptions for sensitivity and specificity:

Table 1: Sample size estimates when power is 80% and confidence interval at 95%

Method	**P_0**	**P_1**	**n**
Sensitivity			
$CuSO_4$	98.8 %	96%	183
HemoCue	99.4%	97%	151
Specificity			
$CuSO_4$	58.1%	68%	180
HemoCue	84.4%	92%	151

The highest sample which is 183 was picked as the minimum participants' sample size number required in this study. The highest sample size of 183 participants is obtained when $CuSO_4$ sensitivity is equal to 98.4 %.

Plus 10% of non-response was included. Non- response in this study was included because the participants have right to withdraw themselves at any time after enrolling in the study.

Non- response formula: Which is 'N'= n/ (1-0.1)

Given that:
'N'= sample size of the study with the inclusion of non- response
n = sample size which excludes non- response (183)
When the numbers inserted into the formula:
'N'= 183/ (1-0.1)
'N'= 203.3 ≈ 204
Therefore, the minimum sample number of participants was 204.

3.5 Sampling Procedure

Convenience sampling was used to obtain 3 blood transfusion centers which is Muhimbili National Hospital (MNH), Eastern Zone Blood Transfusion Centre (EZBTC) and Temeke Regional referral Hospital due to their high contribution of blood donated in Dar es Salaam region (of about 90.2% according to January to December 2017 NBTS annual report).

The distribution of study participants for selected blood donation Centre was selected according to probability proportional to size (PPS) sampling, where participant from MNH, EZBTC and Temeke hospital were distributed in 80, 60 and 64 respectively as shown in Table 2.

Table 2: Representative blood donors from selected study sites of donations

Sites of donation	Population size	Sample size
Muhimbili	29157	80
EZBTC	23354	64
Temeke	21867	60
Total	**74378**	**204**

Note: Source of population data of sites of donation was based on total blood collection from January 2017 to July 2018.

Systematic sampling was used to select the study participants, which was found as follows:

Where: time of data collection was 40 days

: Minimum sample which can be found per day in the field is 10 people

(from Temeke hospital blood donation records)

: Total number of people who can be found within study period will be (40x10) = 400

: A sample size of the study = 204.

Formula: $k = \frac{\text{Population size (400)}}{\text{Sample size (204)}} = 1.96$

$1.96 \approx 2$

Therefore, every 2nd participant was sampled from people who donated blood during the working hours (from 8:00 am – 3:30 pm) from Monday to Friday, in study time frame until the minimum sample of participants was obtained. The first participant every day was obtained by using simple random sampling.

3.6 Inclusion and exclusion criteria

Inclusion criteria: All blood donor candidates who went for hemoglobin screening test at selected blood donation centers during the study period and also who consented to participate in the study population, were included.

Exclusion criteria: The study excluded all clients who came for donation but who fail to either understand the study or provide consent to participate in the study.

3.7 Recruitment and training of research assistants

Three research assistants with the phlebotomist qualifications were recruited from 3 selected blood donation centers found in Dar es Salaam. The set of the training covered, the following: rationale, purpose and scientific objectives of the research; study design methodology and data collection. In sample collection, research assistants were trained to ensure quality assurance of the test reagents and results, maintaining the cold chain, transportation of sample to the testing and storage area. Research assistants were trained on all required procedures; universal precautions, ethical guidelines for research including participants' rights; procedures for obtaining informed consent; and confidentiality requirements.

3.8 Data collection procedure

Capillary blood samples were obtained by lancing a fingertip on the index or middle finger of the left hand using a dry sterile lancet after disinfecting with ethanol and massaging the finger to facilitate blood flow when sitting prospective blood donors. The first drop of blood was wiped off. The second and third drops were used to fill a HemoCue microcuvette and capillary tube for $CuSO_4$ method in alternating order. Capillary blood samples collected and measured its Hb by using $CuSO_4$ and HemoCue.

The venous sample was collected in three milliliters (3 ml) from each of the subjects into Ethylene Diamine Tetra- Acetic acid (EDTA) anticoagulant test tube. The samples were sent to the laboratory and analyzed by using $CuSO_4$, HemoCue and hematology analyzer (Abbott Cell-Dyn 3700).

In this study, the samples and participant's data took place in blood collection centers of Dar es Salaam which include; Muhimbili National Hospital (MNH), Eastern Zone Blood Transfusion Centre (EZBTC) and Temeke Hospital. At the end of each day, the venous blood samples were transported to Muhimbili National Hospital in a cool box at a temperature of 2 to 8^0C in order to be analyzed by automatic hematology analyzer.

3.9 Data management and analysis

3.9.1 Validity and reliability of data

$CuSO_4$ performance was observed under usual blood bank conditions. The calibration of the HemoCue was verified by a control cuvette each day before the first measurement as recommended by the manufacturer. An automated FBP machine (Cell dyne 3700 analyzer) calibration and control were performed each day.

3.9.2 Data analysis

Data were collected in excel sheet where control of data quality was considered through the review of data collection tools. Confidentiality was observed accordingly. The data were entered in Microsoft Excel then exported to Epi Info 7 and Stata 13.1 versions for analysis. The data set copy backup was made for any occasion that may need backup during data analysis.

