"DONOR DEFERRAL DUE TO ANEMIA AND ITS MORPHOLOGICAL PATTERN ANALYSIS IN BIJAPUR- A PROSPECTIVE STUDY"

By

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Dissertation submitted to BLDE University, Bijapur

In partial fulfillment of the requirements for the degree of

MD

in

PATHOLOGY

Under the guidance of

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2015

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ACKNOWLEDGMENT

A line from Sanskrit Shlokha Says “Gurur brahma guru r vishnu gurudevo maheshwaraha, guru ssakshaat parabrahma tasmay shri gurave namaha” - meaning a teacher is next to god and without him knowledge is always incomplete

I wish to take this opportunity to express my indebtedness to my guide **Dr. B. R. Yelikar,** Professor and Head. Department of Pathology, for his resolute guidance, precise approach, constructive criticism and meticulous supervision throughout the course of my work and preparation of manuscripts that have been a valuable part of my learning experience.

I also wish to thank **Dr.Anita. P. Jawalgi**, Assisstant Professor Department of Pathology, for her immense guidance, supervision & approach throughout the study.

I also wish to thank my family members especially Dr.Omprakash Kalyani, My Parents **Mr Vaijinath Kalyane, Mrs Nirmala Kalyane** for their moral support and encouragement during my study.

I am forever grateful to **Dr.S.U.Arakeri** Prof, **Dr R.M.Potekar,** Prof. **Dr.Mahesh H. Karigoudar** Prof, **Dr Surekha Hippargi** Prof, **Dr.Girija Patil** Assoc Prof, **Dr Prakash Patil** Assoc Prof, **Dr.Vijayalaxmi S. Patil** Asst. Prof, for their valuable help and guidance during my study.

I am very grateful to **Praveen Honwad**, all the non teaching staff of Department of Pathology, who has helped me during this work.

I also wish to thank **Mrs. Vijaya Sorganvi** for her valuable help in statistical analysis

Last but not the least**,** My sincere gratitude to all my study subjects whose cooperation has contributed to this study.

Date: **Dr. Mamata Kalyane**

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ABSTRACT

BACKGROUND: The paucity of healthy, safe blood donors has been a serious problem in the blood banks worldwide. The majority of donor populations are deferred due to temporary but easily correctable cause –Anemia. Prevalence rate of anemia in donors is 1.8%. The hemoglobin cut off value for blood donation is 12.5gm% for both male and female.

OBJECTIVES: To determine the prevalence and spectrum of anemia in blood donor deferrals.

METHODS: Prospective donors presented to Blood bank from 1st Oct 2012 to 30th June 2014 and deferred due to anemia in BLDE University, Shri B. M. Patil Medical College, Hospital and Research centre, Bijapur have been included in this study. 5 ml of venous blood sample is collected in EDTA from all the prospective blood donors and sample is processed in automated hematology analyzer, Sysmex KX-21.The data is collected for all hematological parameters & is used to determine the prevalence & pattern of anemia.

RESULTS: A total of 4063 blood donors presented to blood bank during this period, out of which 130 donors are deferred due to anemia. Percentage of donors deferred due to anemia are 48.8% and Prevalence rate of anemia is 3.1% .Most common type of anemia is normocytic normochromic anemia.

CONCLUSION: As anemia is the most common cause for temporary donor deferral. Its appropriate diagnosis and referral for treatment will help in improving health of the donors and motivating them for returning for blood donation and reducing the paucity of blood donors to a large extent.

KEY WORDS: Anemia, donor deferral, paucity

LIST OF ABBREVIATIONS USED
(In alphabetical order)

ACD	Anemia of chronic disease
Fe	Iron
Hb	Hemoglobin
IDA	Iron deficiency anemia
MCH	Mean corpuscular hemoglobin
MCHC	Mean corpuscular hemoglobin concentration
MCV	Mean corpuscular volume
NCNC	Normocytic normochromic
RBC	Red blood cells
TfR	Transferrin
TIBC	Total iron binding capacity
WHO	World Health organization
ZPP	Zinc Protoporphyrin

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INTRODUCTION

Blood transfusion saves lives and improves health, but millions of patients who needs transfusion do not have timely access to safe blood. It is estimated that donation by 1% of the population (10 per 1000 population) is generally the minimum needed to meet a nation's most basic requirements for blood; the requirements are higher in countries with more advanced health-care systems. [1]

National AIDS Control Organization's (NACO) statistics show that the annual rate of blood donation in India is about 7.4 million units, against the requirement of 10 million units. [2]

The state of Karnataka contributes about 500,000 units, with 62% coming via voluntary blood donation.[3]

Pre donation donor selection is done for the safety of the blood donor and recipient. Various causes of the pre-blood donation deferral need to be studied systematically to improve the donation rate. Monthly statistics sent to the drug controller exclusively includes transfusion transmitted diseases data and excludes other major causes of deferral.[1]

Deferral is a painful and sad experience for blood donor as well as the transfusion centre and it necessitates additional efforts towards new recruitments. Moreover, deferring prospective donors often leaves them with negative feelings about themselves as well as the blood donation process. [1]

There are standard guidelines for selection as well as deferral of a prospective blood donor as per the "Drugs & Cosmetic Act 1940 of FDA".[4]

The donors are deferred due to several reasons related to safety of donors e.g hemoglobin level below 12.5gm% or body weight below 45kgs and potential threat to recipient example a prospective donor who is seropositive for HIV.[5,6]

Among the various causes for donor deferral, the most common cause is anemia that is 15.5% to 32.9%. The prevalence of anemia among prospective donors is 1.8%.[5]

Among the various causes of deferrals anemia is the most common cause. The majority of donor population in developing countries like India, are deferred due to temporary but easily correctable cause – anemia.[5]

Accordingly these deferrals can be categorized as temporary which includes short term(1-56 days) due to anemia, long term(57-365days)due to pregnancy and permanent (more than 365 days) due to hepatitis B infection.[5,6]

AIMS AND OBJECTIVES.

1. To determine the prevalence rate of anemia in blood donor deferrals.
2. To determine the morphological pattern of anemia in donors deferred due to anemia.

REVIEW OF LITERATURE

Considering the estimated shortfall of 3-4 million units of blood annually in India, more awareness must be created among all strata of population especially among youngsters on importance of blood donation.[7]

Blood can save millions of life, and young people are the hope and future of a safe blood supply in the world. National blood donation policy of India high lights on the needs of supplying safe and quality blood through collecting blood from regular voluntary donors.[8]

The need for safe and secure supplies of blood and blood products is universal. Worldwide, at least 92 million units of blood are donated each year to save lives and to improve health. However, demand for blood for transfusion continues to increase and many countries cannot meet existing needs.[8]

Today 62 countries have blood transfusion services based entirely on voluntary blood donation, up from 39 in 2002.India repots the greatest increase in the number of voluntary unpaid blood donations from 3.6 million in 2007 to 4.6 million in 2008.[8]

In 2008, 162 countries provided data to WHO on 91.8 million donations. The data comes from countries that account for a total 6.2 billion people, representing 92% of the global population.[8]

In many regions this means inadequate supplies to replace blood lost in child birth and to treat anemia that threatens the lives of children who have malaria or are undernourished. Everywhere blood and blood products are needed for routine and

emergency surgery, including life saving treatment for growing number of people in road traffic accidents, and for treating congenital blood disorders.[9]

India's blood requirement is about 12 million units per year. Blood banks in India are able to collect only 9 million units per year.[9] The shortage of blood is 40%.[9]

WHO recommends developing strategies for 100% voluntary blood donation and states in India achieved more than 80% by voluntary blood donation. More than 38,000 blood donations are needed every day.[8]

In Karnataka every year there is a blood shortage by 4 lakh units. For the last 5 years the shortage of blood in blood banks has become a more serious problem.[10]The status of voluntary blood donation is worrisome especially in Bijapur. Added to this being a economically and development point of view a backward district the deferrals due to anemia is more common.

DONOR DEFERRAL

Deferrals lead to loss of precious blood/ components available for transfusion. For preventing this, we should be having knowledge of causes of deferral and their frequency.[11]

Donors are deferred due to various causes. Among the various causes of deferrals anemia is the most common cause.[5]

The most common cause for deferral is anemia (32.9%)in study done by Bahadur et al[6] similar to that reported in Turkish donors by Arslan[12] (2007) (20.7%) and Custer *et al*[13] (2004) (60% of temporary deferrals) and Halperin *et al* [14](1998) (46%). The second most common cause of deferral was low weight, which accounted for 59.4% of total deferrals. Hemoglobin and low weight combined accounted for 65.3% of total deferred similar to the findings by Chaudhary [15]*et al* (2008)

DONOR DEFERRAL CATEGORIZATION [11]

1. Temporary short term(1-56) days e.g due to anemia
2. Long term(57-365) days due to pregnancy and
3. Permanent (more than 365 days) e.g due to hepatitis B infection.

CAUSES OF TEMPORARY DEFERRAL

1. Hemoglobin less than12.5%,
2. Malaria in last 3 month,
3. Weight lesser than 45kg,
4. Jaundice in last 1year,
5. Alcohol in last 72hrs,

6. On antibiotic/aspirin for last 3 days,
7. Upper respiratory tract infection,
8. Age <18yrs,
9. Previous donation in last 3 months,
10. Syphilis for 1 month,
11. Hypertension,
12. Enteric fever in last one year,
13. Dental extraction/surgery last 6 months,diabetes on insulin therapy,
14. Pregnant and lactating women,
15. Drug addict
16. Cat/dog bite/ rabies vaccination last 1 month
17. Tuberculosis,
18. Stroke,
19. Tattoo/ear piercing last 1 year

CAUSES OF PERMANENT DEFERRAL

1. Sero positive for HBV
2. Age> 60 yrs,
3. HCV positive,
4. Chronic obstructive lung disease,
5. HIV-1,2 positive,
6. Heart disease,HTN
7. Asthma,
8. Epilepsy, leukemia/lymphoma/multiple myeloma,
9. Patient who has received chemotherapy.

ANEMIA

Among the various causes of temporary donor deferrals in India , anemia is most common. [11]

In the study done by Shalini Bahadur et al The percentage of donors deferred due to anemia was estimated to be 15.5%, the prevalence rate was 1.8%[5]

Anemia is functionally defined as an insufficient RBC mass to adequately deliver oxygen to peripheral tissues.[16]

For practical purposes, any of the three concentration measurements performed on whole blood can be used to establish the presence of anemia.

