



**NAMIBIA UNIVERSITY
OF SCIENCE AND TECHNOLOGY**

**AN INVESTIGATION INTO THE AETIOLOGY OF ANAEMIA IN PREGNANT
WOMEN IN NAMIBIA**

By

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Thesis submitted in fulfilment of the requirement for the award of a degree in Master of
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February 2022

DECLARATION

I, Emma Ndakundana Shaduka hereby declare that the work contained in the thesis entitled “An investigation into the aetiology of anaemia in pregnant women in Namibia”, is my own original work and that I have not previously in its entirety or in part submitted it at any university or other higher education institution for the award of a degree.

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DEDICATION

I humbly dedicate this thesis to my beloved mother Fatima Shilongo and my loving grandmother Frodina Hitilasha. Thank you for always believing in education and for ensuring that it always ranks as a priority in our upbringing. Today, I am where I am because of education.

ABSTRACT

BACKGROUND: Anaemia is a condition in which the number of circulating red blood cells (RBCs) and/or haemoglobin (HB) is reduced for the person's age, gender and geographical specifications, which consequently affects tissue oxygenation. The common causes of anaemia are nutritional deficiencies (iron, vitamin B12 and folate), infectious diseases (malaria, Human Immunodeficiency Virus (HIV) and helminths infestation), chronic blood loss and closely spaced pregnancies. Anaemia in pregnancy impairs the health and wellbeing of women, and it is the major cause of morbidity and mortality among pregnant women and elevates the risk of perinatal and neonatal mortality. This study therefore sought to determine the etiology of anaemia in pregnant women and unravel socio-demographic factors associated with its development

METHODOLOGY: A cross-sectional analytical study, was conducted on 336 pregnant women from 4 randomly selected regions of Namibia (Karas, Khomas, Omaheke and Oshikoto). A semi-structured questionnaire was used to obtain quantitative data from 84 pregnant women who participated from the Khomas region. Blood specimens were collected by registered nurses and transported to the laboratory at 2-8⁰C for analysis. The analytical laboratory tests measured in this study were HB, MCV, Ferritin and Vitamin B12. The statistical analysis was performed using the Pearson's chi square test, and a p value of <0.05 was considered statistically significant.

RESULTS: The prevalence of anaemia during pregnancy was 20.8%, with 0% classified severe anaemia, 4.8% moderate anaemia and 16.0% mild anaemia. The prevalence rate per region was as follows: Karas (23.8%), Khomas (21.4%), Oshikoto region (20.2%), and

Omaheke (17.9%). The prevalence of iron deficiency anaemia (IDA) and vitamin B12 deficiency anaemia was 11.9% and 5.6%, respectively. Morphological classification of anaemia revealed that the most prevalent type of anaemia was normocytic normochromic anaemia (86.3%) in Namibia. Karas region (2.4%) reported the least prevalence of IDA, and Omaheke region (33.3%) reported the highest prevalence rate of vitamin B12 deficiency anaemia. Educational status (p -value= 0.003) and gestational status (p -value= 0.0031) were the only factors significantly associated with anaemia.

CONCLUSION: The prevalence of anaemia among pregnant women in Namibia was 20.8%, which is a moderate public health problem. This means that the government still needs to strengthen measures aimed at reducing anaemia in pregnancy. The study revealed that gestational age and educational level were significantly associated with anaemia. Hence, pregnant women should be encouraged to access antenatal care earlier in pregnancy to enable earlier detection and correction of anaemia, and in order to minimize fetal and maternal complications associated with anaemia in pregnancy.

Key Words: Anaemia, haemoglobin, prevalence, pregnancy, iron deficiency anaemia, vitamin B12 deficiency anaemia

TABLE OF CONTENTS

DECLARATION	2
ACKNOWLEDGMENTS.....	4
DEDICATION	5
ABSTRACT	6
CHAPTER 1: INTRODUCTION AND LITERATURE REVIEW	15
1.1 INTRODUCTION	15
1.1.1 Statement of the problem	20
1.1.2 Objectives of the study	21
1.1.2.1 The specific objectives of the study	21
1.1.2.2 Research questions.....	21
1.2 LITERATURE REVIEW	22
1.2.1 Definition of anaemia	22
1.2.2 Prevalence of anaemia during pregnancy.....	23
1.2.3 Factors associated with anaemia during pregnancy	25
1.2.3.1 Nutritional causes of anaemia	26
1.2.3.2 Infectious and parasitic causes of anaemia	32
1.2.3.3 Socio-demographic and socio-economic causes of anaemia	37
1.2.3.4 Other causes of anaemia.....	43
1.2.4 Physiologic anaemia of pregnancy	44
1.2.5 Consequences and burden of anaemia in pregnancy	45
1.2.6 Diagnosis of anaemia.....	47

1.2.6.1 Clinical diagnosis.....	47
1.2.6.2 Laboratory diagnosis.....	48
1.2.7 Prevention and treatment of anaemia	54
CHAPTER 2: METHODOLOGY	60
2.1 Study design and approach	60
2.2 Study area	60
2.3 Population of the study data	62
2.4 Inclusion and exclusion criteria.....	63
2.5 Sample size and sampling method.....	63
2.6 Data collection and procedure.....	64
2.6.1 Data collection method	64
2.6.2 Analytical laboratory methods	65
2.6.3 Data collection tool and procedure	66
2.6.4 Measures to ensure Validity and Reliability	66
2.6.5 Data analysis	67
2.6.7 Ethical Considerations.....	67
CHAPTER 3: RESULTS	68
CHAPTER 4: DISCUSSION AND CONCLUSION.....	79
4.1 Factors associated with anaemia in pregnancy	85
4.2 Conclusion	87
4.3 Limitations and Recommendations	87

5. References	89
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LIST OF FIGURES

Figure 1.1: Algorithm on the interpretation of pathology.....	49
Figure 1.2: Algorithm for the diagnosis of iron deficiency anemia	51
Figure 2.1: Geographic map of the regions of Namibia.....	62
Figure 3.1: Contribution of IDA, non-anaemia iron deficiency and non-IDA.....	73

LIST OF TABLES

Table 1.1: Red blood cell morphological characteristics.....	49
Table 3.1: Frequency distribution of anaemia status among pregnant women.....	68
Table 3.2: Anaemia status per region and within the anaemia status.....	69
Table 3.3: Frequency distribution of anaemia status among pregnant women.....	71
Table 3.4: Distribution of IDA per region.....	72
Table 3.5: Status of vitamin B12 deficiency among anaemic and non-anaemic pregnant women in Namibia.....	74
Table 3.6: Morphological anaemia in pregnant women based on HB and MCV values...	75
Table 3.7: Factors associated with anaemia in pregnancy.....	77

APPENDICES

APPENDIX A: Ethical clearance by the faculty of Health and Applied sciences.....	107
APPENDIX B: Ethical clearance by Ministry of Health and Social Services.....	110
APPENDIX C: Approval by the Namibia Institute of Pathology.....	112
APPENDIX D: Data collection tool.....	113
APPENDIX E: Consent form.....	115

LIST OF SYMBOLS

pmol/L	Picomoles per litre
pg/mL	Picograms per millilitre
%	Percentage
<	Less than
>	Greater than
$\geq$	Greater than or equal to
$\leq$	Less than or equal to
=	Equal to
g/dL	Grams per decilitre
μg	Micro gram
μL	Micro litre
$\mu\text{g/L}$	Microgram per litre
$^{\circ}\text{C}$	Degree Celsius
Pg	Picogram
ng/mL	Nanograms per milliliter

LIST OF ABBREVIATIONS

ACD	Anaemia of Chronic disease
AIHA	Autoimmune Haemolytic Anaemia
ANC	Antenatal care
CI	Confidence Interval
CRP	C-reactive protein
DNA	Deoxyribonucleic acid
FBC	Full blood count
HB	Haemoglobin
HCT	Haematocrit
HIV	Human Immunodeficiency Virus
ID	Iron deficiency
IDA	Iron deficiency anaemia
IQC	Internal Quality Control
LDH	Lactate Dehydrogenase
MCH	Mean Cell Haemoglobin
MCHC	Mean Cell Haemoglobin Concentration
MCV	Mean Corpuscular Volume
MoHSS	Ministry of Health and Social Services
NUST	Namibia University of Science and Technology

RBC	Red Blood Cell
RDW	Red cell Distribution Width
TIBC	Total Iron Binding Capacity
WHO	World Health Organization

CHAPTER 1: INTRODUCTION AND LITERATURE REVIEW

1.1 INTRODUCTION

Anaemia is defined as a condition in which the number of circulating RBCs and/or HB is reduced for the person's age, gender and geographical specifications, which consequently affects tissue oxygenation (Mohamoud, 2020; Omote et al., 2020). In pregnancy, it describes the condition whereby there is a reduction in the concentration of haemoglobin in the blood of a pregnant woman to a level of less than 11 g/dL. Haemoglobin is a large molecule in RBCs that is composed of heme, the iron compound and globin, a simple protein whose main function is to attract oxygen to the iron it carries (Cox, 2016). The World Health Organization (WHO) classifies anaemia in pregnancy as mild when the haemoglobin levels is (9-10.9 g/dL), moderate when haemoglobin levels are between (7.0-8.9 g/dL) and as severe anaemia when haemoglobin levels are less than 7.0 g/dL (Okia et al., 2019).

Anemia in pregnancy is an indicator of both poor nutrition and poor health (Zekarias et al., 2017), and it can result from a variety of causes. However, the common causes are; nutritional deficiencies (iron, vitamin B12 and folate), infectious diseases (malaria, Human Immunodeficiency Virus (HIV) and helminths infestation), chronic blood loss and closely spaced pregnancies (Daru et al., 2018; Obai et al., 2016). Excessive blood loss during women menstrual periods could also result in anaemia. Other factors leading up to anaemia in pregnancy are; unemployment, poor nutritional intake, young age and multigravida (Okubatsion, 2015; Sholeye et al., 2017). Thus, understanding the etiology of anaemia is essential for the implementation of effective interventions needed to address the specific causes of anaemia as well for monitoring and evaluating measures already in place that are targeted to control it.

The signs and symptoms of anaemia are often non-specific and they include; pallor, headache, palpitations, tachycardia, breathlessness and dyspnea (Gudeta et al., 2019a). Fatigue, tiredness, skin paleness, difficulty in breathing and tachycardia are also some of the signs and symptoms experienced by anaemic pregnant women (Achebe & Gafter-Gvili, 2017). Furthermore, anaemia in pregnancy can result in poor performance of the mothers on their day-to-day activities, as they are likely to be weak and get exhausted fast. Depending on the type, duration and severity of anaemia at the time of pregnancy, it could result in various adverse consequences towards the infant such as low birth weight and preterm delivery (Satyam & Khushbu, 2015).

Iron deficiency anaemia which is the main cause of anaemia in pregnancy is often due poor absorption of iron from diet and from poor iron intake (Lumor et al., 2019). Deficiency of iron in pregnancy is reported to result in impaired motor and cognitive development for children, and in increased risk of developing sepsis soon after birth (Okia et al., 2019; Satyam & Khushbu, 2015). Another common cause of anaemia is vitamin B12 deficiency, which mostly results from poor intake of food or supplements rich in vitamin B12. Vitamin B12 is needed for deoxyribonucleic acid (DNA) synthesis and nuclear maturation, and its deficiency in pregnancy is linked to increased risk of neural tube defects, recurrent spontaneous abortions, preterm birth and other neurological symptoms in infants (Achebe & Gafter-Gvili, 2017). Folic acid deficiency is also one of the main causes of anemia in pregnancy. It is an essential vitamin necessary for DNA synthesis and for physiological development of neural tube, and its deficiency can lead to megaloblastic anaemia (Di Renzo et al., 2015). Hence, to overcome these deficiencies (iron, vitamin B12 and folate) in pregnancy it is recommended that pregnant women be advised to eat food rich in iron, vitamin B12 and folate (Assefa et al., 2019). It is also advisable that pregnant women with multiple risk factors such as nutrient deficiency, parasitic and HIV infections, and/or chronic

blood loss that they take iron, folic and vitamin B12 supplements throughout their pregnancy to minimize complications associated with anaemia (Cox, 2016).

Although anaemia affects all age groups, it has been reported to be more prevalent in pregnant women (Obai et al., 2016). This is due to the increased demand and physiological change in pregnancy. During pregnancy there is a physiological increase of plasma volume which surpasses the production rate of RBCs and HB, resulting in haemodilution which subsequently leads to the decline of HB and eventually to the development of anaemia (Pavord et al., 2020). The physiologic anaemia of pregnancy denotes an expansion of plasma volume of 50% and an increase in the RBC mass of 25% (Achebe & Gaftor-Gvili, 2017).

Globally, anaemia affects 1.62 billion people of which 56 million have been identified as pregnant women (Mahamoud et al., 2020). The prevalence of anemia in pregnancy is reported to be 24.1% in the Americas, 48.2% in South East Asia, 25.1% in Europe, 44.2% in East Mediterranean, 30.7% in West Pacific and 52.5% in South East Asia (Okubatsion, 2015; Gudeta et al., 2019b; Zekarias et al., 2017). In Africa 57% of the pregnant women are estimated to be anaemic (Okubatsion, 2015). The prevalence of anaemia during pregnancy is as follows in some selected African countries: Ethiopia (62.7%), Sudan (66.2%), Southeastern Nigeria (40.4%), and 25.8% in Kampala, Uganda, Ghana (56%) (Zekarias et al., 2017; Mohamoud, 2020; Tibambuya et al., 2019). The varying prevalence rates reported from African countries is likely due to different sizes of study populations used in the studies, geographical areas (rural or urban), rates of parasitic infections and socio-economic statuses of participants (Tunkyi & Moodley, 2016). In Sub-Sahara Africa, 55.8% of pregnant women are thought to be anaemic (Ajepe et al., 2020). In three selected southern African countries, the prevalence of childhood as well of maternal anaemia has been reported as; Malawi (63.8%), Mozambique (70%) and Zimbabwe (58.6%) for childhood anaemia, with the prevalence of maternal anaemia reported as 26.7%, 53.4% and 25.1% in Malawi,

Mozambique and Zimbabwe respectively (Ntenda et al., 2018). In Namibia, recent statistics indicate the prevalence of anaemia among pregnant women as 27.1% (World Bank, 2020).

The higher prevalence of anaemia in sub-Saharan Africa in pregnancy has been associated with low intake of iron among pregnant women (Okia et al., 2019; Adam et al., 2018). High burden of HIV infection could also be a contributing factor to the increased prevalence rate of anaemia in pregnancy in sub-Saharan Africa. Malaria infection is endemic in sub-Saharan Africa, and this could be another contributing factor to higher prevalence rate of anaemia in pregnancy (Simo et al., 2020). In southern African countries, the higher prevalence rate could be linked to repeated infections, short intervals between pregnancies, long distances to health care facilities and poor health seeking behaviors among pregnant women (Gudeta et al., 2019a). In Namibia, poverty is more likely to be the leading cause of anaemia in pregnancy. This is because Namibia is a dry country, with the majority of its population depending on seasonal farming for food (Shimanda et al., 2020). Thus, during the non-farming seasons pregnant women who cannot afford to purchase nutritional food are prone to developing nutritional deficiencies such as iron deficiency which is the leading cause of anaemia in pregnancy (Al-Khaffaf et al., 2020).

Moreover, significant efforts have been made into tackling maternal and child health resulting from the effects of anaemia worldwide, and most specifically in African countries where the burden and prevalence of maternal and infant mortality and morbidity continues to be highest. Antenatal care services is one of the measures that can be implemented by countries in order to combat and address the effects of maternal health and to allow for correction of maternal conditions such as anaemia that might exist even before pregnancy (Anlaaku & Anto, 2017).