Descriptive analysis was carried out using frequency and proportion for categorical variables and means (SD) median (IQR) for continuous variables. Logistic regression technique was used to determine the relation of Hb from venous sample and capillary samples. Also, a linear regression model was developed to predict venous blood Hb from capillary blood Hb. A p-value of <0.05 was used as a cut-off for significance test. Also, Cohen's kappa coefficient was analyzed for HemoCue and $CuSO_4$. The value of kappa, 0.1–0.20; 0.21–0.40; 0.41-0.60; 0.61-0.80; 0.81-1.00 were considered slightly, moderate, substantial almost perfect agreement, respectively.

3.10 Ethical Clearance

This study was channeled through the Research and Publication Ethical committee of Muhimbili University of Health and Allied Sciences (MUHAS) for ethical clearance. After clearance, the study was performed after approval of selected blood donation centers managers. All blood donors who participated in this study were well informed about the study before they were asked to take part in the study. Information obtained in this study was kept confidentially. Also, numbers were used in the database instead of respondents' names for the purpose of gathering information. Access to data obtained from questionnaires was restricted to the researcher.

CHAPTER FOUR

4.0 RESULTS

4.1 Socio- demographic characteristics of study participants

A total of 204 blood donors participated in this study. The median age was 30 years (ranging between 18 and 60) years. Males contribute 73% of the study participants. Muhimbili National Hospital (MNH) contributed 39.2% of the participants as seen in table 3.

Table 3: Characteristics of study participants

Variable	Frequency (n=204)	Proportion (%)
Median age (IQR)	30 (18 -60)	
Age		
18-20	45	22.1
21-30	55	27.0
31-40	55	27.0
41-50	32	15.7
< 50	17	8.3
Sex		
Male	149	73.0
Female	55	27.0
Sites of donation		
Muhimbili	80	39.2
EZBTC	60	29.4
Temeke	64	31.4

4.2 Proportion of false eligible and deferred blood donors by CUSO4 method

All the 204 specimens were subjected to $CuSO_4$ and FBP analyser which was used as a reference method. A total of 182 (89.2%) were eligible donors while 22 (10.8%) were deferral donors by $CuSO_4$. Of all the eligible donors, 142 (78%) were qualified as eligible donors by $CuSO_4$ and FBP analyser, whereas 40 (22%) passed through $CuSO_4$ and failed through FBP analyser as seen in Figure 3. Of all the 22 deferral blood donors, 16 (73%) failed in both $CuSO_4$ and FBP analyser whereas 6 (27%) passed through $CuSO_4$ and passed through FBP analyser as seen in Figure 4.

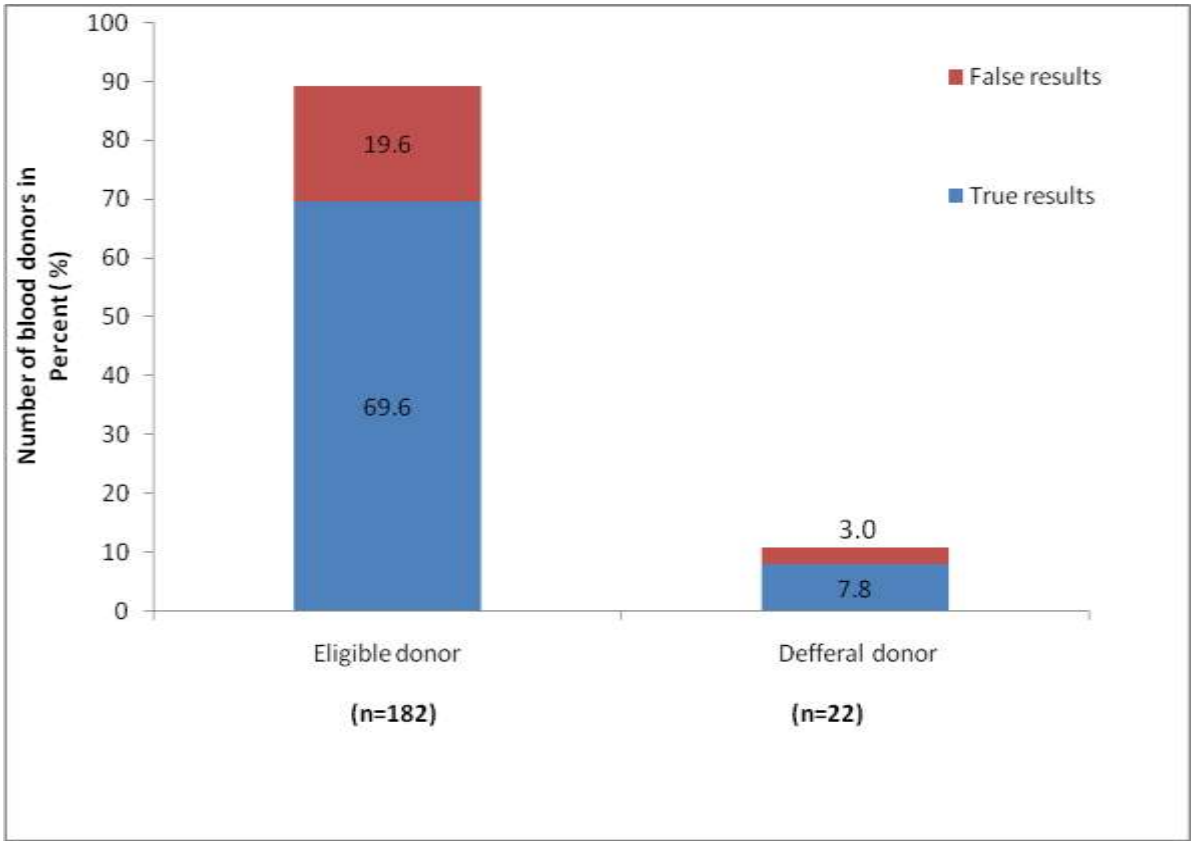
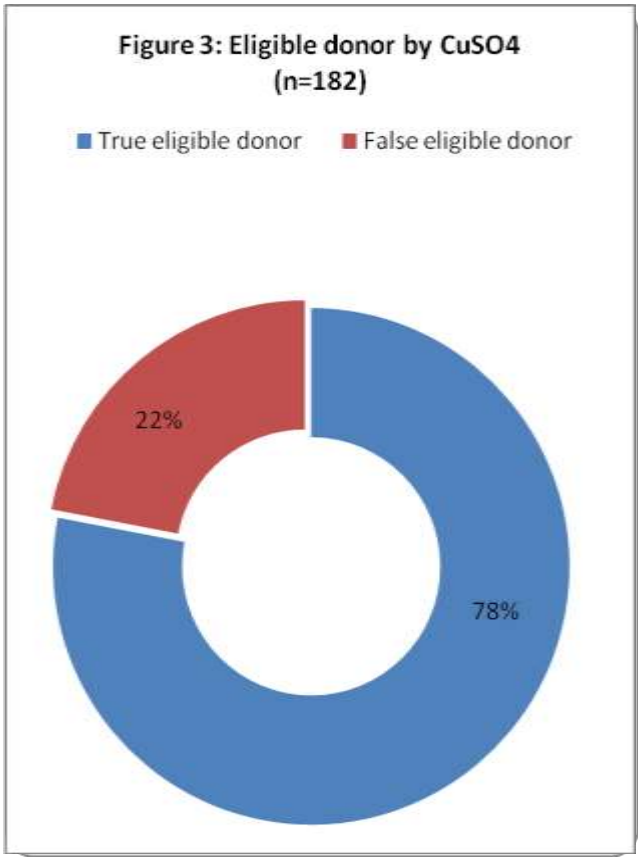