1. The hemoglobin (Hb) concentration typically is expressed as grams Hb per deciliter (g/dl) in the United States and as grams per liter in Europe.
2. The hematocrit (Hct; also called the packed cell volume [PCV] or volume of packed red blood cells [vPRC]) represents the proportion of blood volume represented by RBCs, and is expressed as a percent or as a decimal.
3. The RBC concentration is expressed in cells per microliter (10^6/μl) or cells per liter (10^{12}/L). The red cell concentration is least commonly used in the definition of anemia.[15]

MORPHOLOGICAL CLASSIFICATION OF ANEMIA

Anemias may be initially classified morphologically according to average size and hemoglobin concentration of erythrocytes as indicated by erythrocyte indices.[17]

The general categorization of a morphological classification include

1. Macrocytic normochromic
2. Normocytic normochromic
3. Microcytic hypochromic

Classification of erythrocytes based on MCV

Normocytic : 80-100 fl

Microcytic : < 80 fl

Macrocytic : >100fl

Classification of erythrocytes based on MCHC

Normocytic : 32-36g/dl

Hypochromic : < 32g/dl

Hyperchromic : > 36g/dl

Among these various morphological variants of anemia normocytic normochromic is the most common cause. In the study done by Elhence P et al normocytic normochromic anemia is the most common pattern of anemia.[18]

NORMOCYTIC ANEMIA

Normocytic anemias are those in which the values for MCV are within normal limits, between 80 and 100 fl in adults. At times, however, the anemias that fall into this category also may be macrocytic or microcytic. For example, the anemia associated with hypothyroidism and liver disease may be either normocytic or slightly macrocytic. Also, because of reticulocytosis, the anemia associated with acute hemorrhage or chronic hemolysis may be normocytic or slightly macrocytic. The anemia of chronic disorders, although most often normocytic, is sometimes microcytic, and its pathogenesis is best understood in the context of microcytic anemias, as described above. Last, iron deficiency early in the course of anemia may be normocytic before becoming microcytic.[16]

Anemia of chronic disease

Numerous diseases are associated with anemia of chronic disease, but in many cases an underlying disease is not identified. The hematologic abnormality in anemia of chronic disease is an impaired ability to use the iron stored in the reticuloendothelial system. Erythrocytes are usually normochromic and normocytic, but about one third of patients with anemia of chronic disease have microcytosis,[19,20].

Anemia of renal disease

In renal insufficiency, a normochromic anemia develops which is very similar to Anemia of chronic disease (ACD). In this condition there is a deficiency of erythropoietin production, and there is evidence that the administration of erythropoietin, when the anemia is severe, can partially improve the condition[21,22]

Anemia of endocrine disorders

Endocrine disorders that commonly produce anemia are hypothyroidism, hypopituitarism, and adrenal insufficiency. In all three diseases the anemia is normocytic and normochromic but may be macrocytic.[23]

Anemia of liver disease

In liver diseases, a normocytic or macrocytic anemia develops. There are many causes of anemia in liver disease, including bleeding, iron deficiency, folate deficiency, hypersplenism, and sideroblastic anemia. In liver disease, however, serum iron is increased and there is an increase in transferring saturation. Serum ferritin is also increased and often reflects total body iron overload.[24].

Anemia of collagen vascular diseases

Anemia of chronic disease appears in collagen vascular diseases such as rheumatoid arthritis, polyarteritis, dermatomyositis, systemic lupus erythematosus, and temporal arteritis (including polymyalgia rheumatica).[25]

MICROCYTIC ANEMIA

Microcytic anemia is the one in which the MCV is less than 80fl .[17]

Iron deficiency anemia

Iron deficiency anemia, usually results from nutritional deficiency and other causes includes chronic gastrointestinal blood loss caused by nonsteroidal anti-inflammatory drug induced gastritis, ulcer, colon cancer, diverticulum or angiodysplasia[22]. Chronic blood loss from genitourinary tract cancer, chronic hemoptysis and bleeding disorders may result in iron deficiency but are much less common causes. Older persons may become iron deficient because of inadequate intake or inadequate absorption of iron.[26]

Other Causes of Microcytic Anemia

Anemias caused by abnormal hemoglobin (e.g. sickle cell anemia) are usually not first diagnosed in old age. Thalassemia minor, since it is asymptomatic, may first be found in old age. The anemia of chronic disease may be microcytic, but it is more commonly normocytic. Sideroblastic anemia may be microcytic, normocytic, or even macrocytic.[27]

MACROCYTIC ANEMIA

Macrocytic anemia is described as an anemia in which the MCV is greater than 100 fl. MCV increases slightly with increasing age but usually not enough to produce significant macrocytosis. Relatively few disorders routinely result in macrocytic anemia. The two common disorders that produce macrocytosis are megaloblastic anemias due to either vitamin B12 or folate deficiency. [27,28]

HEMOGLOBIN LEVELS IN DONORS

Blood donors are required to have a hemoglobin level of at least 12.5 g/dL or hematocrit of 38% in order to donate blood.[29] This is to ensure that donors have an adequate number of red blood cells (RBCs) for donation as well as adequate iron stores for erythropoiesis following donation.[29]

Being deferred from donation due to a low hematocrit during screening does not always mean the patient is anemic or has a medical problem. For example, male donors with a hematocrit below the acceptable 38% are considered anemic, but nonanemic women within the normal hematocrit range of 36-37% are not able to donate blood.[29]

Although this practice turns away non anemic women from donating blood, it reduces the chance of depleting their iron stores and potentially causing anemia following donation. Men are allowed to donate when slightly anemic because it is much easier for them to replace the iron lost during donation.[29]

DONATING BLOOD CAN CAUSE IRON DEFICIENCY

A healthy blood donor loses about 200-250 mg of iron per blood donation, constituting a roughly 6% in men and 9% in women with an average loss of 4.0g and 2.5g total body iron, respectively.[30] A double RBC donation, permitted every 16 weeks, results in the loss of up to 500 mg. The body compensates for this loss by mobilizing iron stores in the form of ferritin.[31]

For this reason, mean ferritin levels are significantly lower in blood donors than in non-donors and studies have shown that iron stores decline with repeated blood donation.[31] Men usually have the most dramatic drop in ferritin levels because of higher iron stores before donation. After 6-8 phlebotomies the ferritin level is about 40% lower than at baseline.[31] The proportion of male donors with decreased iron stores went from 8 to 19% with an increase from 5 to 6 donations per year.[31]

HEMOGLOBIN SCREENING METHODS

There is no consensus among blood banks on the best method for blood donor anemia screening.[32] In hospitals and laboratories, the gold standard for hemoglobin detection is the hemoglobincyanide method provided by automatic hematology analyzers.[33]

Screening tests for potential blood donors however require quicker, easier, and more cost-effective testing methods that do not require a venipucture.

The tests which are commonly used for primary screening are as follows,

1. Copper sulfate method
2. Microhematocrit method
3. Hemacue method
4. Automated hematology analyzer

Though the first three tests used for hemoglobin estimation are quick, easy, and relatively inexpensive, their sensitivity, specificity, and accuracy are lower than that of automatic hematology analyzers.[33] These tests are discussed in detail below.

CuSO4 (COPPER SULFATE) METHOD

This is a qualitative screening test based on specific gravity. The density of the drop of blood is directly proportional to the amount of hemoglobin it contains. The sample of donor's blood dropped into copper sulfate solution becomes encased in a sac of copper proteinate, which prevents any change in the specific gravity for about 15 seconds. If the hemoglobin is equal to or more than 12.5 gm/dL the drop will sink within 15 seconds and the donor is accepted.[34]

If the blood drop sinks to the middle and remains or starts to rise, a microhematocrit or comparable test is usually used to confirm the deferral. This is not a quantitative test and will only show that the hemoglobin is equal to, below, or above acceptable limits. Test results that indicate satisfactory hemoglobin levels are usually accurate, but some results that indicate low hemoglobin levels can be false. Repeating the test by a second method is sometimes used as confirmation.[34]

MICROHEMATOCRIT METHOD

Microhematocrit is a method for rapid determination of hematocrit done on an extremely small quantity of blood (one capillary tube of approximately 10 µL) by use of a capillary tube and a high-speed centrifuge. This method is a little more time consuming than other methods. Microhematocrit is often used to confirm failures with the CuSO4 method. A recent study shows a relatively poor correlation of the microhematocrit with the automated hematology analyzer. Anemia screening using this method failed to detect 35.7% of truly anemic donors.[34]

HEMOCUE METHOD

Some blood centers currently use portable equipment that is able to spectrophotometrically determine hemoglobin. These devices use a 10 µL capillary blood sample to determine hemoglobin by measuring the absorbance of azide methhemoglobin, using a cuvette containing a dry reagent system and a dual wavelength photometer. There was a relatively poor correlation of HemoCue 201(+) with the automated hematology analyzer. However, this method was more accurate (56%) in detecting anemia in prospective female blood donors than the microhematocrit method.[34] The HemoCue 201(+) and the microhematocrit method were equivalent in their donor deferral rate.[34]

EFFECT OF WHOLE BLOOD DONATION ON THE IRON STATUS

Blood donation poses a risk of iron deficiency to blood donors. Iron is an important element of the Hb protein, which is found in red blood cells. In humans, most iron is found in red blood cells, incorporated in Hb. Red blood cells contain about 60-75% of the total body iron. In adults, the total body iron content is normally 3-5 g with typically higher values in men than in women.[35]

With a blood donation, a substantial amount of iron is lost. A donation of 500 ml whole blood contains about 200-250 mg iron, which is 4-8% of total body iron. When the iron intake is not sufficient to replenish the iron loss due to donation, a negative iron balance occurs. Subsequent blood donations may then gradually lead to iron deficiency.[36]

In Norashikin J et al study there was a significant correlation between the frequency of donations and serum ferritin levels.The mean ferritin level decreased tremendously in regular blood donors as early as after the first 10 donations and remained stable after 20 or more donations.[30]

Three stages of iron deficiency can be distinguished: iron depletion, iron deficient erythropoiesis and iron deficiency anemia.[37]

Iron depletion is marked by running out iron stores. In the stage of iron deficient erythropoiesis, the iron supply to the erythropoietic bone marrow is becoming insufficient for erythropoiesis. However, Hb levels are still normal. When finally the iron supply becomes insufficient to produce a normal amount of Hb, iron deficiency anemia becomes apparent. Iron depletion and iron deficient erythropoiesis are thus sub-clinical stages; with anemia clinical symptoms appear. The primary

function of Hb is oxygen transport through the bloodstream from the lungs to all other tissues in the body, and one of the first symptoms of anemia is decreased fitness through a diminished oxygen supply to the body tissues. Furthermore, as iron is also an important element of several other proteins, iron deficiency also affect DNA synthesis,[38,39] the immune system[40] and energy metabolism through impaired mitochondrial electron transport.[41,42]

Blood donors on average need several weeks to replenish the lost iron after a blood donation.[43] However, there are wide variations in the duration of the recovery period among individual donors. The European guidelines with relation to a minimum donation interval and a maximum number of donations per year may therefore not be safe for each individual donor. Indeed, depleted iron stores in blood donors are not uncommon[44]and also iron deficient erythropoiesis occurs.[45,46]

DONOR DEFERRAL DUE TO LOW HEMOGLOBIN

To protect donors from developing iron deficiency anemia after blood donations, the iron status of blood donors is assessed prior to donation. Most commonly, this is done by measuring Hb levels. Donors with low Hb levels are deferred from donation to prevent anemia afterwards. Furthermore, deferral of donors with low Hb levels also ensures that blood units for transfusion meet the required standards for Hb content.[47]

A substantial number of donors is deferred from donation because of low Hb levels. In 2011, Dutch male donors were 12,583 (2.2%) deferred because of a low Hb level and for Dutch female donors this number was 23,768 (5.5%). Although deferrals are meant to protect donors, they are also demoralizing for donors. As a consequence, the risk of donor lapse is increased due to deferral.[48,49]

Timely estimating the risk of Hb deferral in blood donors, i.e. before being invited, could be helpful in the management of the donation program and the retention of donors. At the individual level, such predictions of Hb deferral risk may guide the decision whether a donor can be invited for the next donation, or whether it is better to postpone the invitation. From a management perspective, these predictions may decrease the number of donor deferrals for low Hb levels. [31]