In Namibia, ANC is offered to pregnant women without charging a fee at all state hospitals, healthcare centers and clinics. It is used as a tool for prevention, monitoring, early detection, and treatment of general medical and pregnancy related complications including anaemia. Pregnant women who commence with early ANC and regular follow up have plentiful opportunities to obtain delivering health information and interventions which could significantly improve their health and the health of their fetus (Satyam & Khushbu, 2015). During ANC visits, pregnant women can know their status with regards to anaemia and if found anaemic they can be treated and/or advised on the approach to take in order to reverse the anaemia. They can also be advised on the type and frequency of foods to consume that are rich in iron, vitamin B12, or other minerals and vitamins, as well be supplemented with nutritional elements that they can take to address their deficiency.

Early diagnosis and treatment of anaemia can prevent more severe forms of anaemia and its associated risks and complications (Anlaakuu & Anto, 2017). Hence, the need to continuously monitor pregnant women for the likelihood of developing anaemia and treating as well reversing the effects before it progresses to the severe form of anaemia. Furthermore, to date, local statistics on the prevalence, magnitude and the associated risk factors of anaemia in pregnancy have been crucial in the management, prevention and control of anaemia in pregnancy in Namibia. This will subsequently contribute to the reduction in overall maternal and infant morbidity and mortality in the country, southern Africa, sub-Saharan Africa and world at large.

1.1.1 Statement of the problem

Anaemia in pregnancy is a major public health issue affecting both developed and developing countries resulting in high maternal morbidity and mortality (Stephen et al., 2018). There are many causes of anaemia during pregnancy and they can result from a single or combination of factors such as; nutrient deficiency, parasitic and HIV infections, and chronic blood loss (Daru et al., 2018; Obai et al., 2016). The common anaemia types encountered by pregnant women are nutritional anaemias which are mostly caused by deficiencies in iron, folate and vitamin B12 (Serbesa & Iffa, 2018; Ajepe et al., 2020). Iron deficiency is the leading cause of anaemia during pregnancy and has been reported to account for an approximate 75% of all reported cases of anaemia in pregnancy (Al-Khaffaf et al., 2020). Development of anaemia in pregnancy may vary due to dietary practices and socioeconomic background. However, anaemia is largely preventable and completely treatable, if detected on time. A study done in four southern African countries Malawi, Mozambique, Namibia and Zimbabwe on evaluating maternal anemia as a potential risk factor for anemia in children aged 6–59 months in Southern Africa, found that maternal and infant mortality and morbidity are a major cause for concern in Namibia and identified anaemia in pregnancy as a contributing factor (Ntenda et al., 2018). Despite the numerous efforts made to reduce the burden of anaemia in pregnancy in developing countries (Zehra et al., 2014), its etiology has not been studied comprehensively in Namibia and it seems there is limited data on it. This means numerous anemia cases which are potentially preventable are not diagnosed or are poorly managed. This increases the cost of health care, poor treatment outcomes and ultimately high maternal and infant mortality and morbidity. This study therefore sought to determine the etiology of anaemia in pregnant women and unravel socio-demographic factors associated with its development. The findings of the study would provide the necessary information for the interventions aimed at reducing maternal-infant morbidity and

mortality resulting from anaemia. Its findings may also be used as a baseline data for other researchers who might be interested in this study area.

1.1.2 Objectives of the study

The main objective of the study was to determine the aetiology of anaemia in pregnant women in four randomly selected regions of Namibia.

1.1.2.1 The specific objectives of the study

- To determine the prevalence of anaemia in pregnant women in Karas, Khomas, Omaheke and Oshikoto regions.
- To classify the types of anaemia in pregnant women in Namibia.
- To establish the socio-demographic factors associated with development of anaemia during pregnancy in Khomas region.

1.1.2.2 Research Questions

The study sought to answer the following questions:

- What is the prevalence of anemia in pregnant women attending ANC in the 4 randomly selected regions (Karas, Khomas, Omaheke, Oshikoto) of Namibia?
- Which anaemias are common during pregnancy?
- Which socio-demographic factors are associated with anaemia in pregnancy in Khomas region?

1.2 LITERATURE REVIEW

1.2.1 Definition of anaemia

The WHO defined anaemia in pregnancy as having an HB level below 11g/dL and further subdivide it into 3 levels of severity; mild (HB levels 9-10.9 g/dL), moderate (HB levels 7-8.9 g/dL) and severe anaemia (HB levels less than 7 g/dL) (Okia et al., 2019). While the Centers for Disease Control and prevention (CDC) defines anaemia in pregnancy as having a haematocrit (HCT) of <33% during the first and third trimesters, and HCT of < 32% in the second trimester (Achebe & Gafer-Gvili, 2017). Anaemia develops through three main mechanisms; ineffective erythropoiesis (production of insufficient RBCs), haemolysis (destruction of RBCs) and blood loss (World Health Organization [WHO], 2017).

Haemoglobin is a critical component for tissue oxygenation, cellular function and cell development, and its level indicates the amount of circulating HB proteins which are attached to RBCs and constitutes of the body's usable form of iron (Cox, 2016). Among other things the body requires iron, vitamin B12 and folic for erythropoiesis, hence if there is a lack of one or more of these ingredients anaemia may develop (Satyam & Khushbu, 2015). Although anaemia is commonly diagnosed by low HB levels and low haematocrit (HCT), it can also be diagnosed using RBC, mean corpuscular volume (MCV), reticulocyte count and by assessing a blood film made from a full blood count. At population and clinical practice, HB level is the main indicator used to define anaemia (Chaparro & Suchdev, 2019).

In the clinical setting anaemia is mostly diagnosed through measurements of the HB concentration in the blood. However, defining it using the HB concentration require the investigator to know and understand that HB values naturally differ according to age, gender, pregnancy status, genetic and environmental factors, and potentially race. Signs and symptoms can also be used as leading guides in the identification and diagnosis of anaemia.

Additionally, medical history can be used to define anaemia in case of the unavailability of haemological information, but they are limited in their abilities to detect and distinguish the possible anaemia (Helmy et al., 2018).

At the time of pregnancy there is a physiologic expansion of plasma volume of about 50% and an elevation of RBCs to about 30%, and this disproportional elevation in plasma and RBCs decreases the HB concentration of the pregnant woman (Gebreweld & Tsegaye, 2018). Depending on the level of HB concentration before pregnancy, this haemodilution could result in the pregnant women being anaemic (Sabina et al., 2015).

Anaemia has been identified as an indicator of both poor nutrition and poor health, and maternal mortality is a key health indicator in any society (Zekarias et al., 2017). Hence, health services in any country should target to minimize the incidences of anaemia in order to improve the health status of that society.

1.2.2 Prevalence of anaemia during pregnancy

Anaemia in pregnancy has been studied by many researchers worldwide with varying findings. It has a varied prevalence, etiology and degree of severity in different populations, regions, countries and continents as a result of differences in socioeconomic conditions, cultural, lifestyles, health seeking behaviours and geographical variations (Satyam & Khushbu, 2015; Helmy et al., 2018; Omote et al., 2020).

Approximately, 800 women die daily as a result of complications in pregnancy and childbirth and anaemia accounts for 20% of all maternal deaths (Okubatsion, 2015). While on an annual basis more than 11500 maternal deaths and 59100 prenatal deaths in the world are attributed to anaemia in pregnancy and of these 50% of the pregnant women are from the developing

countries (Mohamoud, 2020; Gebreweld & Tsegaye, 2018; Zekarias et al., 2017). The pregnant women from developing countries are vulnerable to anaemia due to low socio-economic conditions, inadequate nutritional intake of food rich in proper nutrients, poor perinatal vitamins intake, low iron and folic acid intake, repeated infections, frequent pregnancies and poor health seeking behaviors (Gudeta et al., 2019a).

The burden of anaemia in pregnancy is more pervasive in Asia and Africa, where majority of the anaemic pregnant women are estimated to be (Ntenda et al., 2018). To ascertain this, one study done in Bathida city in India found the prevalence of anaemia among pregnant women to be 81.8% and of those, 0.2%, 91% and 8.8% were severely, moderately and mildly anaemic (Bansal et al., 2020). Another study conducted at the University of Sindh Jamshoro and Civil hospital in Lumhs, Jamshoro, China on anaemia and its association with parity found anaemia prevalence of 51.5% among pregnant women (Shah et al., 2020).

Many studies done in Africa found relatively high prevalence rates in various African countries with varying severities (Omote et al., 2020; Wemakor, 2019; Tibambuya et al., 2019; Shitie et al., 2018; Shimanda et al., 2020). In Namibia, the prevalence of anaemia among pregnant women has been reported to be 27% (World Bank, 2020). The prevalence of anaemia in pregnancy rises in first trimester, to second and third trimesters from 8%, to 12% and 34% respectively (Al-Khaffaf et al., 2020). A study by Di Renzo et al. (2015), found the prevalence of anaemia highest (24.3%) during the third trimester, followed by the second trimester (20.7%) and the lowest prevalence was in the first trimester (14.6%). The higher prevalence in the third and second trimesters could be as a result of the haemodilution in pregnancy, which increases during the second trimester and reaches its peak point in the third trimester (Gebreweld & Tsegaye, 2018).

The WHO classifies anaemia as a public health concern for a country based on its prevalence. If the prevalence of anaemia is less than 5% is considered as no public health concern, if it is between 5% and 19.9% is indicative of a mild health concern and if it is between 20% and 39.9% is considered a moderate health problem (Ramesh et al., 2017; Adam et al., 2018). However, if the prevalence is 40.0% or beyond, the WHO regards it as a severe public health concern (Okubatsion, 2015). Anaemia prevalence has also been reported to increase with gestational stage, whereby women who seek antenatal care late in their pregnancy usually at 3rd trimester were more likely to develop anaemia in pregnancy than those who seek it at an earlier gestational stage (Ahenkorah et al., 2016). This could be due to that during ANC women are provided with information on anaemia prevention, and given iron supplements to increase their iron stores (Anlaaku & Anto, 2017). Thus, if the pregnant women know about the negative consequences anaemia and the preventive measures earlier in their gestation, they are likely to prevent the predisposing factors which consequently reduces their risks of anaemia.

Furthermore, despite the fact that ANC services are made available in many countries and supplementations such as iron and folic acid are being provided and made available to pregnant women (Sabina et al., 2015). The prevalence of anaemia remained unacceptably high in many African countries including Namibia. Hence, the need to critically evaluate and understand factors associated with anaemia in pregnancy.

1.2.3 Factors associated with anaemia during pregnancy

Some women tend to be anaemic prior pregnancy and others get progressively anaemic during pregnancy (Anlaaku & Anto, 2017). Many factors predispose pregnant women to the development of anaemia. However, these predisposing factors are not universal in their

significance as they vary from one antenatal population to another, and within the same population not all of them might even be significant (Mehrotra et al., 2018). Thus, individual pregnant women may acquire anaemia from a combination of factors making it difficult to pinpoint a single factor as a sole cause of the anaemia.

The common factors reported to result in anemia in pregnancy are nutrient deficiency (i.e inhibitors of iron deficiency, dietary deficiency of iron, folic and vitamin B12), parasitic infestation (malaria, hookworm), HIV infection and chronic blood loss, and closely spaced pregnancies (Daru et al., 2018; Obai et al., 2016; Anjum et al., 2020). Other reported causes of anaemia in pregnancy are; loss of appetite and severe vomiting in pregnancy, and heavy menstrual flow prior pregnancy (Okubatsion, 2015). Anaemia in pregnancy can also result from genetically inherited haemoglobinopathies such as thalassaemia (Di Renzo et al., 2015). Other predisposing factors to anaemia in pregnancy are; low income status, low educational level, poor dietary intake, short pregnancy intervals and young age (Mockenhaupt et al., 2015). Drinking water from unsafe sources increases the risk of gastrointestinal (GI) infections which is highly linked to anaemia as a result of iron stores depletion and haemolysis of RBCs (Ntenda et al., 2018). The prevalence of anaemia is high in pregnant women who have relatively less and late ANC follow up (Shitie et al., 2018). Early and regular ANC visits are critical as they could enable the early correction and treatment of anaemia that might exist even prior the pregnancy (Anlaaku & Anto, 2017).

Below is brief information on how the common factors contribute to the development of anaemia.

1.2.3.1 Nutritional causes of anaemia:

Anaemias resulting from nutritional causes are highly preventable, and in most cases reversible. Iron, vitamin B12 and folic acid deficiencies are the main causes of nutritional anaemia in pregnant women (Mireku et al., 2017). Nutritional anaemias develop as a result of low dietary intake or malabsorption of the required micronutrients, vitamins and proteins, as well from lack of nutrient supplementation. They also result when there is a declined intake of certain nutrients necessary to meet the demands for synthesis of HB and erythropoiesis (WHO, 2017). Women in low and middle income countries have a higher chance of developing nutritional anaemia in pregnancy due to increased deficiency of dietary iron and other macronutrients (Daru et al., 2018).

Iron Deficiency Anaemia (IDA)

Iron is an essential component of HB, that's obtained through the consumption of iron rich food and by recycling iron from old RBCs (Daka et al., 2018). It is essential for all cells and its functions include; energy metabolism, gene regulation, cell growth and differentiation, oxygen binding and transport, muscle use and storage, enzyme reactions, neurotransmitter and protein synthesis (Satyam & Khushbu, 2015). Although iron is more essential through all stages of human development, it is more important during the early stages of life as it regulates brain development (Cerami, 2017). In the blood iron is commonly found in the HB within RBCs and erythroblasts, myoglobin in muscles, iron containing proteins (cytochromes and catalases) and in the storage proteins (ferritin and hemosiderin) (Cerami, 2017). While dietary iron can be acquired in foods such as red meats, sea foods, green leafy vegetables, beef liver and nuts (Cox, 2016).

During pregnancy there is progressive process towards iron depletion in the blood; it commences with the depletion of iron stores, followed by iron deficient erythropoiesis, then a decline in the concentration of haemoglobin which eventually leads to IDA (Ajepe et al., 2020; Chaparro & Suchdev, 2019). Hence, if iron supply does not meet pregnancy iron demand the iron stores get used up faster than they can replenish leading to iron depletion, which if left without any intervention may progress to iron deficiency. It is estimated that 75% of non-physiological anaemia during pregnancy are caused by iron deficiency (Al-Khaffaf et al., 2020). Therefore, pregnant women require more iron compared to non-pregnant women to cater for the formation and growth of the placenta, for the fulfillment of fetus iron requirement and for the increased red cell mass of the woman (Ajepe et al., 2020; Lumor et al., 2019). The iron demand picks up gradually during pregnancy through gestation from 0.8 mg/day to 705 mg/day in the third trimester (Satyam & Khushbu, 2015). The total amount of iron needed during pregnancy is about 900mg, of which 500-600mg required for the uterus and its contents, 150-200mg that is lost in the blood loss during delivery and 150-200 mg that is expended in lactation (Bansal et al., 2020).

Iron requirements for both the mother and fetus are minimal within the first trimester of pregnancy due to absence of menstruation. For this reason, pregnant women who are well nourished can have enough iron stores to sustain the iron demand and decrease their likelihood of developing anaemia in the first trimester. The decline in iron stores is notable during the late first trimester or towards the beginning of second trimester (Mireku et al., 2017). This is mainly due to increase fetal iron requirements to cater for the development and maintenance of the fetal placental unit (Lumor et al., 2019). On the other hand, majority of the women start their pregnancy with poor or depleted iron stores resulting in the amount of iron absorbed from diet together with their mobilized from stores being usually insufficient

to meet the maternal demands imposed by pregnancy and increasing the risks of developing iron deficiency early in pregnancy (Renzo et al., 2015).

Iron deficiency in pregnancy has been associated with adverse fetal and maternal outcomes (Ajepe et al, 2020). Pregnant women with iron deficiency are reported to have one or more of following signs and symptoms; fatigue, headache, decreased mental and work capacity, decreased maternal blood flow (Api et al., 2015). Because of effects of anaemia on work productivity of the pregnant women it has a negative economic impact in terms of her income as a result of decreased productivity (WHO, 2017).