Figure 2: Percentage of eligible and deferral blood donors by $CuSO_4$ method with reference of FBP analyser



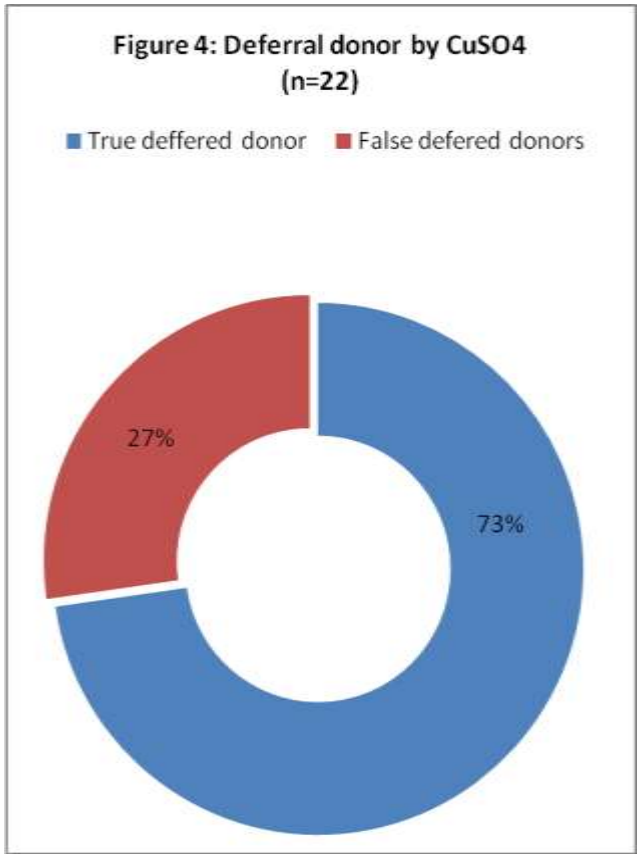


Figure 3: Eligible donor by CuSO4

Figure 4: Deferral donor by CuSO4

Distribution of false eligible and deferred donor by demographic characteristics when performed, it was observed that, out of the 6 blood donors who were falsely deferred by $CuSO_4$ method 80% were male while 47.4% of female donors who pass by $CuSO_4$ method were false eligible. Also, the study shows that 35.5% of eligible donors found in Temeke hospital have false eligible results (Table 4).

Table 4: Distribution of false eligible and deferred donor by demographic characteristics

Variable	**Deferred by $CuSO_4$ N = 22**	**False deferred blood donors N = 6 (%)**	**Eligible donors by $CuSO_4$ N = 182**	**False eligible donors N = 40 (%)**
Sex				
Male	5	4 (80.0)	144	22 (15.3)
Female	17	2 (11.8)	38	18 (47.4)
Centre of Donation				
Muhimbili	2	1 (50.0)	78	11 (14.1)
EZBTC	18	5 (27.8)	42	7 (16.7)
Temeke	2	0 (0.0)	62	22 (35.5)
Age group				
18-20	18	4 (22.2)	40	7 (17.5)
21-30	1	0 (0.0)	48	15 (31.3)
31-40	1	1 (100.0)	53	8 (15.1)
41-50	1	1 (100.0)	27	7 (25.9)
< 50	1	0 (0.0)	14	3 (25.9)

4.3 Overall performance of $CuSO_4$ and HemoCue methods using automatic FBP analyser as standard in all the three sites (Temeke, Muhimbili and EZBTC).