FACTORS ASSOCIATED DONOR DEFERRAL WITH LOW HEMOGLOBIN

Several factors are known to be associated with low Hb levels or Hb deferral. Demographic characteristics such as sex and age are associated with Hb levels. Hb levels rise substantially during childhood. In men, there is a small decrease in Hb levels with increasing age.[50]

Whereas in women, Hb levels rise by the effect of menopause due to hormonal changes and the cessation of iron loss through menstruation.[50]

Despite lower Hb cutoff levels for donation for women in most countries, Hb deferral occurs more frequently in women than in men.[51]

Body mass index (BMI) is also associated with Hb levels: a greater BMI is associated with higher Hb levels.[51] Likewise, blood volume might be associated with Hb deferral in blood donors. The amount of blood given with a whole blood donation is around 500 ml. Donors with a large blood volume lose relatively less blood with a blood donation compared to donors with a small blood volume. Therefore, it is likely that donors with a large blood volume need less time to recover after a blood donation and have a smaller risk of Hb deferral at the next visit to the blood collection center.[51]

Another factor that is associated with Hb deferral is seasonality. Hb levels decrease with increasing daily temperature and are thus lower in warmer seasons. Consequently, in summer months deferral rates are higher.[52,53]

Furthermore, specific characteristics of the donation history might be associated with Hb deferral. Hb levels measured at previous visits to the blood collection center are obviously associated with current Hb levels and previous Hb deferral is likely associated with Hb deferral at a next visit. The longer the time interval between two donations, the more time for the donor to recover from the previous donation; thus time interval between two subsequent donations or visits is also associated with Hb deferral. Studies have shown that an increased donation frequency is associated with lower iron stores.[36,44]

Finally, values of other iron parameters in blood might be associated with Hb deferral. Hb levels are only low in an advanced stage of iron deficiency. As a consequence, it may occur that donors pass the Hb screening test while they have already depleted iron stores or even iron deficient erythropoiesis,[36,37,44,45] as these conditions remain undetected with Hb screening. Especially these donors are at high risk of developing iron deficiency anemia after a blood donation and are more likely to be deferred at their next visit to the blood collection center. Iron parameters that respond in an early stage to a low iron status may therefore be predictive for Hb deferral. There are several iron parameters available to assess iron depletion or iron deficient erythropoiesis. Iron depletion can be assessed by measuring serum ferritin levels.[54] Tests for the diagnosis of iron deficient erythropoiesis include measurement of plasma iron, total iron binding capacity or transferrin, transferrin saturation, soluble transferrin receptor (sTfR) concentration,[55] the sTfR index (sTfR divided by log-transformed ferritin values)[56,57] and zinc protoporphyrin (ZPP).[58]

Another iron parameter with which sub-clinical iron deficiency can be assessed is the recently discovered iron regulatory protein hepcidin.[59,60] Each of the above mentioned tests has its own advantages and disadvantages. ZPP is measured by an automated technology and its attractive features are its ability to perform immediate point-of-care assays and its relative low price. It may therefore especially be useful for donor screening.[58]

DIAGNOSING AND MANAGING ANEMIA AFTER DEFERRAL

It is also important to inform prospective donor that a low hematocrit is not an arbitrary measurement only related to blood donation. Rather, a low hematocrit level or anemia may be an important diagnostic sign that points to a serious and possibly treatable medical condition. The symptoms of anemia can include headache, fatigue, weakness, and difficulty in thinking. With severe anemia, other symptoms, such as shortness of breath and rapid heartbeat, may be experienced. There are several possible reasons that a patient may be anemic and they generally include iron and vitamin deficiencies, chronic illnesses, and gastrointestinal bleeding. Differentiating the types of anemia is important in planning diagnostic testing and in guiding therapy.[31]

Iron and Vitamin Deficiency Anemia

Because it is the most common cause of anemia, iron deficiency must be considered in the evaluation of any anemic patient. Depending upon the criteria used for the diagnosis of iron deficiency, approximately 4-8% of premenopausal women are iron deficient. In men and postmenopausal women, iron deficiency is uncommon in the absence of bleeding.[29]Oral iron supplements, and less frequently, parenteral iron, are used to treat iron deficiency. Supplements are especially important when a patient is experiencing clinical symptoms of iron deficiency anemia to replenish normal iron stores to raise hematocrit levels. A complete overview of oral iron supplements can be found in feature articles.[29]

Existing Chronic Illness and Gastrointestinal Condition

Patients with an existing chronic illness or gastrointestinal (GI) condition are at risk for a low hematocrit level and anemia. If the underlying chronic illness can be effectively treated, the anemia often improves. Conditions such as infections, inflammation, and cancer particularly suppress production of red blood cells in the bone marrow. Renal insufficiency or damage can also lead to low hematocrit level and anemia. There are no specific laboratory tests, so the diagnosis is typically made by excluding other causes. No specific treatment exists for this type of anemia, so treatment of the underlying disorder is usually the first step. Erythropoiesis-stimulating agents (ESAs) may be used to treat more severe cases.[131]

Conditions affecting the digestive tract can often cause bleeding, which commonly lead to anemia. Some conditions which can cause GI bleeding include stomach ulcers, growths in the intestine (polyps), colon cancer, and other less common diseases of the digestive tract. Certain medications can also cause bleeding of the digestive tract. Detection and correction of the source of GI bleeding is the first and most important step in treatment.[31]

Unknown Chronic Illnesses or Gastrointestinal Blood Loss

Anemia is an important sign that can point to a serious and possibly treatable medical condition, such as GI bleeding or a chronic illness like kidney disease or cancer. Non-menstruating women with a blood count below 36%, and men with a blood count below 38% should be evaluated further if they donate blood fewer than 3 times per year and are not already under care for a chronic illness or condition of the

digestive tract. Failure to recognize and evaluate anemia in these patients could lead to delayed diagnosis of treatable conditions.[31]

Although being deferred from donation can be frustrating for the patient, it can call attention to a serious medical condition which otherwise would have gone unnoticed. It may also represent an opportunity for you to educate them on the need for a healthy lifestyle, including a balanced iron-rich diet, and vitamins or supplements if needed. With the right information and help from you, many deferred blood donors can learn how to raise their blood counts, stay healthy, and try to donate again.[31]

PRE DONATION SCREENING FOR TRANSFUSION AND TRANSMISSIBLE DISEASES

All whole blood and apheresis donations should be screened for evidence of infection prior to the release of blood and blood components for clinical or manufacturing use.[61]

Screening of all blood donations should be mandatory for the following infections and using the following markers:

- HIV-1 and HIV-2: screening for either a combination of HIV antigen-antibody or HIV antibodies
- Hepatitis B: screening for hepatitis B surface antigen (HBsAg)
- Hepatitis C: screening for either a combination of HCV antigen antibody or HCV antibodies
- Syphilis (*Treponema pallidum*): screening for specific treponemal antibodies.[13]

Screening of donations for other infections, such as those causing malaria, Chagas disease or HTLV, should be based on local epidemiological evidence.

Where feasible, blood screening should be consolidated in strategically located facilities at national and/or regional levels to achieve uniformity of standards, increased safety and economies of scale.[13]

MATERIALS AND METHODS

7.1 SOURCE OF DATA:

It is a prospective study. In this study the donors presented to blood bank of BLDE University's Shri B. M. Patil Medical College, Hospital and Research Centre. Bijapur have been taken. The duration of study is from 1st October , 2012 to 30th June , 2014.

INCLUSION CRITERIA:

Prospective donors presented to Blood bank, who are deferred due to anemia in BLDE University, Shri B. M. Patil Medical College, Hospital and Research centre, Bijapur have been included in the study.

EXCLUSION CRITERIA:

Prospective donors presenting to blood bank who are deferred due to causes other than anemia e.g fever, underweight, enteric fever, pregnancy, hypertension, hepatitis B , hepatitis C, malaria, syphilis, HIV etc in BLDE University, Shri B. M. Patil Medical College, Hospital and Research centre, Bijapur have been excluded from the study.

METHOD OF COLLECTION OF DATA:

Detailed clinical history & examination of the prospective donor was done & relevant informed consent was taken.5 ml of venous blood sample in ethylene diamine tetraacetic acid [EDTA] was collected from all blood donors and this sample was processed in automated hematology analyzer, Sysmex KX-21.

The following paramaters are used for analysis of the pattern of anemia in donors.

Hemoglobin (g/dl)

Packed cell volume [PCV],

RBCs indices -: MCV, MCHC, MCH .

Peripheral smear study stained with Leishmans stain

Procedure of Leishmans stain

1) Air dried film was flooded with leishman's stain for 2 minutes.
2) Double the volume of buffer water was added for 10 minutes.
3) The stain was washed off with distilled water until it acquired a pinkish tinge.
4) After drying, the back of the slide was wiped clean.
5) The slide was mounted with D.P.X

SAMPLING TECNIQUE: Using systematic random sampling technique data is drawn from total donor population.

STATISTICAL ANALYSIS:

Data is analyzed by tables and Diagramatic (pie chart /bar diagram) representation.

Patterns of anemia are classified based on RBC indices [17]& further correlated by peripheral smear.

Microcytic anemia was defined as MCV below 80 fl,

Normocytic as MCV between 80 and 100 fl and

Macrocytic anemia by an MCV above 100 fl**.**

& then correlated by peripheral smear.

Severity of anemia graded as mild, moderate & severe [5]

Grade 1 (Mild) : 10-12.5gm / dl

Grade 2 (Moderate) : 7-10 gm / dl

Grade 3 (Severe) : More than 7 gm / dl

.

FIGURE 1: HEMATOLOGY ANALYSER SYSMEX KX-21.

RESULTS

VARIOUS CAUSES OF DEFERRAL(Table 1)

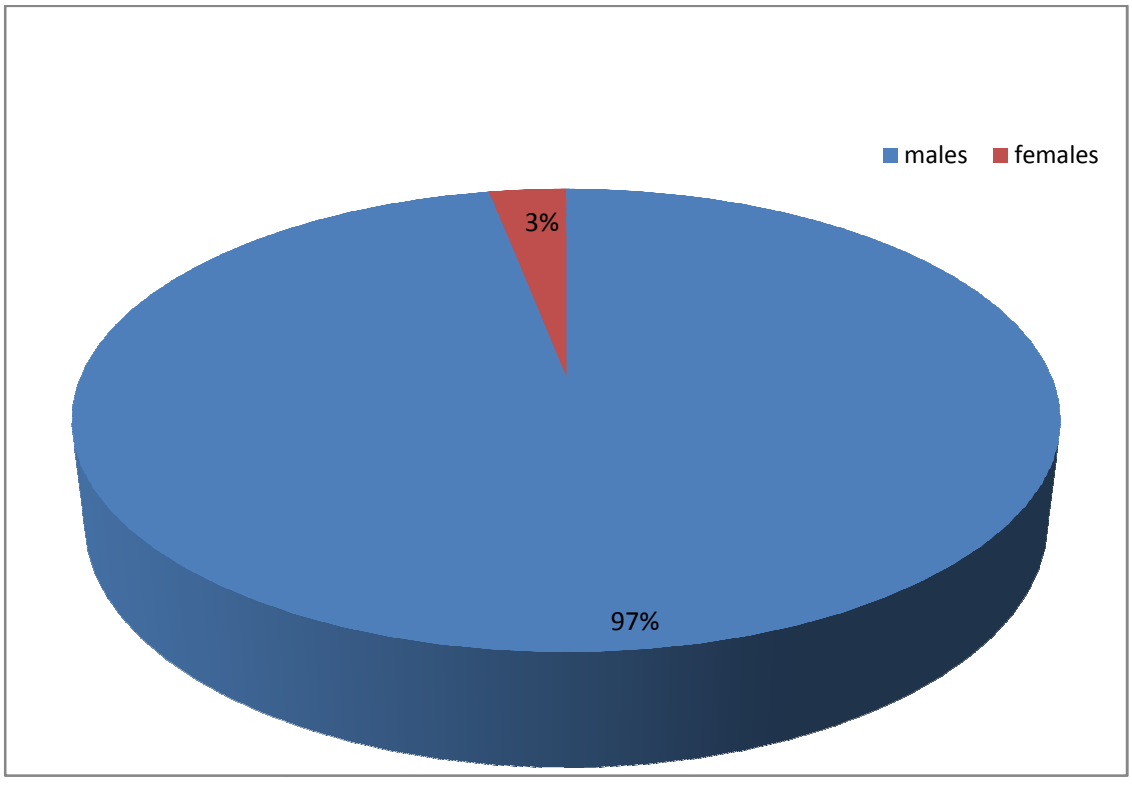
Causes	Number	Percentage %
Anemia	130	48.8
Seropositive for HBsAg	77	28.9
Hypertension	20	7.5
Seropositive forHCV antibodies	11	4.1
Seropositive for HIV	10	3.7
Weight less than 45kg	10	3.7
H/o drug intake	03	1.1
Syphilis	02	0.7
Alcohol	02	0.7
Dog bite	01	0.3
Total deferrals	266(4063)	6.5