Moreover, iron deficiency has been associated with restricted fetal growth, low birth weight, development of sepsis soon after birth, impaired cognitive and motor development in children, (Okia et al., 2019; Satyam & Khushbu, 2015). Iron deficiency can also interfere with defective myelination in infants, causing the infant to have permanent defects in mental development and performance which may have negative impacts towards child learning capacity (Renzo et al., 2015). Reduced myelination, altered temperament and altered dopamine metabolism are also some of the symptoms in infants with iron deficiency (Cerami, 2017). Due to iron effects on dopamine and other monoamines which are closely associated with behavior, an association between behavior outcome and iron status at 12 weeks or 6 months was identified (Moreno-Fernandez et al., 2019). Whereby iron deficiency in infancy was linked to persistency in aggressive and rule breaking behavior until 7 years of age in children.

Vitamin B12 Deficiency

Vitamin B12 also known as cobalamin is a water-soluble vitamin acquired via the ingestion of fish, meat and other daily products, as well through fortified cereals and synthesized by micro-organisms (Strand et al., 2020; Venkatramanan et al., 2016). Due to its primary dietary sources being animal products, individuals with low intake of animal products mainly vegans and vegetarians are at a higher risk of developing vitamin B12 deficiency. Vitamin B12 is necessary for neurologic function, RBC production, differentiation and development, cellular growth and for DNA synthesis (Langan & Goodbred, 2017; Sukumar et al., 2016).

During pregnancy there is a gradual reduction in the serum vitamin B12 intake (Helmy et al., 2018). This is to cater for the rapid expansion of fetal and maternal tissues as well as a result of normal physiological pregnancy changes such as; haemodilution, hormonal changes and maternal to fetus transfer of vitamin B12 (Visentin et al., 2016). Hence, when a pregnant woman has a minimal intake of food rich in vitamin B12 or does not take any vitamin B12 supplements her chances of developing vitamin B12 deficiency anaemia are elevated. Vitamin B12 deficiency can also result from pernicious anaemia, which is caused by an intrinsic factor blocking antibodies that interfere with vitamin B12 absorption (Govindappagari et al., 2019).

Pregnant women with vitamin B12 deficiency experience one or more of the following signs and symptoms; mental slowness, memory defects, hallucinations, numbness or tingling in extremities (Helmy et al., 2018). Thus, it is essential that healthcare providers make it a priority to enquire with pregnant as well lactating women about their diet and medical history in order to identify those at an increased risk of developing Vitamin B12 deficiency.

Vitamin B12 is crucial for DNA synthesis and nuclear maturation, thus its deficiency in pregnancy is associated with increased risk of neural tube defects, recurrent spontaneous abortions, preterm birth and it has been linked to neurological symptoms in infants (Achebe

& Gafter-Gvili, 2017). Its deficiency during pregnancy has also been linked to infant's failure to thrive and haematological disorders (Dayaldasani et al., 2014). Additionally, vitamin B12 deficiency has also been linked with infant small for gestation, growth retardation and children born to Vitamin B12 deficient mothers are at an increased risk for adverse health outcomes such as developmental abnormalities and anaemia (Chandyo et al., 2017).

Moreover, infants who are born by mothers with normal vitamin B12 stores are thought to have sufficient vitamin B12 stores to cover for all their metabolic needs, but those born by mothers with vitamin B12 deficiency are noted to have less stored vitamin B12 and can develop deficiency within the first year of life (Pacholok, 2014). Without intervention vitamin B12 deficiency in pregnancy and early childhood is prone to have lasting effects on infant growth and development, which may be irreversible (Venkatramanan et al., 2016). In one study maternal vitamin B12 supplementation use during pregnancy resulted in higher maternal vitamin B12 with positive birth outcomes (Visentin et al., 2016). While the intervention to vitamin B12 deficiency in early childhood was not found to yield any improvements in neuro development nor did to have any effect on growth or HB concentration (Strand et al., 2020). This might mean that, more research needs to be carried out focusing on the impact of vitamin B12 intervention in early childhood vs intervention in pregnancy.

Folic Acid Deficiency

Folate is essential water soluble vitamin, which acts as a coenzyme in many organic reactions (Ndiaye et al., 2018). Folate occurs naturally in food such as fruits, green leafy vegetables and liver, while folic acid is the synthesized form of folate found in fortified foods and supplements (Khan & Jialal, 2020). Folic acid is an important vitamin necessary for DNA

synthesis and for physiological development of neural tube, and its deficiency can lead to megaloblastic anaemia (Di Renzo et al., 2015). Folic acid plays an important role in the development of neural tube of the neonate, in the production of RBCs, and in the synthesis and repair of DNA and its supplementation in pregnancy has been shown to reduce risks of congenital health defects, cleft lips and limb defects (Okubatsion, 2015). Additionally, when folate levels are deficient there is a higher possibility of megaloblastic anaemia to occur (Alkhaldy et al., 2020).

Furthermore, folic acid deficiency can result from a variety of causes such as dietary intake, presence of diseases inhibiting folate absorption (celiac disease, gastric bypass and short bowel syndrome) and medications, overcooking of food also destroys folic acid (Khan & Jialal, 2020). Additionally, physiological changes at the time of pregnancy, accompanied by fetal growth and development elevates the demands for folic acid (Mireku et al., 2017). There is also an increase of folic acid demand during lactation as a result of transfer of folic acid to maternal milk (Ndiaye et al., 2018). This increased demand cannot be met by diet alone and if the diet is not supplemented by folic tablets, it increases the likelihood of the pregnant woman to develop folate deficiency anemia.

Deficiency in folic acid during pregnancy has been associated with negative pregnancy outcomes for the fetus such as; neural tube defects and spontaneous abortions (Mireku et al., 2017). Neural tube defects such as; spina bifida, anencephaly and encephalocele caused by neural tube closure during early embryogenesis are associated with perinatal mortality (Al-mashhadane et al., 2018). Additionally, preeclampsia, fetal malformation and preterm delivery are also some of the complications linked to folic acid deficiency during pregnancy (Assefa et al., 2019).

Folic acid supplementation preconception and during the first trimester has been found to have a significant impact on the reduction of neural tube defect in infants. A study by Al-mashhadane et al. (2018), found a 72% reduction in neural tube defects in infants born by women that had a previous child with neural tube defect who took folic acid supplementation daily and throughout first semester, compared to infants born by women with a previous child with neural tube defect who never took folate acid supplementation preconception and throughout pregnancy.

1.2.3.2 Infectious and parasitic causes of anaemia

Hookworm infection

Hookworm is the primary soil transmitted helminth linked to anaemia. It lives in the small intestines and is common in tropical regions due to a favorable climate in those regions and in poor countries as a result of socio-economic factors such as; poor sanitation, low quality of water for domestic use and poor personal hygiene including habits of gardening barefooted (Apili et al., 2020). The two species known to cause infections in humans are *Ancylostoma duodenale* and *Necator americanus* (Umbrello et al., 2021). Hookworm sticks to and feeds from the intestinal mucosa leading to gastrointestinal blood loss and depletion on the iron stores, and depending on the iron level of the individual as well the presence of other risk factors it can results in maternal anaemia (Chaparro & Suchdev, 2019; Mockenhaupt et al., 2015). It has been estimated that a single hookworm can lead to a blood loss of 0.03-0.26 mL/day (World Health Organization, 2017).

Hookworm infections in the mother is typically asymptomatic, the symptoms usually present based on the stage of parasite development and the site of the affected host, and the signs may include abdominal pain, diarrhea, tenderness and vomiting (Ghoderf & Jain, 2019).

Furthermore, complications from hookworm infection may not only end at the mother but can also affect her child. Hookworm infection is reported to aggravate anemia in pregnant women and may result in low pregnancy weight gain and intrauterine growth retardation which can be accompanied by low birth weight and higher perinatal mortality rate (Hailu et al., 2019). Hookworm infection in infancy is ought to have long term consequences for iron deficiency anaemia and protein malnutrition accompanied by growth stunting and decreased cognitive capacity (Umbrello et al., 2021).

A study by Shiferaw et al. (2017), found a 20% of pregnant women being infected by hookworm infection. This prevalence of hookworm infections during pregnancy could be attributed to the lower coverage of anthelmintic treatment in maternal health programme (Lebso et al., 2017). It could also be attributed to poor interventions to break transmission routes such as lack of proper sanitation, poor provision of safe water supply for domestic use and poor personal hygiene including not washing hands frequently and properly.

Malaria infection

Malaria is an infective disease resulting from infective parasites belonging to the genus *Plasmodium* (*P. falciparum*, *P. ovale*, *P. malariae*, and *P. vivax*). African regions are documented to have disproportionately high burden of malaria infection accounting for 90% of malaria cases and 92% of malaria deaths (Chaparro & Suchdev, 2019). Malaria infection is one of the causes of anaemia in pregnancy and the primary cause of severe anaemia (Achebe & Gaftor-Gvili, 2017). The etiology of malarial anaemia in malaria endemic regions include various overlapping features such as; increased destruction of infected and uninfected RBCs, splenic sequestration of RBCs, dyserythropoiesis and bone marrow

suppression, infectious diseases and chronic transmission of malaria (Kaur et al., 2017; Ntenda et al., 2018).

In malaria endemic areas the infection caused by *P. falciparum* is often asymptomatic leading to the malaria infection to remain undetected and untreated, however the parasite continues to be incubated in the placenta (M Afolabi & Olukosi, 2018). Despite the malaria parasite being undetected in the blood film, when the malaria infected RBCs pile up in the placenta they lead to adverse birth outcomes (Mlugu et al., 2020). The extent to which malarial infections manifests in the pregnant women and in the fetus varies according to the immunity level, and women in their first pregnancy (primigravidae) are ought be more susceptible to malarial infection than second gravidae or multigravidae (Chandrashekar et al., 2019). The higher susceptibility in primigravidae women is associated with lack of placental parasite-specific immunity (*Plasmodium falciparum* erythrocyte membrane protein 1) which is acquired through repeated pregnancies (Mlugu et al., 2020).

Malaria infection during pregnancy may have severe effects for the mother and her unborn child. It may result in deformities of the genital track to make conception impossible or if conception occurs it may prevent normal implantation and development of the placenta (Okafor et al., 2012). Malaria infection at the time of pregnancy may also cause general decline in the woman's immunity, pulmonary edema, acute respiratory distress syndrome and hypoglycemia (Kaur et al., 2017). The malaria parasites may cause fibrosis of the placenta which consequently prevent sufficient nutrients to be transported to the fetus leading to fetal distress, abortion and in rare cases congenital malformations (M Afolabi & Olukosi, 2018). Moreover, women infected with malaria during pregnancy can pass on the infection to their fetus and this can result in poor birth outcomes such as; intrauterine growth retardation, congenital anaemia, preterm birth, low birth weight and infant mortality (Achebe & Gafter-Gvili, 2017).

Furthermore, a study by Anlankuu and Anto (2017) identified malaria infection as one of the main factors associated with anaemia in pregnancy with those infected found to be 7.2 times more likely to be anaemic in comparison to the pregnant women who were not infected with malaria. This finding could be associated with the intricate link between malaria and anaemia such as; increased destruction of non-parasitized and parasitized RBCs, impaired erythropoiesis and delayed production of RBCs due to bone marrow suppression (Ntenda et al., 2018). Additionally, maternal mortality is reported to be twice in pregnant malaria women compared to non-pregnant women with severe malaria (Okafor et al., 2012). One study found malaria infection during pregnancy contributing 4.5%, 0.3% and 0.3% to the prevalence of mild, moderate and severe anaemia respectively (Fondjo et al., 2020), which is suggestive that the presence of other factors also play significant roles in anaemia during pregnancy.

Human Immunodeficiency Virus (HIV) infection

Human Immunodeficiency Virus is a retrovirus that causes a multi-systemic disease due to viral action on the cells that exhibit CD4 protein in their cytoplasmic membrane and causes hematological changes in the blood (Marchionatti & Parisi, 2020). There is a significant association between HIV and anaemia in pregnancy (Omote et al., 2020). Anaemia was a common clinical finding in pregnant women living with HIV and has been associated with advanced maternal and fetal outcomes (Nandlal et al., 2014). The etiology of anaemia in HIV pregnant women is multifactorial, and could be due to nutrient deficiencies (iron, vitamin B12 or folate deficiencies), anaemia of inflammation from HIV or from opportunistic infections (Finkelstein et al., 2020). Additionally, the virus is ought to have direct effect on

anaemia by affecting the hematopoietic progenitor cells and reduces responsiveness to erythropoietin (WHO, 2017).

There are negative consequences for both the mother and her infant as a result of HIV infection in pregnancy. The adverse pregnancy outcomes linked to anaemia in HIV pregnant women are; pre-eclampsia, preterm labour and low birth weight (Nandlal et al., 2014). Moreover, breastfed infants who are exposed to HIV, despite being HIV negative are reported to be at a higher risk of infectious disease morbidity and more frequent hospital admission when compared with HIV unexposed breastfed infants (Finkelstein et al., 2020).

Furthermore, in one study pregnant women who are HIV positive were found to be 2.5 times more likely to be anaemic than the HIV negative women (Tunkyi & Moodley, 2016). This could be attributed to the fact that HIV positive pregnant women are vulnerable to opportunistic infections, which can aggregate the risk of developing anaemia. Antiretroviral therapy has frequently been linked to the development of anaemia in women and their newborns (Nandlal et al., 2014). The antiretroviral therapy taken by HIV pregnant women also has a negative effect on erythropoiesis as a result bone marrow suppression due to cytokine release causing a reduction in the production of RBCs which is associated with anaemia (Mockenhaupt et al., 2015; WHO, 2017). In study carried out in Durban, South Africa found the prevalence of anaemia in HIV infected women as 64.6% (Tunkyi & Moodley, 2017). The study associated its higher prevalence rate of anaemia in HIV infected women to an increase in the awareness and acceptance of HIV screening.

1.2.3.3 Socio-demographic and socio-economic causes of anaemia

The prevalence and severity of anaemia among pregnant women are greatly attributed to sociodemographic and nutritional factors such as; age, parity, educational and socio-economic status, food insecurity and nutritional deficiencies (Aziz Ali et al., 2019). Cultural factors such as taboos prohibiting pregnant women from consuming meat and egg-based foods put pregnant women at an increased risk of developing anaemia (Ahenkorah et al., 2016). Sociodemographic conditions such as; teenage pregnancy, unemployment, poor nutritional intake, short pregnancy intervals, prim gravida and multigravida are some of the contributing factors of anemia in pregnancy (Okubatsion, 2015; Sholeye et al., 2017).

Socio-economic status of households has been significantly linked to anaemia among pregnant women, with pregnant women from a lower socio-economic status being identified as having a higher prevalence compared to those with a higher socio-economic status (Lebso et al., 2017; Samuel et al., 2020). This finding can be associated to the fact that women in higher economic class can afford to buy food rich in the necessary nutrients and also afford to buy sufficient nutrients rich foods during pregnancy, while the pregnant women in lower economic class cannot afford buy sufficient nutrient rich food (Shimanda et al., 2020).

Most of these predisposing sociodemographic factors are not universal in their significance. Their significance differs from one antenatal population to another, and sometimes even within the same population not all the factors might be significant to all (Ahenkorah et al., 2016).

Below are some of the socio-demographic and socio-economic factors that contribute to the development of anaemia in pregnancy.