Sensitivity and specificity for the HemoCue were 62.5% and 98.7%, respectively for finger prick while 96.6% and 92.9% in venous sample which are greater than sensitivity and specificity of $CuSO_4$ which were 96.0% and 28.6% in finger prick while it was 98.7% and 35.7% in venous samples.

Cohen's Kappa (k) showed higher agreement between venous samples and reference method compared to agreement of results from finger prick samples (Table 5, Column A).

Data was taken from 3 independent blood transfusion donation sites, the performance of the $CuSO_4$ and HemoCue varied between sites as seen in table 5 (Column A, B and C) of Temeke Regional Referral Hospital, EZBTC and Muhimbili National Hospital respectively. EZBTC shows higher performance of $CuSO_4$ compared to Temeke and Muhimbili National Hospital.

Table 5 (Column B, C and D) shows that HemoCue in both pricker and vein have higher performance in terms of sensitivity, specificity, positive and negative predictive value without excluding measures of agreement with Cohen's Kappa method.

In addition, a higher performance is observed when venous sample was used compared to finger prick at NTBS and Temeke centers. In contrast, Muhimbili National Hospital (Column C) which showed no difference in performance when finger prick and venous Hb samples when measured by $CuSO_4$.

Table 5: Performance of $CuSO_4$ and HemoCue methods using automatic FBP analyzer as standard

	Column A				Column B				Column C				Column D			
	Overall performance all the three sites (Temeke, Muhimbili and Ilala)				Temeke Referral region hospital.				Muhimbili National hospital				EZBTC			
	CUSO4 pricker	HemoCue pricker	CUSO4 vein	HemoCue vein	CUSO4 pricker	HemoCue pricker	CUSO4 vein	HemoCue vein	CUSO4 pricker	HemoCue pricker	CUSO4 vein	HemoCue vein	CUSO4 pricker	HemoCue pricker	CUSO4 vein	HemoCue vein
True positive (n)	67	68	67	68	40	40	40	39	67	68	67	68	35	38	39	36
True negative (n)	1	6	1	10	2	16	4	22	1	6	1	10	13	13	16	20
False positive (n)	11	6	11	2	22	8	20	2	11	6	11	2	7	7	4	0
False negative (n)	1	0	1	0	0	0	0	1	1	0	1	0	5	2	1	4
Sensitivity (%)	98.5	100	98.5	100	100	100	100	97.5	98.5	100	98.5	100	87.5	95.0	97.5	90.0
Specificity (%)	8.3	50.0	8.3	83.3	8.3	66.7	16.7	91.7	8.3	50.0	8.3	83.3	65.0	65.0	80.0	100
PPV (%)	85.9	91.9	85.9	97.1	64.5	83.3	66.7	95.1	85.9	91.9	85.9	97.1	83.3	84.4	90.7	100
NPV (%)	50.0	100	50.0	100	100	100	100	95.7	50.0	100	50.0	100	72.2	86.7	94.1	83.3
Cohen's kappa (k)	0.10	0.63	0.10	0.89	0.10	0.71	0.20	0.90	0.10	0.63	0.10	0.89	0.54	0.64	0.80	0.86

4.4 Correlation between hemoglobin from venous and finger prick blood samples

The simple linear regression shows a strong positive correlation (with r = 0.913) between Hb estimate of venous blood and that of finger prick. Prediction formula was determined by the venous blood from the Hb estimate of finger prick through equation of y = 0.913x + 0.685 (Figure 5).