Among the various causes of deferral anemia is found to be the common cause of deferral in this study accounting 48.8% among various deferrals

DISTRIBUTION OF STUDY SUBJECTS ACCORDING TO SEX (Table 2)

GENDER	TOTAL DONORS	DONORS DEFERRED DUE TO ANEMIA
MALE	3858	103
FEMALE	205	27
	4063	130

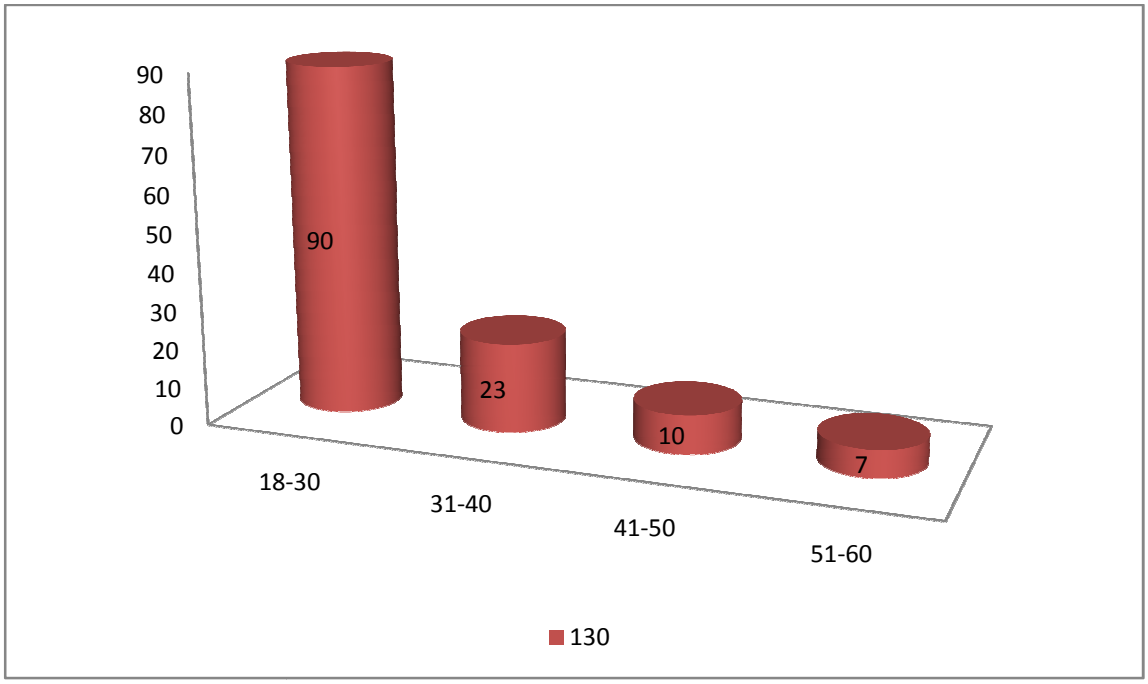
FIGURE 2: PIE CHART SHOWING DISTRIBUTION OF STUDY SUBJECTS ACCORDING TO SEX



DISTRIBUTION OF STUDY SUBJECTS ACCORDING TO AGE (TABLE 3)

Age	No
18-30	90
31-40	23
41-50	10
51-60	07

FIGURE 3: BAR DIAGRAM SHOWING DISTRIBUTION OF STUDY SUBJECTS ACCORDING TO AGE



The mean age of deferrals due to anemia is found to be 18-30 years **.**

PREVALENCE RATE OF ANEMIA IN DONOR DEFERRALS (TABLE 4)

Prevalence of anemia in donors	3.1%
Prevalence of anemia in male donors	2.6%
Prevalence of anemia in female donors	13.2%

Prevalence rate of anemia in donors is 3.1%.Prevalence rate of anemia in female donor deferrals is more as compared to males that is 13.2% and 2.6% respectively.

DISTRIBUTION OF STUDY SUBJECTS ACCORDING TO SEVERITY OF ANEMIA (Table 5)

Sex	Mild	Moderate	Severe
Males(103)	84	18	01
Females(27)	18	08	01
Total(130)	102	26	02
Percentage (%)	78	20	02

Lowest hemoglobin found was 6.5g/dl in females with MCV of 65.9fl and 6.8g/dl in males with MCV of 69.4fl

FIGURE 4: PIE CHART SHOWING DISTRIBUTION OF STUDY SUBJECTS ACCORDING TO SEVERITY OF ANEMIA

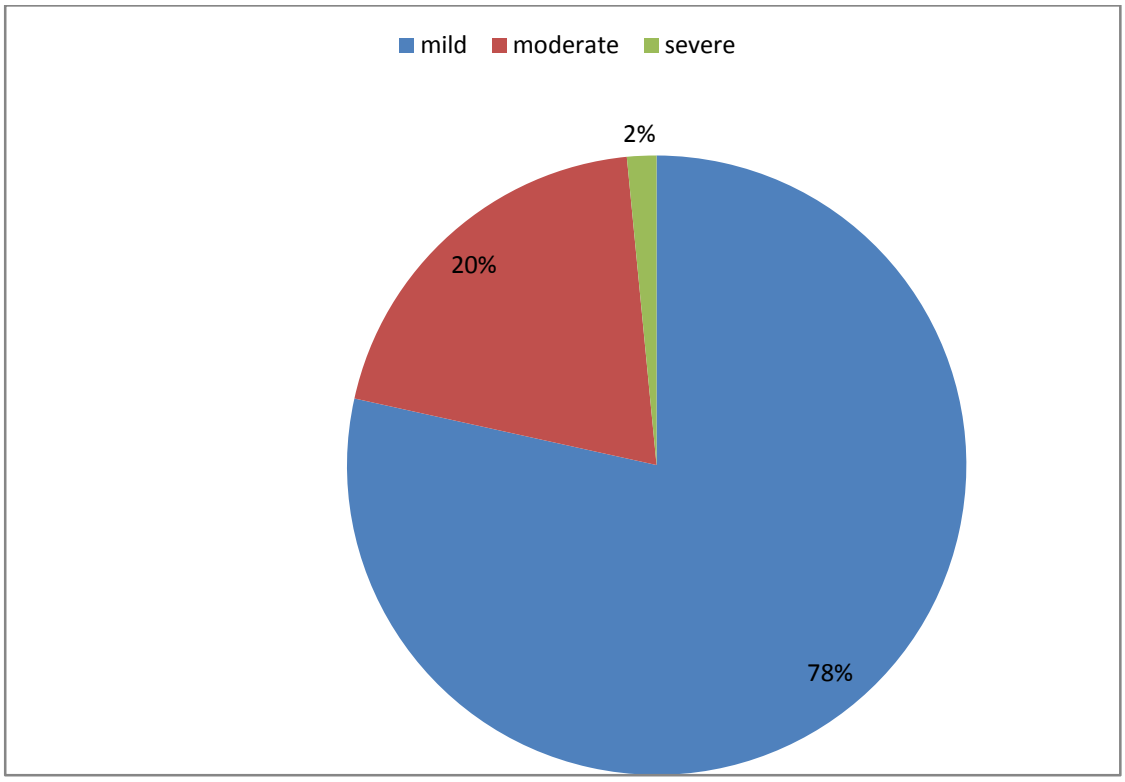
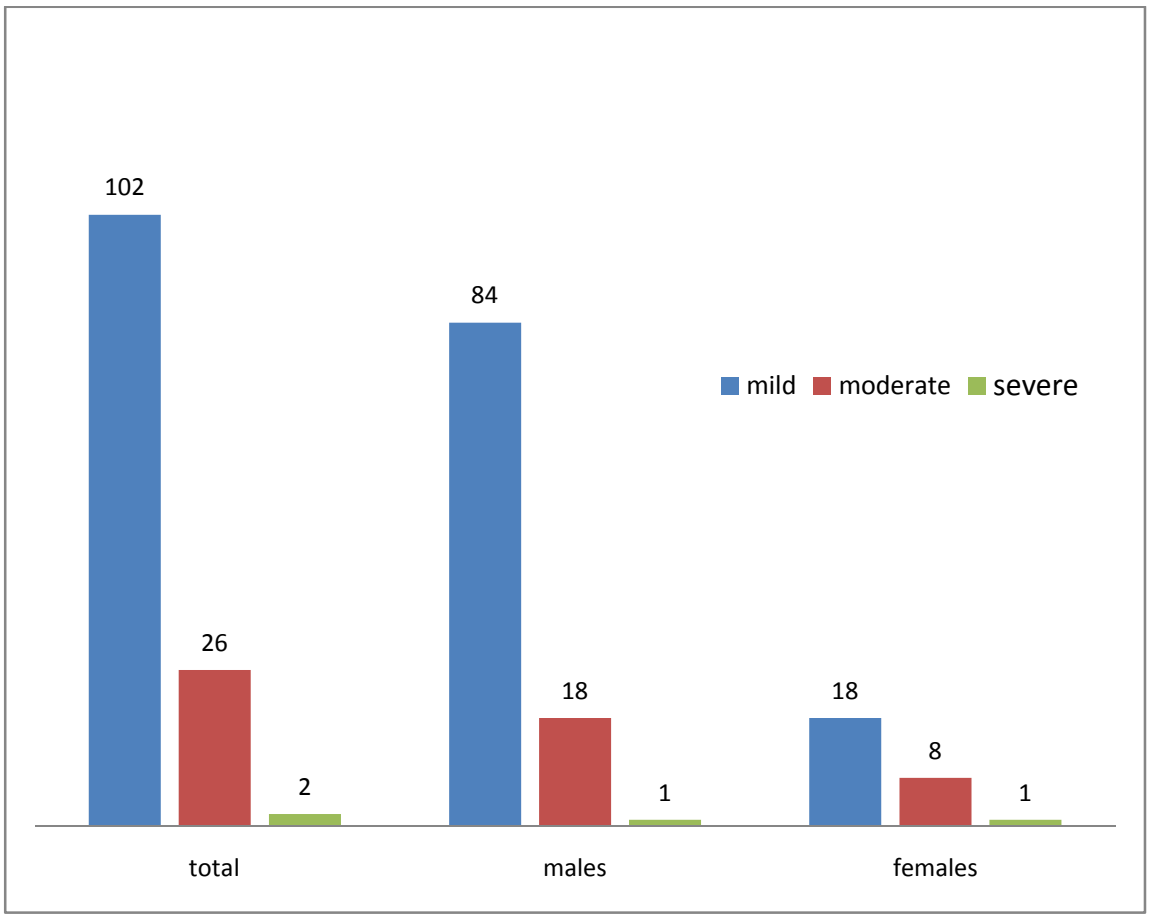