Age

Younger pregnant women are reported to be highly susceptible to developing anaemia than older pregnant women (Ahenkorah et al., 2016). One study found anaemia's prevalence of 73.2% among young pregnant women aged 15-19 years (Tibambuya et al., 2019). A study by Omote et al. (2020), also had a similar finding whereby pregnant women younger than 20 years of age had a prevalence of 60%. The higher prevalence rate among young pregnant women could be linked to the fact that majority of them are still undergoing rapid growth which raises their nutritional needs (for their own growth and for the growth of the fetus) and if left unmet, it elevates their risk of developing anaemia (WHO, 2017). Another probable aspect of younger pregnant women being more susceptible to anaemia is that majority of the younger pregnant women maybe unprepared economically to deal with pregnancy related costs such as transport money to attend ANC regularly and money to buy nutritional food throughout pregnancy. This in turn would lead to poor use of ANC services and poor nutritional intake (Fayed et al., 2017). Another study also found a higher occurrence of anaemia in pregnant women below the age of 20 years (Zehra et al., 2014). The study attributed its finding to the fact that; pregnancies below 20 years of age are teenage pregnancies of which most of them are unplanned or unwanted. Hence, those teenagers may not be emotionally prepared to be pregnant and make changes to their daily habits in line with being pregnant, such as eating nutrient rich food that can minimize the risks of developing nutritional anemia. To the contrary, the very old (40 years and above) were found to have an increased prevalence rate of anaemia in pregnancy (Zehra et al., 2014). This is because with advanced maternal age, old women are likely to be multigravida and this means with every pregnancy there is a reduction of iron reserves causing blood loss at each delivery and predisposing the women to anaemia (Okubatsion, 2015).

Income

Economic empowerment of women, play an important role in reducing the prevalence of anaemia in pregnancy (Mockenhaupt et al., 2015). Economically and socially underprivileged women are at a higher risk of developing anaemia during pregnancy (Obai et al., 2016). Socio-economic deprivation could mean that the mother is unable to afford personal care critical during pregnancy such as purchasing food rich in iron, folic acid and vitamin B 12, and/or for purchasing supplements and this can result in adverse pregnancy outcome such as developing anaemia. A study by Ayele et al. (2020), found pregnant women whose occupation is agriculture to be 2.6 times more like undernutrition than those who are house wife. This increases their likelihood of developing anaemia. The study justified it's finding to the assumption that mothers whose occupation is agriculture spend more time doing agricultural work than caring for themselves and the work intensity might be hard for the pregnant women.

Educational level

Education level is one of the factors associated with anaemia in pregnancy. Pregnant women with secondary and above educational level are found to be less likely to develop anaemia (Sukumar et al., 2016). This is because lack of education and poor understanding regarding health-related issues could lead to delays in seeking care crucial at the time of pregnancy or could result into inappropriate management of life-threatening pregnancy complications. Lack and lower level of education have also been linked to poor awareness on risk factors of anaemia, and the possible strategies to prevent them (Bansal et al., 2020). This could increase the prevalence rate of anaemia among pregnant women with lower education level in comparison to those with higher education level. In addition, less educated pregnant women

were identified to be involved in frequent pregnancies with inadequate spacing which elevates their risk of anaemia (Anjum et al., 2020).

Low level of education is also associated with unemployment a known factor for poverty, which could increase the risk of anaemia in pregnancy (Obai et al., 2016). Moreover, educated pregnant women are more likely to have better income and able to afford and eat nutritious food, making them less likely to develop nutritional anaemia which is the common anaemia type in pregnancy (Stephen et al., 2018). Furthermore, the level of education is thought to significantly contribute to the positive pregnancy outcome in relation to anaemia development (Shimanda et al., 2020). Whereby, pregnant women who had secondary and tertiary education have been linked to several positive maternal and child outcomes, such as higher frequency of exclusive breastfeeding, attending the recommended ANC visits and utilizing of skilled health care services at the time of delivery (Stephen et al., 2018; Omote et al., 2020). Conversely, one study found a weak association between maternal education and related birth outcomes whereby only 1.2% of women with low education level (high school diploma or less) were more likely to have a poor birth outcomes than those with a high education level (Campbell et al., 2018). This weak association was ought to be influenced by other maternal factors like; smoking, drug use and anxiety which were associated with high maternal education.

Nature of diet during pregnancy

Pregnant women require increased dietary diversity and increased nutrient intake to cater for the extra needs during pregnancy (Ayele et al., 2020). This is crucial for their health and for the optimal growth of their infants. Dietary factors such as intake of fruits, red meat, green leafy vegetables, eggs prior and at the time of pregnancy as well as intake of non-food

materials like clay dirt (pica), were linked with anaemia in pregnancy (Sholeye et al., 2017; Tibambuya et al., 2019). Whereby pregnant women who consumed less than 2 meals a day, less diverse meals, less meat, less eggs and/or less green leafy vegetables were at an increased risk of developing anaemia (Anlaaku & Anto, 2017). Another study found an association between anaemia and consumption of green leafy vegetables, whereby 44.8% of pregnant women who never ate green leafy vegetables were anaemic (Bansal et al., 2020). These associations could be attributed to those diets rich in meat, eggs and green leafy vegetables are essential in reducing the risks of developing nutritional anaemias during pregnancy.

Gravidity and parity

The other factors which are significantly linked to anaemia during pregnancy are gravidity (the number of pregnancies), parity (the number of times the pregnancies were carried to a viable gestational age) and short interval between pregnancies (Shah et al., 2020). A significant link between incidence of anaemia and gravidity has been identified. Women with multigravida were found to be more anaemic and the severity increased with the higher gravidity, when compared to women with lower gravidity (Tembhare et al., 2015). This could be linked to that as the number of pregnancies increase there is a higher possibility that the pregnant mother shares the nutrients with her fetus, resulting in loss of iron and other related nutrients subsequently leading to anaemia (Adam et al., 2018). Parity and the birth interval between pregnancies have been associated with anaemia. With increased parity and closer interval between pregnancies especially below 18 months, there is an increased risk of developing anaemia in pregnancy (Zehra et al., 2014). This could be associated with the fact that, increased parity and short pregnancy interval may deprive the pregnant women the time she needs to replenish lost nutrient stores necessary prior another reproductive cycle begins.

(Abriha et al., 2014). In contrast, Mockenhaupt et al. (2015) found inverse relationship between parity and anaemia, whereby anaemia prevalence in pregnancy decreases with increased parity. According to the study, the possible reason to its finding is that with increased parity there is increased awareness of nutritional diets and increased value of drugs used to reduce the risks of anaemia. The study also found that women of higher parity booked for ANC earlier in gestation when the iron requirements are still low compared to women of lower parity who booked later in pregnancy when the iron are higher, which predisposes them to anaemia.

1.2.3.4 Other causes of anaemia

Other causes such as chronic blood loss, gestational age and the stage of ANC commencement we all linked to anaemia in pregnancy (Sabina et al., 2015; Zekarias et al., 2017; Okubatsion, 2015; Tibambuya et al., 2019). Pregnant women with chronic blood loss during or prior pregnancy such as being a frequent blood donor, having abnormally menstrual cycles and having lost large amount of blood either during or after surgery are at an increased risk of developing anaemia at the time of pregnancy. Women in the second and third trimester were found to be 3.09 and 3.68 times more likely to be anaemic than those in their first trimester respectively (Lebso et al., 2017). This finding could be linked to the fact that at the time of pregnancy, the calorie and nutrients requirements peak up to aid the increased maternal metabolism, blood volume and the supply of nutrients to the fetus. These demands increase mostly during the first and second trimesters of pregnancy (Zehra et al., 2014). Moreover, the stage at which pregnant women seek for ANC services also play a crucial role in the prevention of anaemia (Ajepe et al., 2020). Pregnant women who report for ANC early in their pregnancy are offered education on the predisposing factors to anaemia and

how to prevent or manage the occurrence of those factors and screened for early signs of anemia. Hence, less likely for them to be anaemic than those who only start with ANC late their pregnancies (Anlaaku & Anto, 2017). The lower prevalence of anaemia among those who booked for ANC early in pregnancy, could also be due to the haemodilution effect of pregnancy and increased fetal demands that maximizes only after the first trimester (Mockenhaupt et al., 2015).

1.2.4 Physiologic anaemia of pregnancy

In normal pregnancy there is an increase in the RBC mass and plasma volume in order to accommodate the needs of the growing uterus and fetus, but the increase in the plasma volume is more than the RBC mass (Sabina et al., 2015). This increase commence around the 6th week of pregnancy and reaches the maximum value at around 24 weeks gestation, with the maximum value of the plasma volume being 40%-50% more than it was at the start of pregnancy (Means, 2020). However, this disproportionate increase in the plasma volume more than the red cell mass results in haemodilution and decreased haemoglobin concentration causing anaemia in pregnancy, and this process is referred to as physiological anaemia of pregnancy (Gebreweld & Tsegaye, 2018). Haemodilution at the time of pregnancy is followed by an elevation in the iron demand of about 1 000 mg (Mireku, 2016). The required extra 1 000 mg of iron is consisting of 300 mg for the fetus and placenta, 500 mg for increased maternal HB and 200 mg for excretion (Satyam & Khushbu, 2015). Hence, physiological anaemia of pregnancy develops as a result of combined effects of haemodilution and negative iron balance (Sabina et al., 2015).

Despite the physiological decrease in the HB causing anaemia, it has also been reported to have positive outcomes for the baby. The drop in the HB reduces the blood viscosity which is ought to enhance the placental perfusion offering a better maternal-fetal gas and nutrients

exchange (Satyam & Khushbu, 2015; Mireku, 2016). Additionally, it also appears that the increase in plasma volume is an indication of the normal growth of the fetus, and one of the hallmarks of a successful pregnancy (Sifakis & Pharmakides, 2000).

1.2.5 Consequences and burden of anaemia in pregnancy

The WHO has identified anaemia in pregnancy as a global health concern which can result in serious complications for the mothers and their babies (Daru et al., 2018). Anaemia in pregnancy impairs the health and wellbeing of women, and it is the major cause of morbidity and mortality of pregnant women and elevate the risks of perinatal and neonatal mortality (Zekarias et al., 2017; Okubatsion, 2015; Aziz Ali et al., 2019). The risks of maternal and neonatal mortalities increase with the severity of anaemia (Pavord et al., 2020).

Maternal consequences

Anaemia in pregnancy has been linked with increased prevalence of ante-partum and post-partum hemorrhage (Daru et al., 2018). Women who have anaemia postpartum are prone to greater stress and postpartum depression (WHO, 2017). Mild anaemia might not have any effect on pregnancy except that it causes iron stores to be low which can lead to moderate and severe anaemia in subsequent pregnancies (Sabina et al., 2015). Moderate anaemia in pregnancy has been linked to preeclampsia, puerperal sepsis, failure of lactation, delayed wound healing and thromboembolic complications leading into sub involution of the uterus (Bansal et al., 2020). A reduction in maternal breast milk production and maternal depletion of iron stores during and after the postpartum period, as well prolonged hospital stay are some of the negative effects of moderate anaemia towards the mother (Api et al., 2015).

Depleted blood reserves due to anaemia may increase the need for blood transfusion during delivery (Achebe & Gafter-Gvili, 2017). Moreover, pregnant women with severe anaemia are prone to the following signs and symptoms; fatigue, dizziness, weakness, breathlessness, drowsiness, paleness of the skin, increased cardiac stress and cardiac failure (Obai et al., 2016; Sabina et al., 2015; Adam et al., 2018). The signs and symptoms caused by severe anemia if not addressed and treated promptly can result in maternal mortality (Satyam & Khushbu, 2015).

Neonatal consequences

Infancy anaemia can lead to prematurity and perinatal loss (Sabina et al., 2015). The risk of prematurity may increase due to oxidative damage to erythropoiesis (Di Renzo et al., 2015). Children born by mothers who had IDA during pregnancy are observed to have learning and memory impairments, which could persist into adulthood (Achebe & Gafter-Gvili, 2017). It increases the risks of foetal growth retardation and low birth weight, premature delivery, reduces resistance to infection and delay psychomotor development in infancy (Satyam & Khushbu, 2015; Helmy et al., 2018; Ajepe et al., 2020).

Burden of Anaemia

Anaemia is a serious public health problem with severe consequences for human health and impacting socio-economic development due to its costs. Anaemia causes a reduction in the work capacity of an individual (Satyam & Khushbu, 2015). Hence, if work involves manual labour the anaemic pregnant women would have a decline in her income. A study by Shimanda et al. (2020) on socioeconomic factors associated with anaemia, found 2.5%

reduction in the anemic adult income and that anaemia increases the individual's medical expenses. Anaemia is also reported to have a burden at a household level, whereby women who are anaemic find it difficult to cope with household chores and childcare (Satyam & Khushbu, 2015). Additionally, fatigue and depression caused by anaemia may negatively influence the mother to child relationship (Garzon et al., 2020). This likely because most of the times the mother may be too tired to constantly interact or bond with the child, and as a result of depression the mother may be shouting to the child unnecessarily.

The consequences of anaemia in pregnancy are enormous, affecting Namibia as a country, Africa as a continent and the world at large. Because anaemia in pregnancy reduces the physical and productivity of adults, it threatens the household food security and income. This may mean that the government of the countries severely affected by anaemia should have a budget set aside to assist families severely affected by anaemia. This will eventually distort human resource development and economic prosperity of the country (Mehrotra et al., 2018). Anaemia affects the learning and memories of children, which could persist in adulthood (Achebe & Gafer-Gvili, 2017). This specific impact of anaemia in pregnancy, indicates that countries with higher prevalence rate of anaemia are at a risk of having future population with learning and memory problems and this might affect the development of such countries.

1.2.6 Diagnosis of anaemia

The diagnosis of anaemia is carried out into two main ways, through evaluation of the clinical profile (signs and symptoms) and by assessing the full blood picture (blood levels for; HB and RBC indices, iron, ferritin, TIBC, transferrin, folate and Vitamin B 12) of the patient (Helmy et al., 2018).

1.2.6.1 Clinical diagnosis

An accurate physical examination can determine the severity of anaemia, and for this reason a thorough physical examination and detailed history of a pregnant woman are essential steps in the diagnosis of anaemia (Di Renzo et al., 2015). A detailed history includes; enquiring on the present anaemia symptoms and the duration, transfusion history, dietary history like craving for unusual food items (pica), travel history (to malaria endemic areas), presence of chronic diseases (HIV), bleeding (gastrointestinal bleeding) and history of trauma or recent surgery (Alli et al., 2017). These enquiries would serve as a guide in determining whether the pregnant woman is likely to be anaemic or not and propose its cause.

1.2.6.2 Laboratory diagnosis

In order to diagnose and classify anaemia in the laboratory, cut off values are determined and set for the individual's HB and red cell indices, iron, ferritin, TIBC, transferrin, folate and Vitamin B 12. These cut off values vary between groups of individuals (babies, children, adults and according to sex and when there is a change in life such as in pregnancy between pregnant and non-pregnant women). In pregnancy the lower cut off values for Hb are; 11.0 g/dL in the first and last trimester and 10.5 g/dL in second trimester (Satyam & Khushbu, 2015). However, differential diagnosis should be the next step in the laboratory diagnosis. In differential diagnosis anaemia is further classified based on; RBCs characteristics (hypochromic microcytic, normocytic normochromic, macrocytic normocytic), underlying mechanism (decreased bone marrow production, bone marrow aplasia, ineffective hematopoiesis) and peripheral loss or destruction (bleeding, sequestration, haemolysis) (Alli et al., 2017). Figure 1.1 below shows an algorithm on the interpretation of pathology results and Table 1.1 shows RBC morphological characteristics.