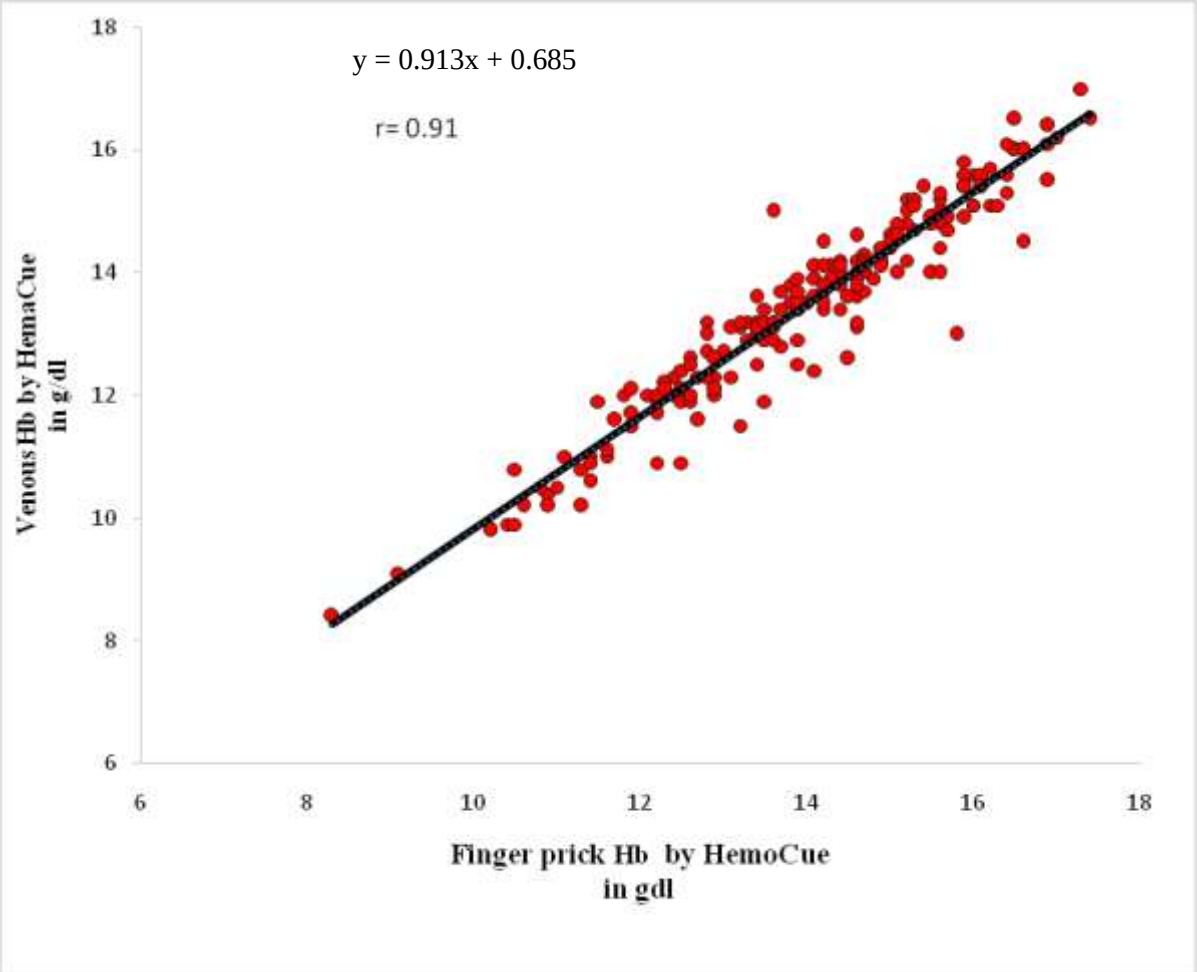


Figure 5: Correlation of Hb estimations via finger prick and venous route as site of blood collection by HemoCue

The median difference of Hb between blood samples from venous route versus from pricker was 0.55 g/dl by using HemaCue method while the median measure of venous sample are the same (13.50 g/dl) for both HemaCue vein and FBP analyzer (Figure 6).

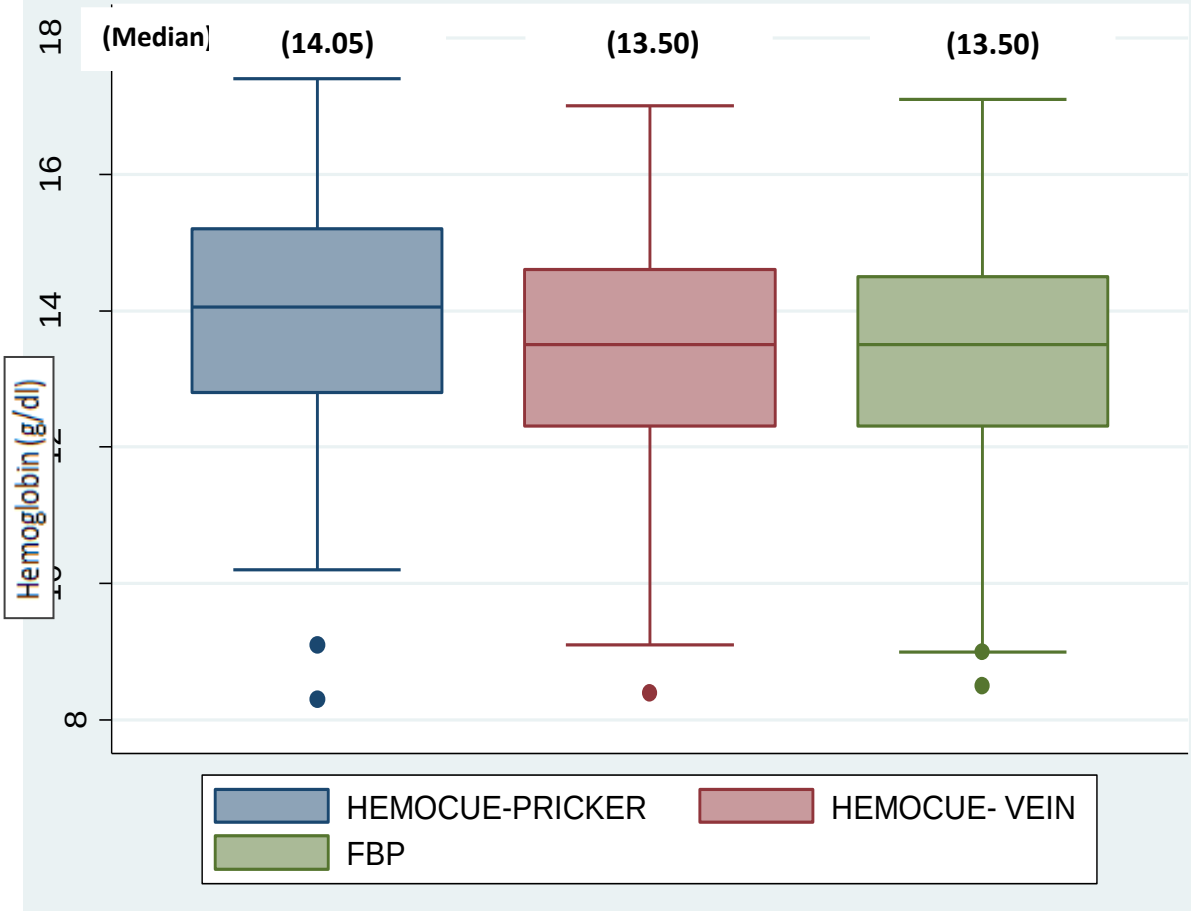


Figure 6: Comparison of hemoglobin estimations by finger prick HemoCue, venous HemoCue and venous Cell-Dyn methods

Paired T- test between Haemoglobin from pricker capillaries and vein blood samples was performed, where by the mean difference of Hb between those blood samples from venous route versus from pricker was 0.56 g/dl and this was statistically significant with p- value of < 0.0001 (Table 6).