FIGURE 5: BAR DIAGRAM SHOWING DISTRIBUTION OF STUDY SUBJECTS ACCORDING TO SEVERITY OF ANEMIA

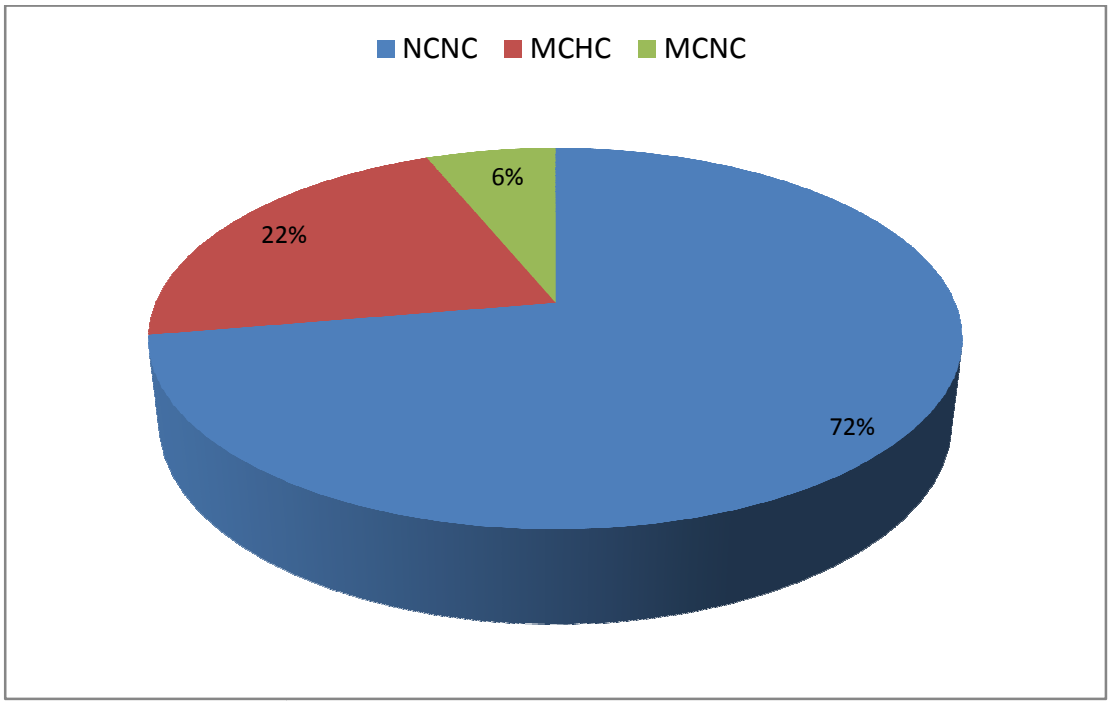


RESULTS OF MORPHOLOGICAL PATTERNS OF ANEMIA(Table 6)

Most common morphological pattern of anemia observed in this study is normocytic normochromic type.

Type of anemia	No	Percentage
Normocytic normochromic anemia(NCNC)	94	72.3
Microcytic hypochromic anemia(MCHC)	28	21.5
Macrocytic normochromic anemia(MCNC)	08	06.2

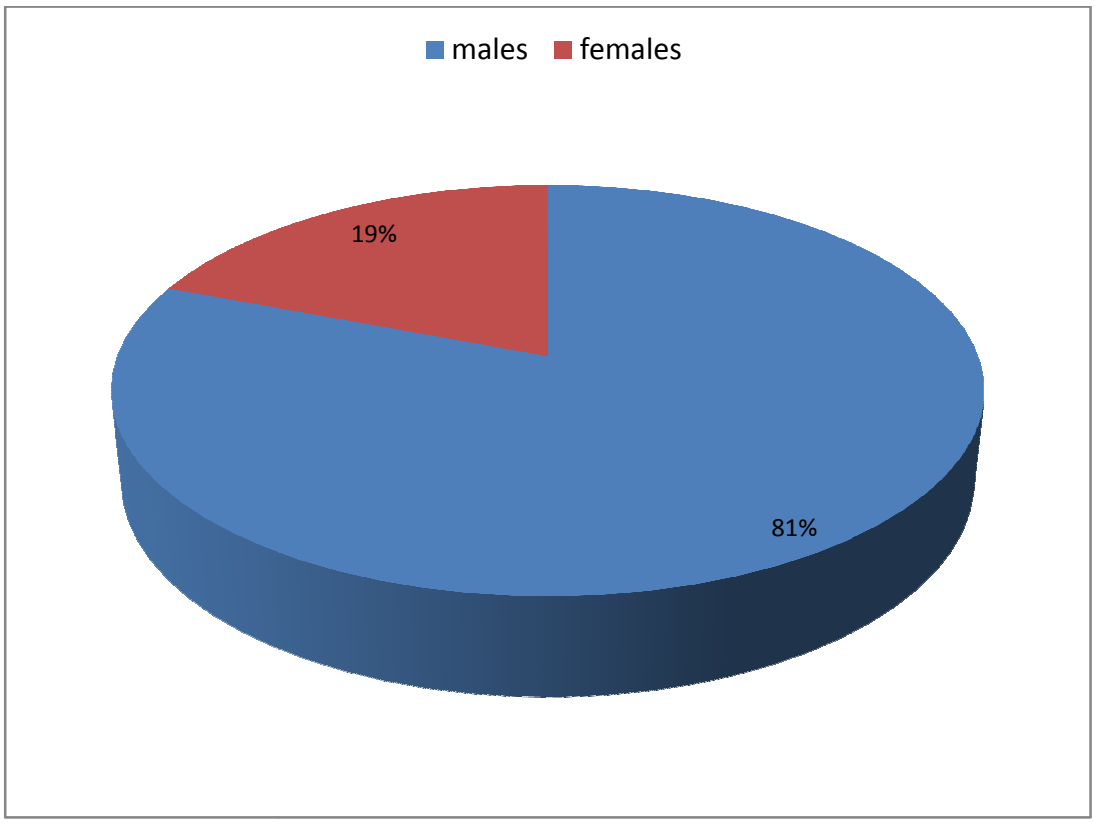
FIGURE 6:PIE CHART SHOWING RESULTS OF MORPHOLOGICAL PATTERN OF ANEMIA



DISTRIBUTION OF NORMOCYTIC NORMOCHROMIC ANEMIA ACCORDING TO SEX (Table 7)

Gender	Number	Percentage
Males	76	80.9
Females	18	19.1

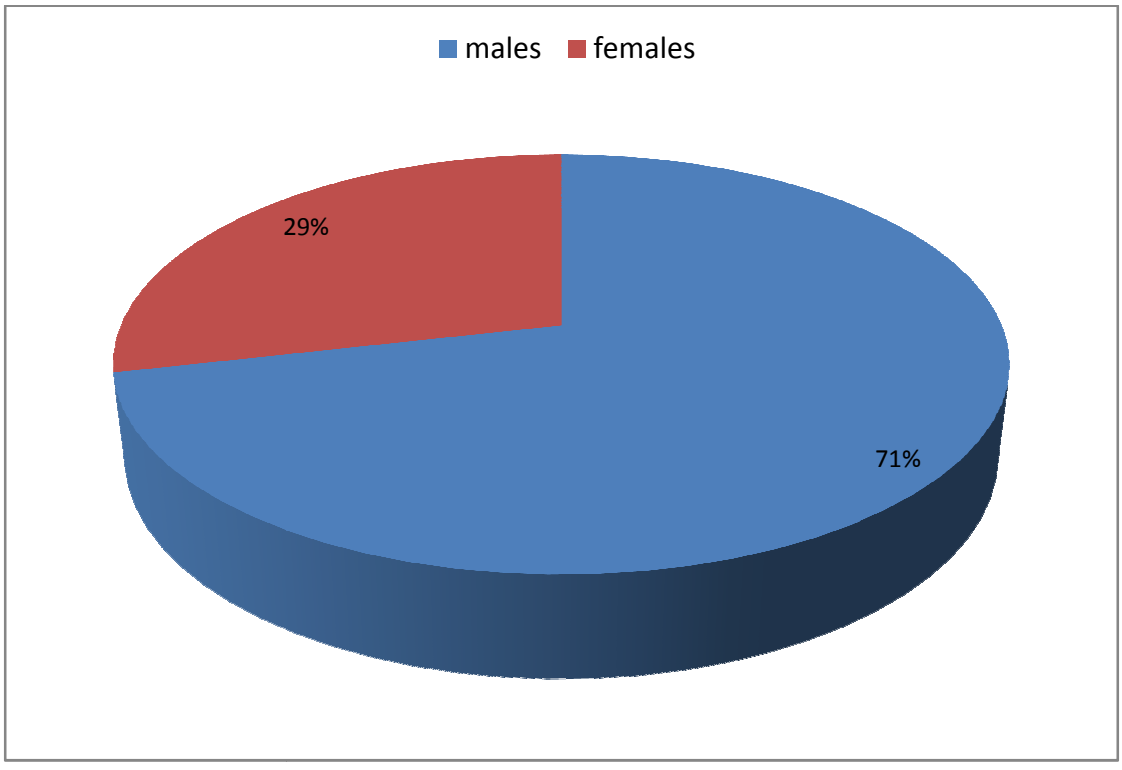
FIGURE 7: PIE CHART SHOWING DISTRIBUTION OF NORMOCYTIC NORMOCHROMIC ANEMIA ACCORDING TO SEX



DISTRIBUTION OF MICROCYTIC HYPOCHROMIC ANEMIA ACCORDING TO SEX (Table 8)