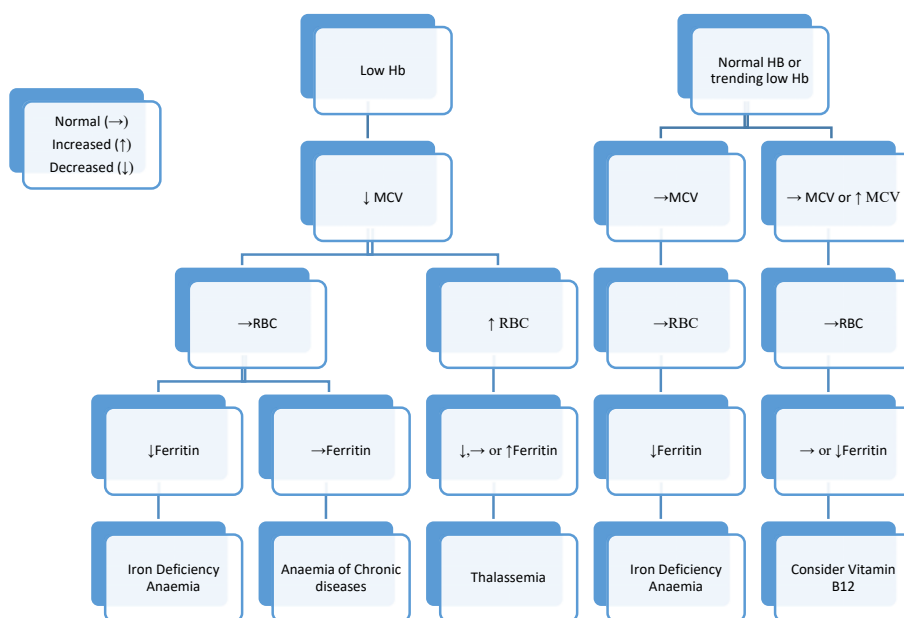


Figure 1.1 Algorithm on the interpretation of pathology results (Adapted from Frayne & Pinchon, 2019).

Table 1.1 Red blood cell morphological characteristics.

Morphological Observation	Significance	Further Tests recommended
Oval macrocytes, tear drops, basophilic stippling, right shift	Macrocytic anaemia	Serum vitamin B12 and folate levels
Hypochromia, microcytosis	IDA, anaemia of Chronic diseases, thalassaemia	Serum ferritin, transferrin, iron levels, HB electrophoresis
Spherocytes	Hereditary spherocytosis, warm autoimmune haemolytic anaemia (AIHA)	Coombs test, red cell membrane analysis
Elliptocytes	Hereditary elliptocytosis	Red cell membrane analysis
Auto agglutination	Cold AIHA	Coombs test

(Adapted from Alli et al., 2017)

Iron Deficiency Anaemia

A patient with IDA usually has the following FBC results; decreased HB concentration, decreased mean cell haemoglobin concentration (MCHC), decreased mean cell haemoglobin (MCH) and mild thrombocytosis (Renzo et al., 2015). Other tests results in IDA are; decreased iron, increased total iron binding capacity (TIBC) and transferrin (Al-Khaffaf et al., 2020). On examination of peripheral blood film, suspected IDA presents the following likely findings; microcytic hypochromic RBC with anisocytosis and poikilocytosis, presence of tea drop cells, pencil cells and occasionally target cells (Adewoyin, 2014). Microcytic hypochromic RBCs can also be found in haemoglobinopathies, thus they need to be interpreted with caution (Pavord et al., 2020). Moreover, the serum transferrin level increases in IDA as a result of the body trying to compensate for low iron levels and it correlates positively with TIBC (Bouri & Martin, 2018). Thus, when IDA is suspected, low serum ferritin is the most specific laboratory test for IDA (Ajepe et al., 2020). This is because ferritin reflects the total body iron stores, and iron deficiency is the only clinical situation associated with extremely low values of ferritin (Achebe & Gafer-Gvili, 2017). The cut off levels to be used for ferritin is an ongoing debate. However, the ferritin levels of $<30\mu\text{g/L}$ is currently the recommended one until good quality evidence suggesting another cut off emerges (Pavord et al., 2020).

To the contrary, serum ferritin is an acute phase reactant and its concentration is elevated when there is infections, systemic inflammation, malignancies, hepatopathies and chronic renal failure, causing ferritin levels to be falsely normal or elevated despite the presence of anaemia making its use for diagnosis in patients with such underlying conditions complicated (Satyam & Khushbu, 2015; Renzo et al., 2015). Thus, low levels of serum ferritin cannot be the only diagnostic test of iron deficiency, it should be accompanied by a reduced transferrin saturation (Api et al., 2015). A transferrin saturation of $<15\%$ is an indication of IDA

(Sukumar et al., 2016). Additionally, the evaluation of C-reactive protein (CRP) level may also aid in obtaining the correct diagnosis of anaemia including infections and inflammations. Whereby if the CRP level is increased, a re-evaluation of ferritin should be carried out after normalizing CRP concentration (Garzon et al., 2020).

Furthermore, serum hepcidin levels use in the diagnosis of IDA is reported to be emerging, however its correlation with clinical outcomes are unknown and there is limited evidence to support its use in pregnancy (Pavord et al., 2020). The red cell distribution width (RDW) is another parameter of the FBC that can prompt for suspected IDA. This is because it shows the variations in the RBCs which is the initial morphological change occurring in IDA, and should be expected to increase due to microcytic population in the blood (Di Renzo et al., 2015). Figure 1.2 illustrates the algorithm for the diagnosis of iron deficiency anemia during pregnancy.

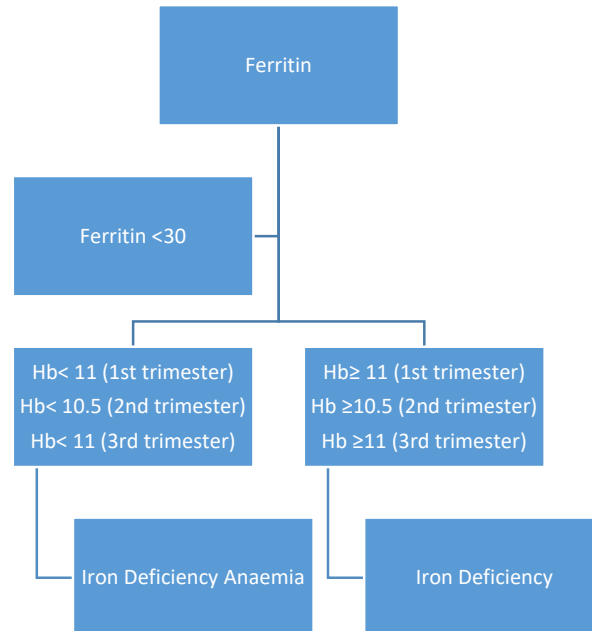


Figure 1.2. Algorithm for the diagnosis of iron deficiency anemia during pregnancy (Adapted from Apili et al., 2020).

Vitamin B deficiency Anaemia

Vitamin B12 deficiency is screened by evaluating the FBC, blood film and determining the serum Vitamin B12 levels. On evaluating the FBC, vitamin B12 deficiency is prompted by having elevated mean corpuscular volume (MCV) of >99 fL, while on assessing the blood film from a full blood count it will show macrocytic RBCs and hyper-segmented neutrophils (Berg and Shaw, 2013). Moreover, during FBC evaluation the RDW is expected to increase in vitamin B12 (Bouri & Martin, 2018). This is associated with the variations in the sizes of RBCs. Despite the FBC and blood film assessment, the most direct analysis and first assay to measure the status of vitamin B12 in the blood is the measurement of total serum vitamin B12 level, of which a level of < 150 pg/mL (111 pmol/L) is considered as diagnostic for vitamin B12 deficiency (Langan & Goodbred, 2017). Moreover, vitamin B12 deficiency levels are categorized by WHO as follow; normal > 221 pmol/L, low 148-221 pmol/L and acute deficiency < 148pmol/L (Hannibal et al., 2016).

Folate deficiency Anaemia

Folate deficiency is screened through assessment of the FBC, blood film and determining the serum folate level. The FBC in folic acid deficiency would show a decrease in HB and haematocrit (HCT) levels, increased MCV to more than 100 fL, on the blood film assessment would show macrocytic RBCs or megaloblasts and hyper-segmented neutrophils, and on analyzing the serum folate a level <2 ng/ml is considered deficient (Khan & Jialal, 2020). Although assessing serum folate level is identified as the most appropriate test for diagnosing folate deficiency (Mireku et al., 2017). It has been reported to show low sensitivity and specificity resulting in the use of other blood parameters like RBC folate and biomarkers like homocysteine (Alkhaldy et al., 2020). Moreover, as a result of some diagnostic features such

as increased MCV, macrocytic RBCs and hyper-segmented neutrophils that are consistent with vitamin B12, requesting vitamin B12 together with folate levels will aid in differentiating between the two.

Thalassemia

Thalassemia is screened by assessing the FBC and blood film, however ferritin level can also guide in thalassemia identification. On evaluating the FBC low MCV ($<80\text{fL}$), low MCH ($<27\text{pg}$) are generally used for positive screening of thalassemia and on assessing the blood film microcytosis, hypochromia and anisopoikilocytosis points towards thalassemia identification (Lee et al., 2019). However, IDA can also present with low MCV and low MCH and microcytosis is also prevalent on the blood film (Renzo et al., 2015). This makes it challenging to use only the FBC and blood film to screen for thalassemia, in this case serum ferritin level can be of use. Whereby if the serum ferritin is normal and the MCV is low in the absence of inflammation the diagnosis is pointing towards thalassemia and other investigations are required (Api et al., 2015). The other investigations that are very useful in the confirmation and characterization of the thalassemia are the advanced electrophoresis techniques including high performance liquid chromatography (for diagnosing β -thalassemia) and molecular characterization (for diagnosing α -thalassemia) (Lee et al., 2019).

Anaemia of Chronic diseases (ACD)

An assessment of the FBC, morphology of the blood film, reticulocyte count, serum ferritin, serum bilirubin and lactate dehydrogenase (LDH) are critical in the diagnosis of ACD (Madu

& Ughasoro, 2017; Di Renzo et al., 2015). Anaemia of chronic diseases is typically a normocytic normochromic with low reticulocyte count, characterized by mild to moderate HB levels (8-10 g/L) (Chaparro & Suchdev, 2019). Assessing the blood film may provide the underlying cause of ACD, with thrombocytosis indicative of chronic hemorrhage, toxic granulation in neutrophils indicative of severe sepsis and hyper-segmented neutrophils directive of mixed nutritional deficiencies found in malignant conditions (Madu & Ughasoro, 2017). Moreover, the ferritin level is increased in ACD due to an increased in the iron regulator hepcidin which is elevated in infection or inflammation, and as a result of failure to release iron from the ferritin stores there is a low iron, low transferrin saturation and low TIBC in ACD (Bouri & Martin, 2018).

Therefore, the correct anaemia diagnosis and treatment lead to proper management of fetal and maternal risks, and improved perinatal outcome (Helmy et al., 2018).

1.2.7 Prevention and treatment of anaemia

Preventive and control measures of anaemia

Anaemia in pregnancy resulting from nutritional deficiencies, malaria infection and helminth infestation are preventable causes (Mockenhaupt et al., 2015). Thus, in order to effectively prevent and address anaemia especially at a country level effective assessments of individual contributing factors and identifying the most appropriate interventions are essential. Commencing with ANC early in gestation is one of the key components for prevention and controlling anaemia in pregnancy. This is because ANC visit enables early detection and correction of anaemia, as well allow for the identification and immediate treatment of malaria and helminths infections which are common in pregnancy and predisposes the pregnant

women to anaemia (Anlaaku & Anto, 2017). Additionally, access to quality and timely healthcare services has been noted as also one of the effective measure in the reduction of risks associated with anaemia in pregnancy (Omote et al., 2020).

Nutrition specific preventive and control measures

Nutrition specific interventions are those measures directed towards the primary causes of nutritional anaemia, mainly from poor dietary intake or malabsorption of essential nutrients (iron, vitamin B12 and folic acid). Thus, the preventive measures for nutritional deficiency anaemias are aimed at increasing dietary diversity, improve bioavailability of nutrient rich food, make fortified food accessible to all and advocate for the diets that improve absorption (WHO, 2017). This can be achieved by improving the year-round local availability of nutrient rich food, reducing the prices of such food in retailer shops and by including components in the diet that improve absorption. Additionally, pregnant women are recommended to take the iron and folic acid supplementation early in pregnancy to avoid complications related to iron and folic deficiencies (Okubatsion, 2015).

Furthermore, the recommended dietary folate intake in people beyond the age of 18 years is 240 μ /day, but the requirements in pregnancy is up to 400 μ /day (Alkhaldy et al., 2020). This means diet alone cannot meet the pregnancy folate requirements and should be supplemented by folic acid tablets. Moreover, as part of the nutrition specific preventive measures the WHO recommends intermittent iron supplementation for menstruating women living in areas where the prevalence of anaemia is 20% or more (Wemakor, 2019).

Furthermore, as part of addressing nutritional anaemia it is also recommended that pregnant women be screened for anaemia at booking and at 28 weeks gestation and be given

information regarding diet in pregnancy with details of food rich in iron, folate, vitamin B12 and other necessary micronutrients (Sabina et al., 2015). Thereof, at each ANC visit healthcare workers should assess and document compliance to medication and dietary recommendations, as well assess and document side effects and provide advice for management of the side effects (Satyam & Khushbu, 2015).

Infectious and parasitic preventive and control measures

Infectious and parasitic interventions are those measures that directly relate to the prevention of anaemia through vector control and periodic treatment of at-risk populations such as pregnant women and children, living in endemic areas (WHO, 2017). Thus, in order to prevent and control anaemia resulting from parasitic infections eliminating the breeding grounds for mosquitoes such as improving sanitation and offering health education on personal malaria protection (using mosquito repellents, wearing long-sleeved shirts and long pants) are amongst the malaria control effective measures (Di Renzo et al., 2015). Issuing insecticide treated nets to pregnant women in the initial stages of antenatal visits is also one of the essential interventions for anaemia resulting from parasitic infections (Stephen et al., 2018). This is because insecticide treated nets reduced the risks of the pregnant women being bitten by mosquitoes while sleeping which can result into malaria one of the risk factors for anaemia (Okia et al., 2019). Additionally, intermittent preventive treatment of asymptomatic pregnant women and case management of malaria are also reported as effective measures for controlling malaria infection (Mockenhaupt et al., 2015; Mlugu et al., 2020).

Moreover, as a preventive measure for helminths infestation, antihelminths treatment should be issued through health services (maternal and child health), school health programme and community interventions targeting other vulnerable groups such as adolescent girls

(Shiferaw et al., 2017). Additionally, interventions to break the transmission routes should be prioritized such as provision of safe use and drinking water, improvement of community sanitation and advocating for good personal hygiene. Ongoing efforts should also be made towards HIV prevention and control which is among the risks factors of anaemia (Mockenhaupt et al., 2015).

Despite the various nutrition-specific, infectious and parasitic interventions on anaemia, studies on prevalence and etiology of anaemia in pregnancy are recommended for the ongoing management of anaemia (Mockenhaupt et al., 2015).

1.2.8 Treatment of anaemia

Each case of anaemia should be addressed and treated in order to prevent adverse perinatal outcomes associated with anaemia. The early treatment of mild anaemia prevents anaemia to progress to moderate and severe forms (Daka et al., 2018), which are highly associated with increased risk of fetal and maternal mortality and morbidity requiring long term treatment which can be very costly to both the mother and supplier of funds.

As the need to iron picks up at the time of pregnancy, prophylactic oral iron therapy is provided to all pregnant women with normal laboratory values, but when severe anaemia is detected intravenous (IV) iron therapy should be the priority option due to its effectiveness in resolving anaemia in comparison to oral therapy (Api et al., 2015; Renzo et al., 2015). Intravenous iron therapy promotes a higher and faster, increase of HB concentration and serum ferritin levels which subsequently correct the anaemia (Garzon et al., 2020). However, the mode of iron therapy is mostly determined by the degree of anaemia, the stage of the pregnancy and the factors that affects the gastrointestinal absorption of iron (Achebe &

Gafter-Gvili, 2017). Thus, it is crucial to treat every case of anaemia to avoid and minimize adverse perinatal outcomes related to this disorder.