Table 6: Paired T- test between Haemoglobin from pricker capillaries and vein blood samples

Variable	Mean	Standard Error	Standard Deviation	95% CI	T value	P value
HemoCue-Pricker	13.92	0.12	1.70	13.69 - 14.15	15.75	< 0.0001
HemoCue Vein	13.37	0.11	1.62	13.17 - 13.62		
Difference	0.56	0.03	0.48	0.46 - 0.59		

Some of factors which may affect the performance of $CuSO_4$ were noted during data collections procedure from three (3) selected blood donation centers. Most of factors that can improve the performance of $CuSO_4$ are present at EZBTC while absent at Temeke blood donation centre. (Table 7)

Table 7: Factors which can affect the performance of CUSO4 by site of blood donation

FACTORS WHICH CAN AFFECT PERFORMANCE OF CUSO4	MUHIMBLI	EZBTC	TEMEKE
Standard Operating Procedure (SOP) for preparation of $CuSO_4$	Present	Present	Absent
Standard Operating Procedure (SOP) for Hemoglobin screening using the $CuSO_4$ test	Present	Present	Present
Evidence of training for usage of $CuSO_4$ as screening test to blood donors	Absent	Present	Absent
Evidence of competence assessment to the health personnel	Absent	Present	Absent
Evidence of quality control of $CuSO_4$ before usage	Present	Present	Absent
Equipment and instruments for $CuSO_4$ preparation and quality control	Present	Present	Absent

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CHAPTER FIVE

5.0 DISCUSSION

It is very important to choose a method for Hb estimation in blood donors that is safe and with minimal discomfort to donors, quick, applicable in a mobile setting, accurate enough to avoid accepting anemic individuals for donation, and not unnecessarily reject eligible donors. Such method will assist in providing best estimates of venous Hb of blood donor by using fingers capillary Hb, hence contributing towards the improvement of quality of blood transfusion services. In addition, the method should have high sensitivity, specificity, PPV and NPV enough to avoid donation induced iron deficiency anemia but no less important, a method that doesn't defer prospective donors inappropriately because blood donors tend to be lost permanently after deferral.

In Tanzania, $CuSO_4$ is still used as a method for measuring hemoglobin for blood donors. This method was relatively cheap compared to other method employed for measuring blood Hb, but the effect of its performance can cripple both the safety of donors and performance of blood transfusion services.

In this study, the overall proportion of false eligible donors by $CuSO_4$ method in all the three sites was 19.6% meaning that $CuSO_4$ passed 19.6% of the blood donors that were not supposed to donate. The findings in this study differ from those reported in Mumbai and Northern India where proportion of false eligible donors in the study done in India found that $CuSO_4$ inappropriately passed 3.8% (19/500) of donors (23) while another study found 6.9% of the blood donors were false eligible (4). Proportion of eligible donors varied between the three sites where half of the false eligible donors were observed in Temeke Hospital (35.5%) followed by NBTS (16.7%) and MNH (14.1%). Blood donors who are falsely passed by $CuSO_4$ compromise their safety as they will be prone to anemia.

This study also revealed the overall proportion of deferred blood donors by $CuSO_4$ as 3% meaning that the $CuSO_4$ method misses 3% of blood donors who must have qualified to donate the blood. This proportion was higher when compared to the study done in India where the proportion of the deferred blood donors was only 1.4% (23). The high proportion of the false eligible and deferred donors may be caused by the lack of quality management checks of the CUSO4 method as the calibration and quality control of $CuSO_4$ method before data collection have been observed to reduce the proportion of the false and deferred donors (23).

The study findings showed that the $CuSO_4$ method had 96% sensitivity. A previous studies conducted in India showed 98.4% and 98.8 % sensitivity when automated Cell Counter is used as gold standard (4,23). The $CuSO_4$ methods is highly sensitive to detect eligible blood donors, therefore it will enable distinction between the eligible donors and non-eligible donors, thus avoiding compromising safety of the blood donors. The small difference observed between the current study and the studies done at India may be caused by usage of different type of reference machine in which this study we used Cell dyne 3700 while Indians' studies they used automated Cell Counter as reference.

In the present study, $CuSO_4$ method exhibited the specificity of 28% which is in contrast of the findings from the previous evaluation done in India where they found the specificity of 58.1% and 62% (4,23). Therefore, $CuSO_4$ method showed low ability to measure actual deferral blood donors that were correctly identified by $CuSO_4$. The low ability of $CuSO_4$ method may be explained by an absence of equipment for preparation of $CuSO_4$ reagent, lack of quality control and training for health personnel that was observed in the facilities during the current study.