Gender	Number	Percentage
Males	20	71.4
Females	08	28.6

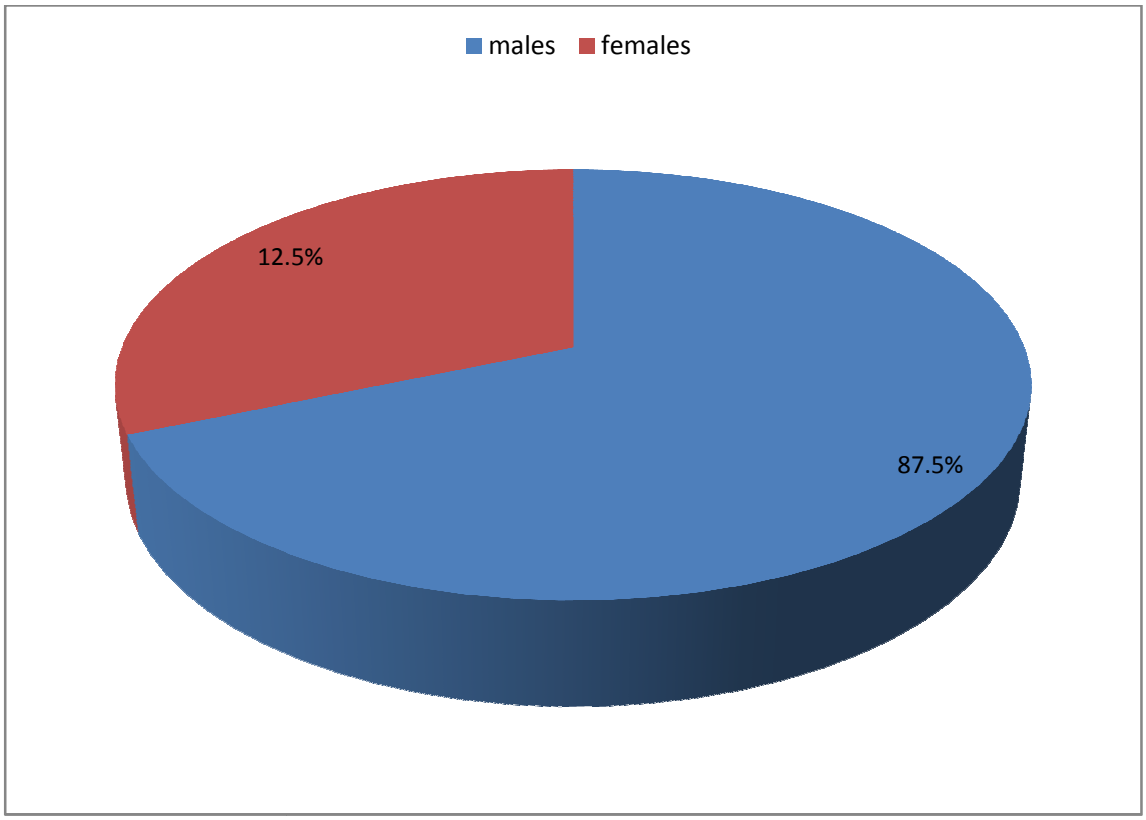
FIGURE 8: PIE CHART SHOWING DISTRIBUTION OF MICROCYTIC HYPOCHROMIC ANEMIA ACCORDING TO SEX



DISTRIBUTION OF MACROCYTIC NORMOCHROMIC ANEMIA ACCORDING TO SEX (Table 9)

Gender	Number	Percentage
Males	07	87.5
Females	01	12.5

FIGURE 9: PIE CHART SHOWING DISTRIBUTION OF MACROCYTIC NORMOCHROMIC ANEMIA ACCORDING TO SEX



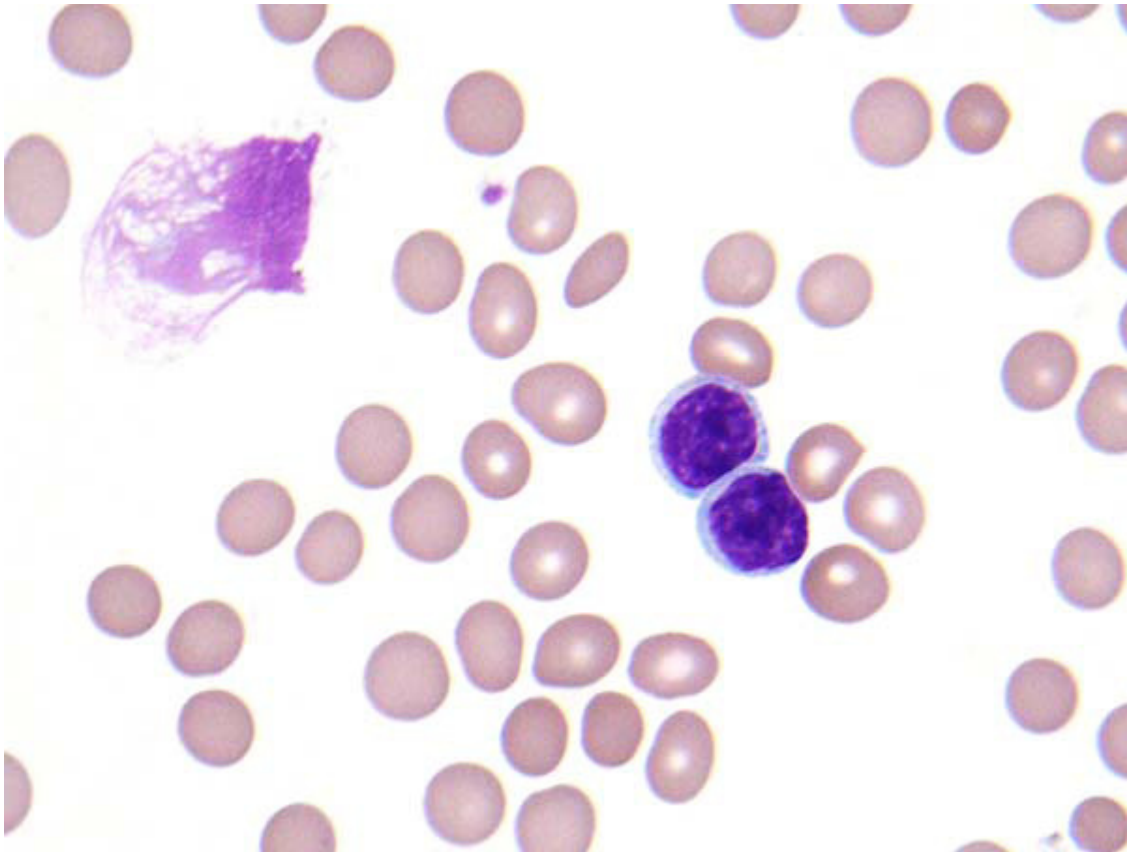


FIGURE10: NORMOCYTIC NORMOCHROMIC ANEMIA - LEISHMAN'S STAIN (100X)

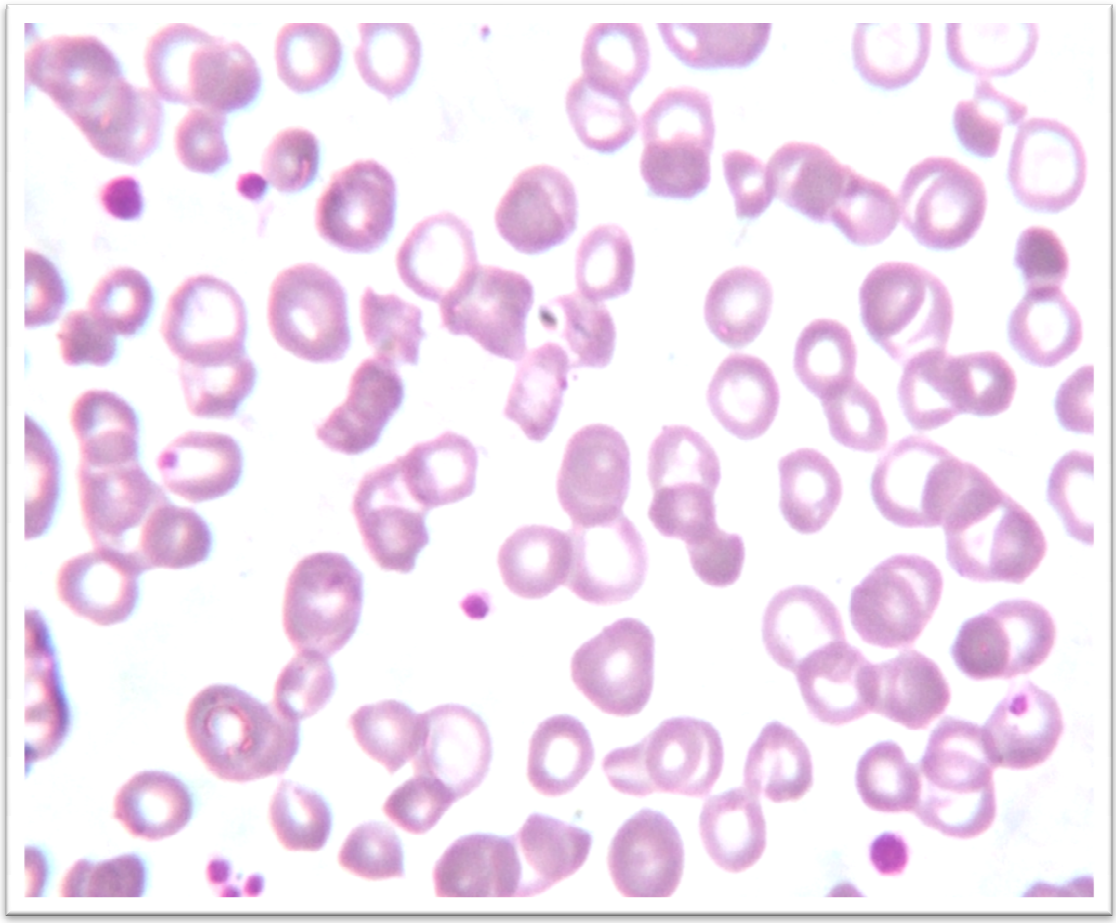


FIGURE 11: MICROCYTIC HYPOCHROMIC ANEMIA LEISHMAN'S STAIN (100X)

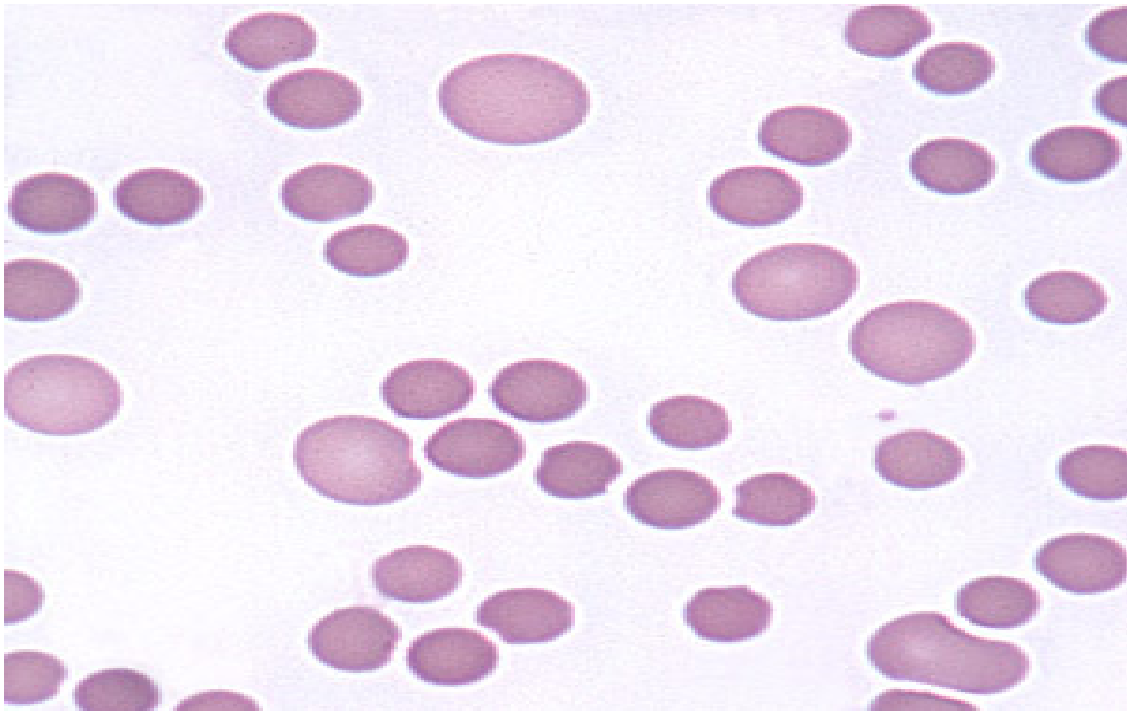


FIGURE – 12. MACROCYTIC ANEMIA – LEISHMAN'S STAIN (100X)

DISCUSSION

The paucity of healthy, safe blood donors has been a serious problem in the blood banks worldwide.[1] India is in deficit of approximately 3 million units of blood annually. While losses resulting from consequences of rigorous screening for transfusion transmitted infections have been the focus of our attention for more than a decade, other reasons for donor deferral have not received as much attention.[2]

Hemoglobin assessment is an important criterion for blood donor selection. The minimal hemoglobin cutoff is set at 12.5 gm%, which is done to ensure both donor safety and appropriate hemoglobin content in the donated unit. A healthy blood donor loses about 200–250 mg of iron per unit of blood donated, constituting to roughly 6% and 9% iron loss in men and women with an average of 4.0 g and 2.5 g total body iron, respectively.[5] The body compensates for this loss by mobilizing iron stores in the form of ferritin. For this reason, the mean ferritin levels are significantly lower in blood donors than in non-donors and studies have shown that iron stores decline with repeated blood donation.[6]

There is no consensus among blood banks on the best method for blood donor anemia screening.[34] In hospitals and laboratories, the gold standard for hemoglobin estimation is the use of automated hematology analyzer.