The average daily absorption of iron from diets is only 1-5 mg, however the iron demands in pregnancy increase gradually from 0.8 mg per day in the first trimester to 7.5 mg per day in the third trimester (Achebe & Gafter-Gvili, 2017). Making it difficult for pregnant women to satisfy their iron requirement just from normal food intake. Hence, the Centre for Disease Control (CDC) recommends that all pregnant women should commence with 30 mg per day of iron supplements at the first prenatal visit, while the WHO suggests 60 mg per day for all pregnant women (Achebe & Gafter-Gvili, 2017).

When it comes to the treatment and monitoring of Vitamin B12 deficiency, an oral administration of high vitamin B12 has been reported to be as effective as intramuscular administration for correcting anaemia and neurological symptoms (Langan & Goodbred, 2017). However, intramuscular therapy is linked to more rapid improvement and needs to be considered in critical patients (Renzo et al., 2015).

Moreover, pregnant women with folate deficiency should be given supplemental folic acid in correlation with severity of the deficiency (Assefa et al., 2019). Typically, oral folic acid suffices for the treatment but in pregnant women who cannot tolerate oral medications, intravenous, subcutaneous or intramuscular formulation of folic acid can be utilized (Khan & Jialal, 2020).

Furthermore, when it comes to malaria and helminths treatment the WHO recommended intake of sulphadoxine pyrimethamine (antiparasitic drug used to treat malaria) as intermittent preventive treatment in pregnancy and antihelminthics in the second trimester of pregnancy (Lumora et al., 2019). Blood transfusion containing 10mL/kg of packed cells administered over a period of 2-4 hours is recommended for anaemia due to malarial infection in high

transmission settings (HCT of less than 15%) or in low transmission setting (HCT less than 20%) and for patients with hypovolemic whole blood transfusion might be suggested (Kaur et al., 2017). Additionally, pregnant women living in areas where helminths are common, should be recommended to deworm after the first trimester to minimize the risk of severe maternal anemia and to reduce infant mortality (Shiferaw et al., 2017).

CHAPTER 2: METHODOLOGY

2.1 Study design and approach

This was a cross-sectional analytical study, which was conducted in randomly selected regions of Namibia. There are 14 regions in Namibia namely: Erongo, Hardap, Karas, Kavango East, Kavango west, Khomas, Kunene, Ohangwena, Omaheke, Omusati, Oshana, Oshikoto, Otjozondjuba and Zambezi region. The regions were alphabetically arranged, and every 3rd region was selected as a study region. Therefore, Karas, Khomas, Omaheke and Oshikoto were the randomly selected regions for the study. A cross-sectional study measures variables at a single point in time. Cross-sectional study designs are used for population-based studies and assess the prevalence of diseases in clinical based samples (Seratia, 2016). With a cross-sectional study approach one can also measure the association between variables and identify the significance difference between, exposure and health outcomes of exposed and unexposed groups (Alexander et al., 2014). Therefore, a cross-sectional study was selected to be a design for this study, as the study aims at providing the estimate of anaemia prevalence among pregnant women attending ANC in Namibia. The study also intended to find the association between anaemia and the different exposures (socio-demographics), thus a cross-sectional study was the appropriate design for this measure.

2.2 Study area

The study was carried out in the 4 randomly selected regions of Namibia, namely, Karas, Khomas, Omaheke and Oshikoto regions (Figure 2.1). Karas is situated in the south of Namibia an area covering 161 514 km². Khomas region is in the central highland of the country with area of 36 964 km², and it is the region with the highest population in the country. Omaheke region is situated on the eastern boarder of Namibia and the western

extension of the Kalahari Desert and has an area of 84 981 km². The fourth region of the study is Oshikoto which is located in the central part of Namibia and has an area of 38 685 km². According to the Namibia inter-census demographic survey report of 2016, the regions of study have the following total populations and the female percentages respectively; Karas 85 759 and composed of 49.5% females, Khomas 415 780 of which 50.4% are females, Omaheke 74 629 with 47.2% being females and Oshikoto region 195 165 with 51.8% of the population being females (NSA, 2016). On the other hand, the study area for the socio-demographic aspects of the study was only Khomas region. This has been due to Covid-19 restrictions during the data collection process. Thus, the researcher could not travel to all 4 regions of the study in order to collect socio-demographic information from the study participants as originally intended. Data on sociodemographic factors was only collected from Khomas region at the Katutura State Hospital ANC clinic. Thus, the inference on the association between anaemia and socio-demographic factors could not be made for all regions but only for Khomas region. Katutura hospital was purposively selected as a study area because it is the largest hospital not only in Khomas region, but in the whole country. It is also one of the hospitals that had more ANC attendees compared with any other healthcare center in the region, and on the days allocated for ANC visits (Tuesday and Wednesday) it receives between 30 to 40 pregnant women attending their first ANC. Hence, it is thought to give a good sample that represents the ANC population in Khomas region. Katutura hospital is located between Independence Avenue and Hans Dietrich Genscher Street in Windhoek.

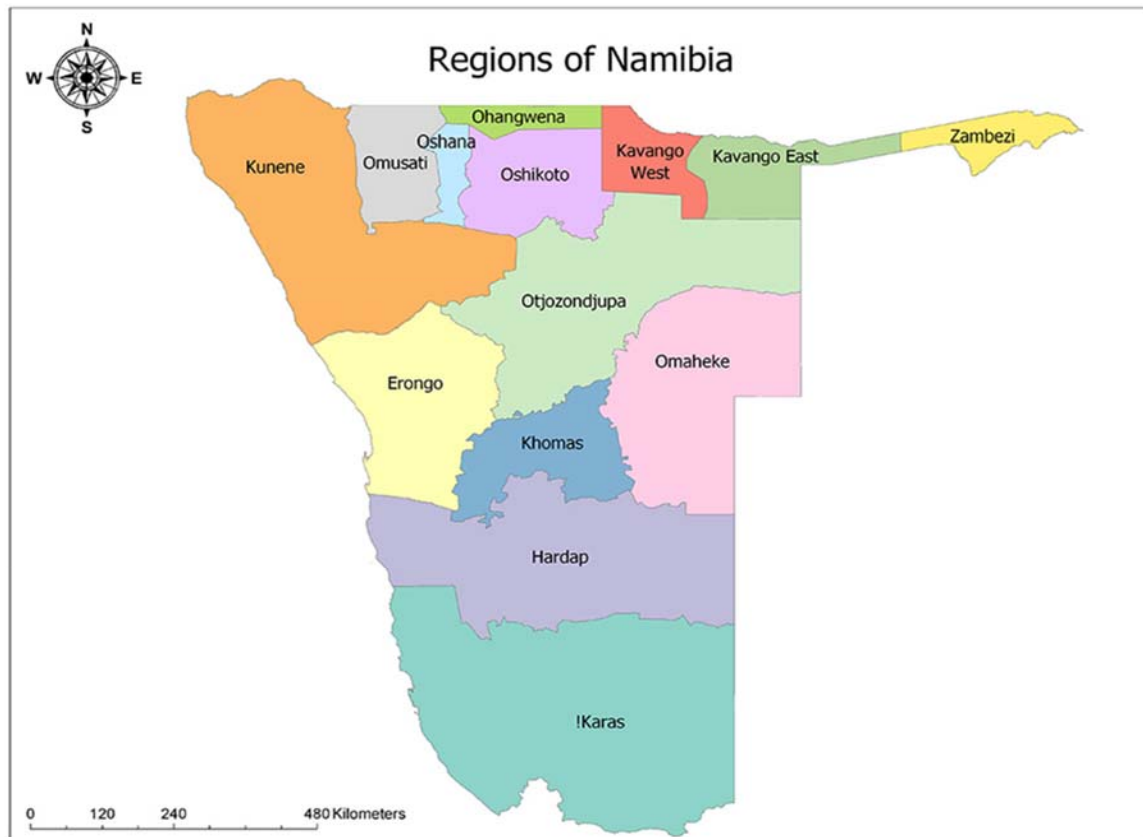


Figure 2.1: Geographic map of the regions of Namibia

Source: <https://www.mappr.co/counties/namibia/>, retrieved 17 June 2021.

2.3 Population of the study

The study population was comprised of pregnant women who reside in the study areas. Whole blood specimens were collected from pregnant women attending ANC at selected study regions during the study period. The sample population for the data collected by means of a questionnaire were the pregnant women attending their first ANC visit at Katutura state hospital in Khomas region at the time of data collection. The participants for the questionnaire were selected based on convenience, whereby only pregnant women available

and who consented at the time of data collection were included. Each participant was only recruited once.

2.4 Inclusion and exclusion criteria

All specimens from selected study regions collected from women attending the respective ANC clinics in the region were eligible for the study. The laboratory information system (LIS) was used to scan for the history of anaemia in the selected pregnant women. Those found with a history of anaemia, having malaria infection and those who were identified as HIV positive were excluded from the study.

On the other hand, the inclusion criteria for the pregnant women to complete the questionnaires were those who were on their first ANC visit, the pregnant women who did not start with any iron supplements, those with no history of anaemia and those who were aged 18 and above who consented to the study. The exclusion criteria for partaking in the questionnaire completion were the critically ill pregnant women, recently transfused and the ones that declined consent.

2.5 Sample size and sampling method

The sample size was determined using Epi info version 7 with 95% confidence interval (CI), 5% marginal error, and the prevalence of anaemia among pregnant women in Namibia as reported by World Bank, (2017) which was 27%, an assumption of 10% non-response rate and a design effect of 1. A total sample size of 333 was calculated. Multistage sampling technique was used to select study participants, of which 4 regions (Karas, Khomas, Omaheke and Oshikoto) were selected from the 14 regions of Namibia using a randomized

method. A proportional allocation was used to obtain the sample size from each selected region, and a total of 84 specimens per region was determined. The sample for the questionnaire were all 84 pregnant women who consent to partake in the study from whom the specimens for Khomas region were drawn. The first interviewed participant for the completion of the questionnaire was randomly selected by a lottery system, and afterwards every second pregnant women who consent to partake in the study was selected as a study participant and issued with a questionnaire to complete. This process was carried out until the required sample size was achieved.

2.6 Data collection and procedure

2.6.1 Data collection method

Blood samples were collected by registered nurses for routine analysis at NIP from pregnant women in the selected regions who come for routine ANC testing. Serum samples were used to test for Hepatitis B surface antigen and when the testing was completed, the serum specimen remaining was used for research purposes to test for ferritin and vitamin B12. The results for the HB and MCV were analyzed using Sysmex XN-1000 (Sysmex Cooperation, Windhoek, Namibia). The specimens from the regions outside Khomas were packed in cooler boxes labelled with biohazard stickers and transported at 2-8°C to the laboratory.

Data on socio-demographic information, socio economic status and nutritional were collected through self-administered questionnaire-based interview. The questionnaire for this study was developed based on the WHO guidelines as per study by Jufar & Zewede, (2014).

2.6.2 Analytical laboratory methods

The analytical laboratory tests measured in this study were HB, MCV, Ferritin and Vitamin B12. The HB and MCV were carried out at various Namibia Institute of Pathology (NIP) laboratories within the selected regions by qualified medical technicians and technologists in the haematology departments. While, Ferritin, and Vitamin B12 were done on the Alinity instrument (Abbott, Windhoek, Namibia) at the NIP reference laboratory central hospital in the Chemistry department in Windhoek. The serum specimens for Ferritin and Vitamin were transported to the laboratory for analysis at 2-8°C.

Anaemia in pregnancy was defined as HB level less than 11 g/dL, and further classified based on the severity as per WHO criteria; mild anemia (HB between 10-11 g/dL), moderate anaemia (HB between 7.0-9.9 g/dL) and severe anaemia (HB <7.0 g/dL) (Asrie, 2017; Anlaaku & Anto, 2017). The criteria for the diagnoses of Vitamin B12 deficiency was as follow; Vitamin B12 level between 200-300 pg/mL was defined as low, vitamin B12 level >300 pg/mL as normal and vitamin B12 level <200 pg/mL as diagnostic of vitamin B12 deficiency (Achebe & Gaftor-Gvili, 2017). Furthermore, low serum ferritin (<30 ng/mL) together with HB levels were used to diagnose for iron deficiency and iron deficiency anaemia as adopted from the criteria developed and previously used in the study by (Al-Naseem et al., 2021). When the serum ferritin level is low (<30 ng/mL) but the HB is normal ≥ 11 g/dL the diagnosis is iron deficiency, however if the serum ferritin is low (<30 ng/mL) and the HB is also low <11 g/dL then the diagnosis is iron deficiency anemia (Api et al., 2015; Short & Domagalski, 2013). A low serum ferritin levels depict depleted iron stores, and hence is a best indicator for detecting iron deficiency (WHO, 2011). Thus, when a serum ferritin levels goes below 30 ng/mL, iron deficiency can be ascertained (Al-Naseem et al., 2021; Soppi, 2019). Moreover, the MCV level together with HB value were used to classify anaemia as either; microcytic anaemia (MCV <80 fL; HB <11 g/dL), normocytic

normochromic anaemia (MCV between 80-100fL; HB ≥ 11) or macrocytic normocytic anaemia (MCV >100 fL; HB ≥ 11) (Achebe & Gaftor-Gvili, 2017; Alkhaldy et al., 2020; WHO, 2011).

2.6.3 Data collection tool and procedure

A self-administered structured questionnaire was used to obtain the sociodemographic, nutritional intake and obstetric information of the study participants. The sociodemographic questions of the questionnaire assessed the participant's age, marital status and educational level. The nutritional section of the questionnaire evaluated the frequency in consumption of animal products and green vegetables by participants. The obstetric section assessed the gestational stage as well identified the current gravidity of the participants. The questionnaire was translated into the relevant languages and was administered by people who were fluent in the language and the researcher was always available to give clarity on the questions that the participants might have during the process of completing the questionnaire.

2.6.4 Measures to ensure Validity and Reliability

The validity and reliability of the analytical laboratory methods were ensured by confirming that, prior to analyzing of the research samples the maintenance and calibrations records of the instruments were up to date, and the internal quality controls (IQC) for each specific instrument to be used for the tests of interest have been run on that day and were within the acceptable limits of the test.

On the other hand, the exclusion criteria as mentioned in the “inclusion and exclusion” section, were used to ensure validity and reliability of data obtained via questionnaires. The

questionnaire used for data collection was administered in a trial run before final administration to study participants, hence ensuring its validity and reliability.

2.6.5 Data analysis

The data from the questionnaires as well instruments were entered using Microsoft excel, cleaned and sorted. The cleaning was carried out by ensuring that the data with missing and inconsistent values were identified and rectified. Finally, the data was analyzed using SPSS software version 27. The statistical analysis was performed using the Pearson's chi square test, and a p value of <0.05 was considered statistically significant. Proportion was done to describe the relation of the study participants to the relevant variables.

2.7 Ethical Considerations

Prior to commencement of the study ethical clearance was obtained from Namibia University of Science and Technology (NUST) ethics committee (Reference: FHAS 21/2019), the Ministry of Health and Social Services (MOHSS) and NIP research committee (Reference: 17/3/3 ES). A written informed consent was obtained from each participant, as an acknowledgement of voluntary participation. Participants' information and data were kept strictly confidential. Participants who completed questionnaires were informed to not write their names on the questionnaires, but instead were given a laboratory requisition number as their individual identification. Laboratory requisition numbers were also used on blood samples, instead of patient names making it difficult to link the individual patients to their questionnaires. All the extracted data were entered and kept onto a personal computer, with protected passwords and only the researcher had access to it.