The study exhibited the PPV and NPV of 78% and 72.7%, respectively. This was in contrast with the previous studies that showed PPV of 92.3% and 95.8% and NPV of 90.7% and 81% respectively (4,23). The low PPV explains the probability of $CuSO_4$ to detect eligible donors which are above 12.5 g/dl while the low NPV explains the probability of truly deferred blood

donors with a less than 12.5g/dl (NPV) to be deferred by $CuSO_4$. Implication of low PVP and NPV may cause donation induced iron-deficiency (DIID) anemia (5) due to negative iron balance which have several complications to blood donors such as increased thirst , fatigue, shortness of breath, lower leg cramps , weak and rapid pulse , chest pain, heart murmur, enlarged heart, neurological malfunction and heart failure (10,11). The difference in PPV between the present study and the previous studies might be due to nonuse of Standard Operating procedure (SOP) that was observed during the current study.

The current study obtained the findings from different independent blood donations centers located in Dar es Salaam region. It was observed that EZBTC has higher performance of $CuSO_4$ method compared to Muhimbili National Hospital and Temeke Regional Hospital. The higher performance of EZBTC can be explained by presence of trained and competent staff and the performance of quality control of $CuSO_4$ before usage.

The difference in the performance of Hb method between finger prick and venous sample was observed as vein samples had high performance regardless of the method used (both $CuSO_4$ and HemoCue). Other similar studies revealed similar results such as the study done in Bethesda (USA) and Sao Paulo (Brazil) (4,23). The higher performance obtained by venous samples can be explained by the usage of reference (Cell dye 3700) method which uses only venous sample. The Hb of venous sample was not affected by dilution from extracellular tissue fluid exuded due to skin pressure, skin temperature and depth of skin penetration compared to finger capillary sample.

In order to investigate whether this difference in the performance of hemoglobin measurement was due to the blood sampling site or the analytical instrument used, we assessed capillary and venous Hb by HemoCue Hb 301 and compared these with venous Hb concentration by the Abbott cell dyne 3700 as the reference method. Capillary blood was observed as an estimate rather than a measure of the true venous Hb and cutoff values in the guidelines have been set on the based on venous Hb levels. The difference observed in this study was significant of about 0.525 g/dl whereby venous samples have lower Hb results compared to pricker samples

although they have good correlation. The significant difference of Hb results from pricker and venous can change the performance of the method by increasing performance of method when the type of sample used match with sample used by reference method. Previous study done at Department of Transfusion Medicine at the National Institutes of Health (NIH), Bethesda (USA) (37) and from Sao Paulo (Brazil) reported the same results as the current study (34).

5.1 Study strength

This is the first study conducted in Tanzania among blood donors on the performance of Copper Sulphate and HaemaCue methods for hemoglobin estimation, thus it can be used as baseline information for further studies. Also, the study results can be generalized since selected blood donations centres contribute to 90.2% of donated blood in Dar es Salaam region.

5.2 Study limitation and mitigation

This study used the gold standard that is only applied to the venous sample due to the fact that Cell dyne analyser need a larger amount of blood and therefore the cell dyne analyser could not be used to test for the finger prick samples.

Limitation of the study was mitigated by repeating measurement of finger capillary of $CuSO_4$ and HemoCue method by using vein sample and hence removes the results which were influenced by the anatomical site of the sample.

CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATION

6.1 Conclusion

$CuSO_4$ method was found to have lower performance for hemoglobin measurement compared to HemoCue since false positive and negative results are common in both venous and pricker samples. This was attributed to the absence of equipment and competence in preparation and use of $CuSO_4$ method. In order to save the pool of eligible blood donors The Hb level of donors rejected by $CuSO_4$ may be reassessed by HemoCue. These findings could be of value to blood centers with limited resources especially for camp donations where mass donor Hb screening is carried out. Also, the findings may change perspectives of NBTS and related stakeholders (including planners and policymakers) of blood donation to consider this significant change of Hb results when reviewing the guidelines for screening of Hb for blood donors.

Further studies are needed to explain blood donor return following temporary deferral by $CuSO_4$ method and clinical manifestation of anemia for blood donors who are deferred by $CuSO_4$.

6.2 Recommendation

We recommend the following to improve performance of $CuSO_4$ as the method used for measuring Hb of blood donors:

- National Blood Transfusion Services (NBTS) as the main stakeholder of blood transfusion services, it should review, improve and monitor the performance of CuSO4.
- NBTS should conduct continual trainings and supervision at blood donations centers,

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APPENDICES

Appendix I A: Consent Form (English Version)

STUDY TITLE: "Evaluation of the performance of Copper Sulphate and HemoCue methods for hemoglobin estimation among blood donors in Dar es Salaam, Tanzania"

Principle investigator	Institution	Mobile phone number
Goodluck E. Mwanga	Muhimbili University of Health and allied sciences	0767 159 580

Investigator's statement

My name is Goodluck E. Mwanga, am doing this study as part of the requirements to complete Master degree of Public health in Muhimbili University of Health and Allied Sciences (MUHAS). The purpose of this form is to give you information about the study. Kindly read it carefully, and ask me questions about anything that is not clear to you, regarding what I will ask you to do, the risks and benefits involved and your rights as a volunteer. You can also ask about anything you wish to know about the study. When all is well understood so you can make an informed consent whether to participate in the study or not. If you wish to be contacted with the results of the tests, you will be requested to provide your mobile telephone numbers. You will also be asked to sign or thumbprint on the form as a sign that you have accepted by your choice to participate in the study.

Purpose of the Study

The results obtained from this study will assist National Blood Transfusion Services (NBTS) and others who are engaged in blood donation activities to opt for the best methods of Hb estimation with minimal discomfort to donors, quick, safe as possible, applicable in a mobile setting, accurate enough to avoid accepting anemic individuals for donation, and not

unnecessarily reject eligible donors, hence contributing towards improvement of quality of blood transfusion services in Tanzania.