Screening tests for potential blood donors, however, require quicker, easier, and more cost-effective testing methods that do not require a venipuncture and cause

minimal discomfort to the donor. Three tests that are commonly used for primary screening are Copper sulfate method, Hemocue, and Microhematocrit, which uses a capillary tube and high speed centrifuge. Although these tests are quick, easy, and relatively inexpensive, their sensitivity, specificity, and accuracy are lower than that of an automated hematology analyzer.[34]That is why at our center, we used Copper sulfate as primary screening methods, but the results were ultimately confirmed by running the EDTA venous sample of the subject on an automated analyzer.

In this study, we analyzed donor deferral patterns in an attempt to provide insight into the reasons for donor deferral, where blood donors are usually just relatives of patients admitted to the hospita.[2]

Safe donor selection is the first step towards safe transfusion services. National and international efforts are on to ensure safe blood supply through screening, education and strict criteria laid down by the Directorate General of Health Sciences, Ministry of Health and Family Welfare (2003) and Eligibility criteria for blood donation American Red Cross.[1]

Studies done by Bahadur et al, Ramesh et al[5,6,18] female donor population was very low i.e. 1.7% and 10% respectively similar to present study with female donor population of 5%% **[Table 2]** .This less population of female donors can be attributed due to ignorance, fear, lack of awareness and motivation among them. Most common cause of deferral was anaemia with common morphological pattern of microcytic hypochromic anemia among females[5,6] reflecting ill health, poor

nutritional status and higher prevalence of anaemia which is also observed in our study

Among the various causes for donor deferral, the most common cause is anemia that is 15.5% to 32.9%, Prevalence rate of anemia is found to be 1.8%.[5]

The most common cause for deferral was low hemoglobin (32.9%) **in study done by Bahadur et al** similar to that reported in Turkish donors by Arslan (2007) (20.7%) and Custer *et al* (2004) (60% of temporary deferrals) and Halperin *et al* (1998) (46%). The second most common cause of deferral was low weight, which accounted for 59.4% of total deferrals. Hemoglobin and low weight combined accounted for 65.3% of total deferred similar to the findings by Chaudhary *et al* (2008). Most of these deferred donors (89.7%) were age 18-40 years old. This highlights the fact that a sizeable proportion of youth in this part of the world are malnourished, reflecting the impact of low socioeconomic status on the health of Indian youth.[6, 12-16]

In present study commonest cause of deferral was anemia with male predominance which was also observed by the study done by **Shalini Bahadur et al**[5]

The percentage of donors deferred due to anemia was estimated to be 15.5%, the prevalence rate was 1.8%, in present study percentage of donors deferred due to anemia ia 48.8% **[Table 1}**which is more as compared to Shalini Bahadur et al; the prevalence rate was found to be 3.1%[**Table 4**].

In the study done by **Elhence P et al ,**the prevalence rate was found to be 2.5% Most common cause of anemia was normocytic normochromic anemia . Among females it was microcytic hypochromic anemia. The present study the common pattern of anemia is normocytic normochromic [Table6] overall. Microcytic hypochromic anemia was common in females[Table 8]. These findings are in concordance with study done by **Elhence P et al**[18]

In the study done by **Timothy Acnor Ekwere et al**[62] and **Nagrekha et al 63** anemia was the common cause of temporary deferral i.e 39% and 34.3% respectively and decreased weight as second common cause of temporary deferral which is in concordance with our study results which showed anemia(48.8%) **[Table 1]** as first common cause and decreased weight (3.7%) **[Table 2]** as second common cause among the temporary deferrals.

In the study done by **Ramesh S Patil et al**[64]in Solapur district anemia was found to be the common cause of temporary deferral (64.9%) with prevalence rate of 3.95% which is in concordance with our study which showed anemia as common cause(48.8%) **[Table 1]** and prevalence rate in our study is 3.1% **[Table 4].**

In the study done by **Girish P J**[1] et al most common cause was found to be hypertention(39.9%) followed by anemia(19.4%) which is in contrast with present study with anemia as most common cause followed by decreased weight**[Table 2].**

The present study has also showed that higher percentage of donors are deferred on account of being positive for Hepatitis B i.e 28.9% **[Table 1]** in concordance with the study done by **Rehman et al**[11] with 23.5% of donors deferred for being positive for Hepatitis B and is in discordance with **Ramesh p et al**[64] study with only 0.5% of donors were being deferred for Hepatitis B infection.

SUMMARY

- Present study was conducted as prospective study from Oct 2012-June2014
- Total 4063 donors were Registered
- Out of 4063 donors, 266 donors were deferred
- Commonest cause for donor deferral is anemia i.e. 130 donors constituting (48.8%).
- Prevalence rate of anemia among donor deferrals due to anemia is 3.1%
- Male deferrals are predominant.
- Commonest morphological pattern of anemia is normocytic normochromic anemia.
- Other causes for deferral are underweight, hypertension and infections .

CONCLUSION

Anemia is the most common cause for temporary donor deferral.

Timely diagnosis & treatment of such donor deferrals, helps in reducing the strain to a certain extent on the demand & supply mis-match by converting the temporary deferral to a healthy donor status.

Analysis of rejection patterns may help medical personnel to be more focused in donor screening. This will not only help in improving donor and recipient safety but also in maintaining a healthy donor pool in the long run, provided the potential donors are appropriately counselled and managed to improve the efficiency of the donor program. Temporary donor deferrals need to be actively and aggressively managed so as not to lead to a diminished supply of future donors.

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ANNEXURES

MASTER CHART

Sl.No	Age	Sex	Weight	Hb	Hct	MCV	MCH	MCHC	Plt	Type of anemia on PS
1	43	F	95	12	37.4	91.7	29.4	32.1	2.78	NCNC
2	18	M	63	11.9	37.8	67.7	21.3	31.5	2.89	MCHC
3	19	F	62	12.4	38	85.8	28	32.6	4.53	NCNC
4	25	F	62	9	26.1	102.4	35.3	34.5	4.02	MCNC
5	22	M	52	11.6	34.8	96.7	32.2	33.3	3.02	NCNC
6	18	M	56	11.3	35.4	80.1	25.6	31.9	2.04	NCNC
7	20	M	85	10.7	35.5	63.4	19.1	30.1	2.5	MCHC
8	21	M	55	11.7	37.4	106.9	33.4	31.3	2.12	MCNC
9	19	M	50	10	29.5	88.9	29.9	33.9	2.58	NCNC
10	22	M	54	10.4	32.2	90.4	29.2	32.3	1.93	NCNC
11	22	F	60	8.6	27	97.5	31	31.9	1.94	NCNC
12	21	M	76	11.3	36	96.3	30.2	31.4	2.69	NCNC
13	19	M	60	12.2	41	79.9	23.8	29.8	2.8	NCNC
14	19	F	50	11.5	35.2	78.2	22.1	27.6	3.2	NCNC
15	38	F	55	6.5	24.5	65.9	17.5	26.5	4.44	MCHC
16	28	M	58	9.6	33.5	86.6	24.8	28.7	3.5	NCNC
17	32	M	62	9.6	32.5	85.8	25.7	29.9	4.11	NCNC
18	27	M	60	10.3	32.9	81	25.4	31.3	2.62	NCNC
19	37	M	65	10.1	35.7	83	23.5	28.3	3.44	NCNC
20	37	M	65	11.3	34.9	100.6	32.6	32.4	2	MCNC

21	23	M	66	11.7	37.4	86.2	27	31.3	2.3	NCNC
22	21	M	53	11.1	36.4	80.7	24.6	30.5	3.06	NCNC
23	22	M	56	11.5	41.5	75.2	20.8	27.7	2.44	MCHC
24	21	M	68	10.9	34.9	86.6	27	31.2	2.33	NCNC
25	34	M	69	11.6	41.1	91.1	25.7	28.2	2.54	NCNC
26	25	M	61	11.3	39.5	77.3	22.1	28.6	3.5	MCHC
27	29	M	55	11.7	38.1	83.9	25.8	30.7	2.75	NCNC
28	25	M	70	11.8	39.6	81.5	24.3	29.8	3.38	NCNC
29	21	M	58	12.1	40.6	84.1	25.1	29.8	2.75	NCNC
30	39	M	65	12.4	40.9	82.1	24.9	30.3	2.89	NCNC
31	32	M	70	12.4	40.7	80.4	24.5	30.5	2.65	NCNC
32	26	M	68	11.3	39.4	74.3	21.3	28.7	3.25	MCHC
33	28	M	63	12.1	40.4	79.7	23.9	30	2.78	NCNC
34	20	M	58	12.4	40.5	86.9	26.6	30.6	2.21	NCNC
35	27	M	63	11.7	39.3	80.2	23.9	29.8	2.76	NCNC
36	55	M	66	7.7	33.3	79.3	18.3	23.1	2.35	NCNC
37	42	M	58	11.3	42.8	83.1	21.9	26.4	3.23	NCNC
38	36	M	62	12.2	44.5	100	27.4	27.4	3.2	MCNC
39	32	M	70	9.6	32.1	85.8	25.7	29.9	4.11	NCNC
40	28	M	60	11.8	37.6	83.9	26.3	31.4	2.69	NCNC
41	29	M	65	11.9	42.4	88.1	24.7	28.1	3.67	NCNC
42	26	M	55	11	36.9	83.9	25	29.8	2.76	NCNC
43	27	M	60	12.3	38.6	81.1	25.8	31.8	2.61	NCNC
44	34	M	70	12	43	85	23.7	27.9	2.75	NCNC
45	24	M	58	12	42.7	79.5	22.3	28.1	3.18	NCNC
46	35	M	65	12	42.6	86.9	24.5	28.2	3.82	NCNC
47	36	M	60	11.3	38.5	92.3	27.1	29.4	3.35	NCNC