CHAPTER 3: RESULTS

A total of 336 pregnant women who attended ANC in the 4 randomly selected regions (Khomas, Karas, Omaheke and Oshikoto) of Namibia, were screened for anaemia. Their ages ranged from 13-44 years, with 27 years as their mean age.

The prevalence of anaemia during pregnancy in Namibia in the current study was 20.8%, with 0% severe anaemia, 4.8% moderate anaemia and 16.1% mild anaemia. The frequency distribution of anaemia status among pregnant women in Namibia is shown in Table 3.1.

Table 3.1: Frequency distribution of anaemia status among pregnant women in Namibia

Anaemia status	Frequency (n)	%	Cumulative %
Moderate anaemia	16	4.8	4.8
Mild anaemia	54	16.1	20.8
Non-anaemic	266	79.2	100.0
Total	336	100.0	

Most of the pregnant women in Namibia were not anaemic and for those who were, they had mild anaemia.

Prevalence of Anaemia per Region

Karas region reported the highest prevalence of anaemia in pregnancy (23.8%), followed by Khomas region (21.4%), Oshikoto region (20.2%), and Omaheke region reported the least prevalence (17.9%).

Out of the total prevalence (20.8%) Karas region contributed the major portion to the overall prevalence contributing 28.6%, followed by Khomas region which contributed 25.7%, Oshikoto and Omaheke region each contributed less than 25% to the overall prevalence, with Oshikoto contributing 24.3% and Omaheke contributing 21.4%. The anaemia status per region and within the region anaemia status is presented in Table 3.2.

Table 3.2 Anaemia status per region and within the region anaemia status.

Region		Anaemia Status		
		Non- anaemic	Anaemic	Total
Khomas	Count	66	18	84
	Expected count	66.5	17.5	84
	%within count	78.6	21.4	100
	%within anaemia status	24.8	25.7	25
	% of total	19.6	5.4	25
Karas	Count	64	20	84
	Expected count	66.5	17.5	84

	%within count	76.2	23.8	100
	%within	24.1	28.6	25
	anaemia status			
	% of total	19	6	25
Omaheke	Count	69	15	84
	Expected count	66.5	17.5	84
	%within count	82.1	17.9	100
	%within	25.9	21.4	25
	anaemia status			
	% of total	20.5	4.5	25
Oshikoto	Count	67	17	84
	Expected count	66.5	17.5	84
	%within count	79.8	20.2	100
	%within	25.2	24.3	25
	anaemia status			
	% of total	19.9	5.1	25

Table 3.2 above, illustrates that most of the non-anaemic pregnant women were from Omaheke region.

Moreover, mild anaemia was reported per region as follows: Karas (19.0%), Khomas (16.7%), Oshikoto (15.5%) and Omaheke (13.1%). The frequency distribution of anaemia status among pregnant women per region is presented in Table 3.3.

Table 3.3 Frequency distribution of anaemia status among pregnant women per region

Anaemia status	Khomas		Karas		Omaheke		Oshikoto	
	Frequency	%	Frequency	%	Frequency	%	Frequency	%
	(n)		(n)		(n)		(n)	
Severe anemia	0	0.0	0	0.0	0	0.0	0	0.0
Moderate anaemia	4	4.8	4	4.8	4	4.8	4	4.8
Mild anaemia	14	16.7	16	19.0	11	13.1	13	15.5
Non- anaemic	66	78.6	64	76.2	69	82.1	67	79.8
Total	84	100	84	100	84	100	84	100

Table 3.3 above, shows that none of the regions reported with a severe form of anaemia. However, the moderate form of anaemia was consistent throughout the regions (4.8%).

Iron deficiency anaemia (IDA)

One of the types of anaemia found among pregnant women in the current study is IDA, with a prevalence of 11.9%. The regions that contributed the least to the total prevalence are Karas (2.4%) and Oshikoto (2.7%), while the regions that contributed the most to the total prevalence are Khomas (3.3%) and Omaheke (3.6%). The distribution of IDA per region is presented in Table 3.4.

Table 3.4 Distribution of IDA per region.

IDA	Region				
	Khomas	Karas	Omaheke	Oshikoto	Total
Count	11	8	12	9	40
% within IDA	27.5	20.0	30.0	22.5	100.0
%within Region	13.1	9.5	14.3	10.7	11.9
% of Total	3.3	2.4	3.6	2.7	11.9

Most of the pregnant women did not have IDA, and those who had majority were from Omaheke region.

Moreover, 25.6% of the pregnant women had non-anaemia iron deficiency, which means they have iron deficiency in the absence of anaemia. Iron deficiency can occur in the absence of anaemia when iron stores are absent but ongoing iron deficient is not sufficiently critical to produce a decreased HB concentration (Means, 2020). Additionally, 8.9 % of the pregnant

women were found to have non-iron deficiency anaemia, which means they have anaemia which is not iron deficiency. Majority of the women were categorized as normal (53.6%), indicating absence of IDA or non-IDA. The contribution of each region to IDA, non-anaemia iron deficiency, non-IDA and normal is presented in Figure 3.1

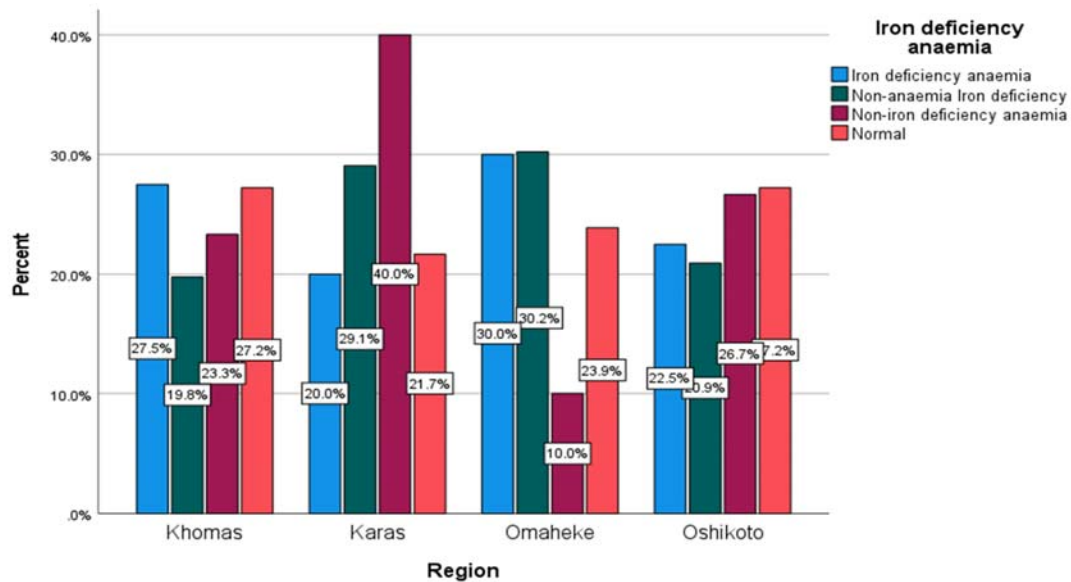


Figure 3.1 Contribution of IDA, non-anaemia iron deficiency, non-IDA and normal per region

Vitamin B12 deficiency anaemia

Vitamin B12 deficiency anaemia is another type of anaemia among pregnant women determined in the current study. Out of the 336 total pregnant women assessed in this study, 5.56% of the anaemic pregnant women presented with vitamin B12 deficiency anaemia and 7.58% of the non-anaemic pregnant women were found with vitamin B12 deficiency. Oshikoto region reported 5.97% of vitamin B12 deficiency among non-anaemic pregnant

women. Additionally, Karas and Omaheke regions reported the following prevalence of vitamin B12 deficiency among anemic pregnant women 10.00% and 33.33%, respectively and among non-anaemic pregnant women 4.69% and 10.14, respectively. The status of vitamin B12 deficiency among anaemic and non-anaemic pregnant women is presented in Table 3.5.

Table 3.5 Status of vitamin B12 deficiency among anaemic and non-anaemic pregnant women in Namibia

Anaemic Status	Vitamin B12 status	Region				
		Namibia	Khomas	Karas	Omaheke	Oshikoto
Non- anaemic	Vitamin B12 deficiency	7.58%	0%	4.69%	10.14%	5.97%
	Low	22.73%	0%	35.94%	17.39%	14.93%
	Normal	69.7%	0%	59.38%	72.46%	79.10%
Anaemic	Vitamin B12 deficiency	5.56%	0%	10%	33.33%	0%
	Low	50.0%	0%	30%	26.67%	23.53%
	Normal	44.44%	0%	60%	40.00%	79.47%

There was no case of vitamin B12 deficiency reported in Khomas and Oshikoto regions in anaemic pregnant women. Khomas region did also not report any case of vitamin B12 deficiency in non-anaemic pregnant women.

Morphological anaemia

The morphological classification of anaemia was determined. The morphological anaemia in pregnant women according to HB and MCV values per region is presented in Table 3.6.

Table 3.6 Morphological anaemia in pregnant women based on HB and MCV values per region

Morphological anaemia		Region				
		Khomas	Karas	Omaheke	Oshikoto	Total
Microcytic	Count	6	8	14	7	35
hypochromic						
	%within	17.1	22.9	40.0	20.0	100.0
	morphological					
	anaemia					
	% within region	7.1	9.5	16.7	8.3	10.4
	% of total	1.8	2.4	4.2	2.1	10.4

Normocytic	Count	78	75	70	67	290
normochromic						
	%within	26.9	25.9	24.1	23.1	100.0
	morphological					
	anaemia					
	% within region	92.9	89.3	83.3	79.8	86.3
	% of total	23.2	22.3	20.8	19.9	86.3
Macrocytic	Count	0	1	0	10	11
normocytic						
	%within	0.0	9.1	0.0	90.9	100.0
	morphological					
	anaemia					
	% within region	0.0	1.2	0.0	11.9	3.3
	% of total	0.0	0.3	0.0	3.0	3.3

Majority of the pregnant women had normocytic normochromic anaemia.

Socio-demographic factors associated with development of anaemia during pregnancy

The association of various factors such as socio-demographic, obstetric and nutritional factors to anaemia during pregnancy in Khomas region was assessed.

The factors associated with anaemia in pregnancy (non-anemic vs anaemic pregnant women) is presented in Table 3.7.

Table 3.7 Factors associated with anaemia in pregnancy

Variables	Categories	Anaemia status						Chi-square
		Non-anaemic		Anaemic		Total		(P-value)
		Count	%	Count	%	Count	%	
Age	Below 25	17	81.0	4	19.0	21	25.0	0.951
	25-35	39	78.0	11	22.0	50	59.5	
	Above 35	10	76.9	3	23.1	13	15.5	
Marital Status	Single	62	78.5	17	21.5	79	94.0	0.936
	Married	4	80.0	1	20.0	5	6.0	
Educational Status	Primary and below	3	37.5	5	62.5	8	9.5	0.003*
	Secondary and above	63	82.9	13	17.1	76	90.5	
Gestational Status	1 st Trimester	43	87.8	6	12.2	49	58.6	0.0031*
	2 nd Trimester	20	69.0	9	31.0	29	34.5	
	3 rd Trimester	3	50.0	3	50.0	6	7.1	

Gravidity	Primigravida	14	77.8	4	22.2	18	21.4	0.926
	Multigravida	52	78.8	14	21.2	66	78.6	
Eating animal products	Daily	37	78.7	10	21.3	47	56.0	0.987
	Weekly	19	79.2	5	20.8	24	28.6	
	Occasionally	10	76.9	3	23.1	13	15.5	
Eating green vegetables	Daily	21	67.7	10	32.3	31	36.9	0.18
	Weekly	39	84.8	7	15.2	46	54.8	
	Occasionally	6	85.7	1	14.3	7	8.3	

Table 3.7 above, illustrates that out of the total variables assessed in this study only educational and gestational statuses were found to be significantly associated with anaemia in pregnancy, with educational level having a p-value of 0.003 and gestational status having a p-value of 0.031.

CHAPTER 4: DISCUSSION AND CONCLUSION

Anaemia is one of the leading causes of complications in pregnancy and could lead to adverse pregnancy outcomes (Lin et al., 2018). It is a public health concern in both developed and developing countries. This study sought to compare the prevalence of anaemia in pregnancy in 4 randomly selected regions of Namibia. In the current study the overall prevalence of anaemia in pregnancy was 20.8%, and according to the WHO classification of anaemia a prevalence between 20% and 39.9% is considered a moderate health problem (Ramesh et al., 2017; Adam et al., 2018). This means that the Namibian government still needs to strengthen measures aimed at reducing anaemia in pregnancy. Namibia is considered an upper middle-income country (World Bank, 2021), which means it has resources to support its population and consequently contributed to the prevalence of anaemia falling under the classification of a moderate health problem. However, despite being an upper middle-income country there is still poverty in Namibia caused by skewed distribution of wealth with a lot of people being poor (Tjitemisa, 2020). Hence, the prevalence of anaemia not lower in this study. The finding on the prevalence is comparable with similar studies conducted in Tanzania (18%) and Pakistan (24%) (Stephen et al., 2018; Zehra et al., 2014). However, the prevalence of anaemia in the current study was higher than 7.4% anaemia prevalence obtained in a study carried out in South Western Uganda among pregnant women attending antenatal care (Okia et al., 2019). The study attributed its lower anaemia prevalence finding to improved maternal healthcare services in the study area due to the United States Agency for International Development (USAID) funded project in Uganda. In addition, the present study's finding on anaemia prevalence was also higher than that of the study carried out in Ethiopia which found the prevalence of 11.6% among pregnant women attending ANC in the governmental health facilities (Gebreweld & Tsegaye, 2018). Earlier commencing of ANC visit might be the probable reason for the lower prevalence rate in Ethiopia.

Moreover, the result of the study was considerably lower than the previous studies done in other African countries; Ghana (41.5%), Sudan (53.0%) and South Africa (42.7%) (Anlaaku & Anto, 2017; Adam et al., 2018; Tunkyi & Moodley, 2016). The possible reason for the lower prevalence of anaemia in the present study, might be due to the administration of iron supplements to pregnant women in most of the state health facilities in Namibia which is a low and cost-effective method to reduce the burden of maternal anaemia (Kanyangarara et al., 2019). Low sociodemographic factors, low obstetric history and poor dietary habits of study participants in the previous studies might be linked to their higher anaemia prevalence rates in comparison to the current study.

Furthermore, the classification of anemia according to the level of severity in the present study was as follows: 0% severe, 4.8 % moderate and 16.1% mild. The zero case of severe form of anaemia reported in this study might be due to the strategies regarding primary health care in Namibia such as: promotion of proper nutrition, promotion of safe drinking water and promotion of maternal and child health services (Christians, 2020), which seems effective and assessable to most of the pregnant women in the regions of the study. However, the finding on anaemia classification in some previous studies had contrasting findings to the ones of the current study (Shitie et al., 2018; Tunkyi and Moodley; Zecharias, 2017; Okia et al., 2019). Geographical differences maybe the possible reason for the varying prevalence in the level of anaemia severity in pregnant women across countries. Additionally, the utilization of various anaemia cut offs in different studies may also contribute to the varied prevalence rates (Abriha et al., 2014). Poor utilization of antenatal care services, lack of knowledge about anaemia, poor adherence to iron supplementation and inability to purchase supplementation during pregnancy in some sub-Saharan- African countries might also be attributed to varied prevalence rates (Djibril et al., 2019).

The prevalence of anaemia in pregnancy across the 4 regions of the study was as follows: Karas (23.8%), Khomas (21.4%), Oshikoto (20.2%) and Omaheke (17.9%). The regional findings were comparable to each other, and this could be due to maternal health education that is made universal across the country. The findings on the prevalence across the regions in Namibia were similar to findings in Jamaica, which found the prevalence across its regions almost the same (South East region 47.0%, North East region 47.9% and Western region 41.8%) (Wright et al., 2017). However, there were no apparent reasons attributed to the anaemia prevalence rates being almost the same across the different regions of Jamaican.