Risks or discomfort

There is no anticipated mediate, short-term, or long-term risks or distresses that may arise out participation in this study. But should you be uncomfortable with any of the questions or procedure asked, you are free to decline and withdraw your consent. It will not in any way affect your access to services in this department

Benefits and compensations

There is no additional cost to you due to your participation in the study, and no physical injuries are anticipated. There will be no compensation.

Confidentiality

Participation in this study is voluntary and you can decline and your consent without loss of any benefits or any penalties. Your name will not be used on the study data forms only study numbers will be used. All your personal information will be treated confidentially. The investigator may use the data for analysis and quality control purposes or publication but your identity will never be reported.

Who to call in case of a query or problem after the study?

You can call the investigator on the phone lines listed below and additional contacts for the supervisor

Name of supervisor	**Institution**	**Contact /Mobile number**
Dr. Doreen Kamori	Muhimbili University of Health and Allied Sciences (MUHAS)	0711 954 661
Ms. Loveness Urio	Field Epidemiology and Laboratory Training. Program	0756 750 727

Participants' statement and signature

The study described above has been explained to me. I have had a chance to ask questions and feel satisfied to make an informed consent to take part in this study. If in future I wish to ask any questions about the study I can contact the investigator through the provided contacts.

……..	…….	……
Participant signature/thumb print	Mobile number	Date

Appendix II B: Consent Form (Swahili Version)

FOMU YA IDHINI

Jina la Utafiti: "Tathmini ya utendaji wa Kopa Salfeti na HemoCue kama njia za makadirio ya hemoglobin kwa watu wachangiaji damu Dar es Salaam, Tanzania"

Utambulisho wa mtafiti

Naitwa Goodluck E. Mwanga , ninafanya utafiti huu kama sehemu ya mahitaji ya kukamilisha shahada ya ya uzamili katika Afya ya umma katika Chuo Kikuu cha Afya na Sayansi shirikishi cha Muhimbili. Kusudi la fomu hii ni kukupa taarifa kuhusu utafiti huu. Tafadhali isome fomu hii kwa uangalifu, na uulilize maswali ya chochote ambacho hukielewi, kuhusu kile nitakachokuomba kufanya, hadhari na faida zinazohusika na haki zako kama kujitolea. Unaweza pia kuuliza juu ya chochote unataka kujua kuhusu utafiti huu. Ukiisoma na kuridhika unaruhusiwa kushiriki katika utafiti au la. Ikiwa ungependa kuwasiliana na matokeo ya vipimo, utaombwa kutoa nambari za simu za mkononi. Pia utaombwa kusaini au kutia kidole kwenye fomu kama ishara kwamba umekubali kushiriki katika utafiti.

Kusudi la utafi ti huu

Matokeo yanayotokana na utafiti huu utasaidia huduma za mpango wa Taifa wa Damu Salama na wengine wanaohusika katika shughuli za mchango wa damu ili kuchagua kipimo bora zama kadirio ya wingi wa damu ambacho kina usumbufu mdogo kwa wachangiaji damu, kipo haraka, salama iwezekanavyo, inayofaa katika mazingira ya kuhama hama , sahihi kwa kiwango cha kutosha ili kuepuka kukubali watu wanaoathiriwa na uchangiaji damu, na sio lazima kukataa wafadhili wanaostahiki kwa kuzingatia ufanisi wa gharama, na hivyo kuchangia kuboresha huduma za uchangaji wa damu nchini Tanzania.

Hadhari au usumbufu

Hakuna hadhari wala mataatizo ya muda mrefu au mfupi, yatakayotokana kwa kushiriki katika utafiti huu. upo huru kukubali au kujitoa katika ushiriki wako muda wowote. Na haitakuadhiri kupata huduma katika kitengo hiki muda wowote.

Faida na fidia

Hakuna gharama ya ushiriki wako katika utafiti huo, na hakuna majeruhi ya kimwili yatakayosababishwa na utafiti huu. Hakutakuwa na fidia yeyote kwa ajili ya utafiti huu.

Usiri

Kushiriki katika utafiti huu ni kwa hiari na unaweza kujitoa bila kupoteza na faida yoyote au kupata adhabu yoyote. Jina lako halitatumiwa kwenye fomu za data za utafiti huu ila namba maalumu zitatumika. Maelezo yako yote yatakua siri. Matokeo ya vipimo yanweza kuchapwa kwenye jarida la mafunzo.

Unaweza kumpigia simu kwa maswali au kujifunza juu ya utafiti huu

Unaweza kupiga simu kwenye namba zilizoorodheshwa hapo chini na anwani za ziada kwa

Wasimamizi wa Utafiti

Jina la msimamizi	Taasisi/Ofisi	Namba ya simu
Dr. Doreen Kamori	Chuo Kikuu cha Afya na Sayansi shirikishi cha Muhimbili	0711 954 661
Ms. Loveness Urio	Field Epidemiology and Laboratory Training. Program	0756 750 727

Taarifa ya mshiriki na saini

Maelezo yaUtafiti yalioelezwa hapo juu nimeelewa vizuri. Nimeuliza maswali na nimekubali kwa hiari yangu kushiriki katika utafiti huu. Ikiwa katika siku zijazo napenda kuuliza maswali yoyote kuhusu utafiti huu ninaoweza kuwasiliana na watafiti namba za simu zilizotolewa.

....

Saini/ Dole gumba la mshiriki tarehe Namba ya simu