48	21	m	76	11.3	36	96	30.2	31.4	2.69	NCNC
49	22	M	80	10.7	34.8	92.6	28.5	30.7	2.69	NCNC
50	19	M	56	11.8	36.7	112.9	36.3	32.2	2.75	MCNC
51	29	M	68	11	36.7	80.1	24	30	1.82	NCNC
52	55	M	81	11.6	37.5	99.5	30.8	30.9	1.68	NCNC
53	19	M	67	10.1	34.2	85.1	25.1	29.5	2.22	NCNC
54	19	F	53	11.2	35.5	86	27.1	31.5	2.55	NCNC
55	19	F	53	10.5	32.7	89.8	28.8	32.1	1.86	NCNC
56	19	M	50	7.5	25.3	74.9	22.2	29.6	3.13	MCHC
57	19	M	50	8.5	28.1	91.2	27.6	30.2	2.78	NCNC
58	19	M	70	11.7	38.9	93.5	28.1	30.1	1.4	NCNC
59	37	M	80	10.7	36.3	77.4	22.8	29.5	2.28	MCHC
60	33	F	60	9.4	29.5	83.8	26.7	31.9	1.5	NCNC
61	30	M	71	8	28.8	65.3	18.1	27.8	2.99	MCHC
62	29	F	59	11.6	34.9	100.6	33.4	33.2	2.65	NCNC
63	22	F	59	9.2	29.8	84.4	26.1	30.9	2.67	NCNC
64	24	M	58	8.9	29.7	86.1	25.8	30	2.86	NCNC
65	24	M	62	10.9	33.2	90.7	29.8	32.8	2.63	NCNC
66	23	M	53	12.1	37.5	94.7	30.6	32.3	2.86	NCNC
67	20	M	68	12	36.2	88.9	29.5	33.1	2.5	NCNC
68	19	F	57	10.6	33.8	81.8	25.7	31.4	2.89	NCNC
69	19	F	50	7.4	24.9	78.8	23.4	29.7	2.2	MCHC
70	25	F	62	10.8	32.7	86.7	28.6	33	2.24	NCNC
71	20	M	75	12.2	35.5	92.2	31.7	34.4	2.72	NCNC
71	20	M	85	10.5	32.2	91	29.7	32.6	3.44	NCNC
73	22	M	65	9.9	32.5	75.2	22.9	30.5	4.37	MCHC
74	22	M	52	12.3	40.2	58.9	18	30.6	2.57	MCHC

75	33	M	60	10.5	33.9	85.4	26.4	31	2.08	NCNC
76	56	F	75	12	39	91.3	28.1	30.8	2.27	NCNC
77	21	M	82	12.2	37.3	86.5	28.3	32.7	2.49	NCNC
78	19	F	53	12.1	44.1	76.3	20.9	27.4	2.51	MCHC
79	18	M	52	12	40.2	110.1	32.9	29.9	2.58	MCNC
80	20	F	52	12.3	39.3	88.3	27.6	31.3	2.78	NCNC
81	28	M	64	12	37.1	86.1	27.8	32.3	2.84	NCNC
82	33	M	85	12.4	39.6	82.8	25.9	31.3	2.82	NCNC
83	38	M	75	7.4	25.5	67.8	19.7	29	6.52	MCHC
84	44	M	91	9.6	30.5	78.6	24.7	31.5	2.69	MCHC
85	26	M	60	12	35.2	90.7	30.9	34.1	1.5	NCNC
86	25	M	65	11.5	37.4	80.8	24.8	30.7	2.43	NCNC
87	48	M	79	11.6	32.6	98.8	35.2	35.6	1.79	NCNC
88	25	M	55	12.3	36	88	30.1	34.2	2.81	NCNC
89	22	M	62	10.2	31.9	102.9	32.9	32	2.38	MCNC
90	20	M	70	11	33.4	93.6	30.8	32.9	2.23	NCNC
91	25	M	70	12.1	38.1	81.1	25.7	31.8	3.11	NCNC
92	25	M	70	11.1	36	83.7	25.8	30.8	4.48	NCNC
93	43	M	72	11.3	35.3	86.5	27.7	32	3.55	NCNC
94	48	M	79	9.6	30.9	84.9	26.4	31.1	3.4	NCNC
95	25	M	65	11.3	34.8	85.5	27.8	32.5	3.56	NCNC
96	24	F	55	10.4	30	99	34.3	34.7	1.54	NCNC
97	22	F	60	10.9	33	89.4	29.5	33	1.5	NCNC
98	23	F	52	9.3	29.7	66.4	20.8	31.3	3.25	MCHC
99	35	M	70	11.8	35.4	81.9	27.3	33.3	2.88	NCNC
100	59	M	65	10.7	30.3	89.1	31.5	35.3	1.43	NCNC
101	26	F	55	11	32.6	82.3	27.8	33.7	1.97	NCNC

102	24	F	50	8.1	28.3	61.3	17.5	28.6	2.97	MCHC
103	28	F	52	10.4	33.1	67.6	21.2	31.4	1.79	MCHC
104	58	F	55	10.1	31.1	86.6	28.1	32.5	2.43	NCNC
105	51	M	65	10.8	31.4	76	26.2	34.4	2.84	MCHC
106	50	M	85	11.2	35.5	91.7	28.9	31.5	1.34	NCNC
107	34	M	73	11.4	36.6	90.1	28.1	31.1	2.85	NCNC
108	18	M	70	11.7	35.5	90.1	19.7	33	2.77	NCNC
109	29	M	46	11.5	35.2	82.4	26.9	32.7	2.41	NCNC
110	33	F	57	11.7	35.5	91.5	30.2	33	1.4	NCNC
111	30	M	66	11.1	30.6	101.7	36.9	36.3	2.04	MCNC
112	34	M	85	11.5	35.3	87.2	28.4	32.6	2.15	NCNC
113	50	M	55	11.9	36.7	90.8	29.5	32.4	2.26	NCNC
114	26	M	58	11.70%	36.8	79	29.1	31.1	2.21	NCNC
115	21	F	50	10.40%	32.1	74.3	27.9	31.2	1.76	MCHC
116	23	M	60	7.70%	24	62.1	28.4	29.1	2.61	MCHC
117	21	M	59	11.50%	34.6	76.9	30.4	32.1	2.2	MCHC
118	23	M	55	11.10%	33.1	80.4	27.1	32.1	1.9	NCNC
119	43	M	60	9.10%	30.4	79.4	28.7	32.1	2.61	NCNC
120	21	M	54	9.90%	32.9	81.4	27.7	31.7	1.5	NCNC
121	19	F	48	7.40%	23.9	61.1	18.4	22.2	2.43	MCHC
122	43	M	63	11.60%	38	78.9	29.1	32.7	1.97	NCNC
123	52	M	60	11.80%	38.6	73.4	32.1	34.3	3.76	MCHC
124	25	M	57	10.70%	34.2	78.2	28.6	32.4	2.67	NCNC
125	38	M	61	10.90%	35.1	89.4	30	33.6	3.75	NCNC
126	27	M	59	9.70%	30.1	75.7	29.3	32.7	1.65	MCHC

127	21	M	52	9.90%	29.9	74.5	29.9	33.9	3.11	MCHC
128	33	M	58	10.10%	31.2	79.5	23.9	30.1	2.51	NCNC
129	20	M	51	6.80%	20.4	69.4	29.6	30.2	2.54	MCHC
130	20	M	55	8.80%	27.1	61.9	24.3	28.9	1.56	MCHC

ANNEXURE II

PROFORMA FOR STUDY :

Demographic Details:

1. Name:

2. Age :

3. Sex: M/F

4. OPD / IPD no.:

5. Present history:

6. Past history :

7. History of intake of drugs:

8. General physical examination**:**

Pallor :

Icterus :

Built :

Nourishment :

9. Vitals:-

PR :

BP :

RR

Temp :

Weight :

10. Investigations:

11. Hematological examination

Hb :

MCV :

MCH :

MCHC :

Peripheral blood smear:

ANNEXURE III

PROCEDURE :- Leishman's Stain

REAGENTS:-

Leishman powder - 0.15gm

Methyl Alocohol (acetone free) – 100ml.

Leishman powder :- Equal quantities of polychromed Methylene Blue (1 %) and Eosin B (0.1%)

PROCEDURE :-

1) Air dried film was flooded with leishman's stain for 2 minutes.
2) Double the volume of buffer water was added for 10 minutes.
3) The stain was washed off with distilled water until it acquired a pinkish tinge.
4) After drying, the back of the slide was wiped clean.
5) The slide was mounted with D.P.X

ANNEXURE IV

Parameters		Reference values
Hemoglobin	Males	13-17.2
	females	12-15.1
Hematocrit	Males	40.7-50.3
	Females	36.1-44.3
MCV		80-97.6
MCH		27.6-33.7
MCHC		32.7-33.5

ANNEXURE V

BLDE University's

SHRI B.M.PATIL MEDICAL COLLEGE HOSPITAL &

RESEARCH CENTRE, BLOOD BANK, BIJAPUR-586 103

CONSENT FORM

It has been explained to me in the language I understand and that :

1. Since blood can not be sterilised, there is possibility of transmitting any agent present in the red cells or plasma, other than transfusion - transmissible infections including HIV, Hepatitis B, HCV, Syphilis and Malaria, which are detected by routine screening tests as per drugs and cosmetic rules.

2. Blood is aproduct obtained from voluntary blood donors and may carry virusses of infective hepatitis and HIV. It has been further explained that necessary tests have been done to eliminate these. It has been further explained that these viruses can be transmitted by transfussion in window period, inspite of the screening procedures done to avoid these , as per drugs and cosmetic act.

3. Though extreme care is taken in every case of blood grouping cross matching and atypical antibody identification, transfusion reactions may occure.

Declaration by Patient

I Shri /Smt _____age _____ yrs sex _____ date _____ of ward _____ with IP No. _____ have been explained that regarding benefits and risks of blood transfusion.

I am prepared to take the risk of those transfusion hazards as a life saving measures in the view of emergency.

Signature of Patient **Attested by Unit Head**

ANNEXURE VI

ETHICAL CLEARANCE

Institutional Ethical Committee
BIJAPUR-586 103.
OUTWARD
No. 046/12
Date 18/10/12
BLDE U's Shri B.M. Patil Medical College

B.L.D.E. UNIVERSITY'S
SHRI.B.M.PATIL MEDICAL COLLEGE, BIJAPUR-586 103
INSTITUTIONAL ETHICAL COMMITTEE

INSTITUTIONAL ETHICAL CLEARANCE CERTIFICATE

The Ethical Committee of this college met on 18-10-2012 *at* 3-30pm *to scrutinize the Synopsis of Postgraduate Students of this college from Ethical Clearance point of view. After scrutiny the following original/corrected & revised version synopsis of the Thesis has been accorded Ethical Clearance.*

Title Donor deferral due to anemia and its Morphological pattern analysis in Bijapur - A prospective study

Name of P.G. student Dr. Mamata Kalyane
Pathology

Name of Guide/Co-investigator Dr B.R. Yelikar
Prof & HOD, Pathology

DR.TEJASWINI. VALLABHA
CHAIRMAN
INSTITUTIONAL ETHICAL COMMITTEE
BLDEU'S, SHRI.B.M.PATIL
MEDICAL COLLEGE, BIJAPUR.

Following documents were placed before E.C. for Scrutinization
1) Copy of Synopsis/Research project.
2) Copy of informed consent form
3) Any other relevant documents.