According to the WHO classification of anaemia in pregnancy, Karas, Khomas and Oshikoto regions fall under the moderate health problem (prevalence between 20%-39.9%), and only Omaheke region is considered to have anaemia in pregnancy as a mild health problem (prevalence between 5%-19.9%) (Ramesh et al., 2017; Adam et al., 2018). Majority of the population in Omaheke region have irrigated household gardens which provide them with nutritious food, and might be the contributing factor to the lower anaemia prevalence rate in comparison to the other regions of the study (Iileka, 2020). Furthermore, no severe form of anaemia was found in any of the regions of study. However, the intriguing find was that each region equally contributed to the moderate form of anaemia (4.8%) in the country which meant that each region had a prevalence of 1.2% moderate anaemia. However, this is not a common finding whereby each region of study in the country equally contribute to the prevalence of moderate anemia. Previous studies done in other parts of the world had comparable results to the current study's result on severe form of anaemia in pregnancy (Tunkyi & Moodley, 2016; Tibambuya et al., 2019). Moreover, the prevalence of mild anaemia per region is as follows; Omaheke (13.1%), Oshikoto (15.5%), Khomas (16.7%) and Karas (19%). The variations in the prevalence of mild anaemia across the different regions of Namibia may be linked to the different level of poverty in the different regions.

Iron deficiency anaemia (IDA)

The prevalence of IDA among pregnant women in this study was 11.9%. The lower IDA prevalence rate in the current study might be linked to the provision of free antenatal care services in the state healthcare centers, clinics and hospitals, and the provision of food aid to the low-income families across the country (MoHSS, 2020). The prevalence of IDA obtained is almost similar to 12.3% prevalence found in a study conducted in Lagos, Nigeria on the prevalence and foetomaternal effects of IDA among pregnant women (Ajepe et al., 2020). The study's finding was also comparable to study done in Sudan which found a prevalence of IDA (13.6%) among pregnant women (Adam et al., 2018). The prevalence of IDA recorded in the current study was much lower than that obtained by Abu et al. (2016), who found that the prevalence of IDA among Egyptian pregnant women was 52.35%.

Additionally, 25.6% of the pregnant women in this study had depleted iron stores (iron deficiency). This is lower than 30-40% of the pregnant women estimated by WHO to have depleted iron stores (Loy et al., 2019). The finding was inconsistent with the study by Srour et al. (2018), who found more than half (52.0%) of the pregnant women with depleted iron stores. The variation in iron deficiency prevalence rates from the current study to that of the previous study might be linked the free supply of iron supplements to pregnant women in Namibia, which increases iron stores (Georgieff et al., 2019). Iron demands increases for pregnant women compared with nonpregnant women, and if there is no iron supplementation, the iron ingested from food sources is not adequate to meet their needs (Alene & Dohe, 2014).

The prevalence rates of IDA were almost similar across the regions of study, from the lowest in Karas (9.5%), Oshikoto (10.7%), and Khomas (13.1%) to the highest in Omaheke (14.3%). The study's findings across four regions, was contrary to a previous study carried out by Tan

et al, (2020), that found IDA prevalence varied substantially among its six regions of study. Dietary habits among the pregnant women who participated in the present study, and those who participated in the previous study might play crucial roles in the diverse distribution of IDA across the different regions of the studies. Dietary habit is the key in preventing IDA in pregnancy (Abu-Ouf et al., 2015). A well-balanced diet goes hand in hand with iron rich foods and has proven to increase the iron status of pregnant women and gradually providing better pregnancy outcomes (Banjari, 2018). Therefore, dietary habits play an important role in the diverse distribution of IDA.

Vitamin B12 deficiency anaemia

The prevalence of vitamin B12 deficiency anaemia in the present study was 5.56%. The lower prevalence rate of Vitamin B12 deficiency anaemia among pregnant women reported in the current study is similar to a previous study done in South-India among rural pregnant women with a prevalence of 11.7% (Barney et al., 2020). Malnutrition, mediated malabsorption of vitamin B12 and vegetarianism are some of the main contributing factors to vitamin B12 deficiency anaemia (Sukumar et al., 2016). Malnutrition, mediated malabsorption of vitamin B12 and vegetarianism appears to not be a big concern among pregnant women in Namibia, which might be the contributing factor to the lower prevalence rate of vitamin B12 deficiency anaemia in the current study.

The finding of the prevalence of vitamin B12 deficiency anaemia was lower than the previous study done by Karaoglu et al. (2010), which obtained 34.5% vitamin B12 deficiency anaemia among pregnant women. The varied prevalence rates may be due to different sample sizes and study settings used in the current study to the previous study.

Furthermore, the prevalence of vitamin B12 deficiency in the present study was recorded as 7.58%. The finding contrasted that of a 25% prevalence of vitamin B12 deficiency found among pregnant women in a study by Sukumar et al, (2016), differences in dietary consumption of pregnant women from the different studies might have attributed to the varied prevalence rates.

The prevalence of vitamin B12 deficiency anaemia varied across the 4 regions of study, with zero prevalence reported in Khomas region, Oshikoto region (5.97%), Karas region (10.00 %) and Omaheke region (33.33%). Accessibility to retailer shops might be the attributing factor to the varied prevalence rates across the different regions. Pregnant women leaving in Khomas region (urban area) might be more accessible to retailer shops enabling them to purchase nutritional food on a regular basis when compared to those leaving in Omaheke region (semi-rural) who might be staying far from retailer shops, making it difficult to purchase nutritional food on a regular basis. Thus, a higher prevalence of vitamin B12 deficiency anaemia in Omaheke region in comparison to Khomas region.

Morphological anaemia

Out of 20.8% anaemic pregnant women, 10.4% had microcytic hypochromic anaemia, 86.3% had normocytic normochromic anaemia and 3.3% had macrocytic normocytic anaemia. Macrocytic normocytic anaemia was the least morphological anaemia probably due to the low proportions of vitamin B12 deficiency among pregnant women of the study. Vitamin B12 deficiency is one of the main causes of macrocytic anaemia (Karaoglu et al., 2010). The finding of the current study on morphological anaemia in pregnancy correlated with findings of a previous study that had majority of the pregnant women who participated in the study with normocytic normochromic anaemia (56.5%), followed by microcytic hypochromic

anaemia (38.1%) and minority of the participants had macrocytic normocytic anaemia (0.9%) (Karaoglu et al., 2010). However, the findings of the study were contrary to a study by Okia et al. (2019), which found majority of the pregnant women with microcytic hypochromic anaemia (50.0%), followed by those with macrocytic normocytic anaemia (41.7%) and the least prevalence was noted in the pregnant women with normocytic normochromic anaemia (8.3%). The study attributed its variations of morphological anaemia to the socio-economic differences and dietary variations of its participants.

4.1 Factors associated with anaemia in pregnancy

In the multiple logistic regression, no significant association was found between age, marital status, gravidity, income level, the frequency of animal products consumption and green vegetables consumption (p values >0.05). A smaller sample size used in the current study might be the possible factor to most of the variables being not significantly associated with anaemia. However, majority of these variables (age, marital status, income level) were also not found to be significantly associated with anaemia in a study by Alene and Dohe (2014). The study concluded that differences between sociocultural and behavioral characteristics of its participants might be the main contributing factors to the lack of association between the variables and anaemia. Contrary, similar studies done elsewhere had significant association between age, marital status, gravidity, income level, frequency of animal product consumption and green vegetable consumption, partly due to larger sample size used (Abriha et al., 2014; Anlaakuu & Anto, 2017; Bansal et al., 2020).

The only association found by the current study was on education level and gestational status. Pregnant women with primary and below level of education were found 17 times more likely to be anaemic in comparison to those with secondary and above level of education. This

finding meant that women with lower education level, might have poor knowledge on the proper nutrition required during pregnancy. Additionally, women with secondary and above level of education are more likely to have a better income enabling them to afford purchasing nutritious food and hence, less likely to develop nutritional anaemia. The finding is comparable to previous studies which found majority of their participants with below secondary level of education being at a higher risk of anaemia than those with higher level of education (Fondjo et al., 2020; Stephen et al., 2018). Moreover, pregnant women in their second and third trimesters were 31 and 50 times respectively, more likely to develop anaemia than those in their first trimester. During pregnancy, there is a physiologically plasma volume increase by 25–80% of pre-pregnancy volumes between the second trimester and the middle of the third trimester of pregnancy which causes a decrease in the Hb levels (Lin et al., 2018). Additionally, at the time of pregnancy the calorie and nutrients requirements peak up to aid the increased maternal metabolism, blood volume and the supply of nutrients to the fetus (Ahenkorah et al., 2016; Alene & Dohe, 2014). These demands increase mostly during the first and second trimesters of pregnancy (Zehra et al., 2014).

Despite the lack of association on most of the current study's variables which is likely caused by the smaller sample size 84 (pregnant women) used. The incidence of anaemia in pregnancy has been reported to increase as gravidity increases, while expected to decrease as the age and income level increase. As the number of pregnancies increase (multiparous), there is a repeated drain on the iron reserves which increases the risk of the pregnant women to be anaemic (Asrie, 2017). Moreover, younger pregnant women belong to the physically active group and most are still undergoing rapid growth which increases their nutritional requirements, predisposing them to anaemia (Fondjo et al., 2020). In addition, women with higher income level are more likely to afford diets rich in nutrients essential in the reduction

of anaemia. Thus, as age and income increase the likelihood of being anaemic ought to decrease, while it is likely to increase with increasing gravidity.

4.2 Conclusion

In the current study there was a moderate problem of anaemia among pregnant women in Namibia (20.8%). This means that the government still needs to strengthen measures aimed at reducing anaemia in pregnancy. Additionally, IDA (11.9%) and vitamin B12 deficiency anaemia (5.56%) were considered as a mild health problem, and the most frequent morphological anaemia was normocytic normochromic anaemia (86.3%). Moreover, the prevalence of anaemia in pregnancy across the 4 regions of study; Karas (23.8%), Khomas (21.4%), Oshikoto (20.2%) and Omaheke (17.9%) were comparable to each other. Apart from Omaheke region's anaemia prevalence rate which falls under mild health problem, the other 3 regions' prevalence rate is classified as a moderate health problem.

The prevalence rates of IDA were almost similar across the regions of study with Karas region reporting the lowest prevalence rate (9.5%). Furthermore, the study found no case of vitamin B12 deficiency in Khomas and Oshikoto regions in anaemic pregnant women. In addition, educational and gestational statuses were the only factors found significantly associated with anaemia in pregnancy (p-value of <0.05).

4.3 Limitations and Recommendations

Due to Covid-19 restrictions during the data collection procedure, the researcher could not travel to all 4 regions of the study in order to collect socio-demographic information from the study participants. Socio-demographic information was only collected from Khomas

region. Thus, the inference on the association between anaemia and socio-demographic factors could not be made for other regions but only for Khomas region. Additionally, due to lack of funds toward this project other classification of anaemia could not be assessed. Further studies exploring and classifying other types of anaemia such as; thalassemia, anaemia of chronic disease, haemolytic anaemia, pernicious anaemia, sickle cell anaemia and aplastic anaemia in pregnancy are recommended. Having such information will enable the government and other stakeholders to implement effective measures aimed at reducing infant and maternal mortality associated with anaemia in pregnancy. Further studies classifying other types of anaemia in pregnancy will also provide information to health care workers about the prevalent anaemia type in their region for the better management and treatment of their patients. Additionally, assessing majority of the different types of anaemia could have contributed significantly to the study's finding in determining whether they are a cause of concern among pregnant women in the regions of study.

Moreover, pregnant women should be encouraged to access antenatal care earlier in pregnancy to enable earlier detection and correction of anaemia, and in order to minimize fetal and maternal complications associated with anaemia in pregnancy. Funding towards projects of this kind should be prioritized to enable assessment of majority of the anaemia types.

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APPENDICES

APPENDIX A: Ethical clearance by the Faculty of Health and Applied sciences



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FACULTY OF HEALTH AND APPLIED SCIENCES RESEARCH ETHICS COMMITTEE (FHAS-REC)

DECISION/FEEDBACK ON RESEARCH PROPOSAL ETHICAL CLEARANCE

Dear: Ms

Emma Shaduka

Research Topic:	An investigation into the Aetiology of Anaemia in pregnant women in Namibia.
Supervisor	Dr Munyaradzi Mukesi
Qualification registered for:	Master in Health Sciences

Re: Ethical Screening Application No:

FHAS: 21/2019

The Faculty of Health and Applied

Sciences Research Ethics Screening Committee has reviewed your application for the above-mentioned research project. Based on the recommendation of the expert reviewer, the research as set out in the application is hereby:

(Indicate with an X)

3.	No Ethical issues noted	Nil
Approved: i.e. may proceed with the project		X
Approved provisionally: i.e. may proceed but subject to compliance with recommendation(s) listed below		
Not approved: Not to proceed with the project until compliance with recommendation listed below and resubmit ethics application for consideration		

It is important to note that as a researcher, you are expected to maintain ethical integrity of your research. You are encouraged to strictly adhere to the research ethics policy of NUST. You should remain within the scope of your research proposal and support evidence as submitted to the FHASREC. Should any aspect of your research change from the information as presented, which

Ethical issues that require compliance/ must be addressed : None		
No.	Ethical issues	Comment/recommendation
1.	No Ethical issues noted	Nil
2.	No Ethical issues noted	Nil

could have an impact or effect on any research participants/subjects/environment, you are to report this immediately to your supervisor and to the FHAS-REC as applicable in writing. Failure to do so may result in withdrawal of approval. Kindly consult the committee if you need further clarification in this regard.

We wish you success in your research endeavor and are of the belief that it will have positive impact on your career as well as the development of NUST and the society in general.

Sincerely,

A handwritten signature in black ink, appearing to be 'S. R. Moyo', with a long, sweeping horizontal line above it.

Name: Prof Sylvester R Moyo

Signature:

Date:

25/10/2019

Chairperson: FHAS Ethics Screening Committee.



REPUBLIC OF NAMIBIA

Ministry of Health and Social Services

Private Bag 13198
Windhoek
Namibia

Ministerial Building
Harvey Street
Windhoek

Tel: 061 – 203 2507
Fax: 061 – 222558
E-mail: itashipu87@gmail.com

OFFICE OF THE EXECUTIVE DIRECTOR

Ref: 17/3/3 ES
Enquiries: Mr. A. Shipanga

Date: 12 February 2020

Ms. Emma Shaduka
PO Box 62200
Katutura

Dear Ms. Shaduka

Re: An investigation into the Aetiology of Anaemia in pregnant women in Namibia.

1. Reference is made to your application to conduct the above-mentioned study.
2. The proposal has been evaluated and found to have merit.
3. **Kindly be informed that permission to conduct the study has been granted under the following conditions:**
 - 3.1 The data to be collected must only be used for academic purpose;
 - 3.2 No other data should be collected other than the data stated in the proposal;
 - 3.3 Stipulated ethical considerations in the protocol related to the protection of Human Subjects should be observed and adhered to, any violation thereof will lead to termination of the study at any stage;

- 3.4 A quarterly report to be submitted to the Ministry's Research Unit;
 - 3.5 Preliminary findings to be submitted upon completion of the study;
 - 3.6 Final report to be submitted upon completion of the study;
 - 3.7 Separate permission should be sought from the Ministry for the publication of the findings.
4. All the cost implications that will result from this study will be the responsibility of the applicant and **not** of the MoHSS.

Yours sincerely,

BEN NANGOMBE
EXECUTIVE DIRECTOR





OFFICE OF THE CHIEF OPERATIONS OFFICER

Enquiries: Mr Boniface Makumbi; Tel.: 061-295 4210

Date: 03 August 2020

Ms Emma Shaduka
Namibia University of Science and Technology
Windhoek
Namibia

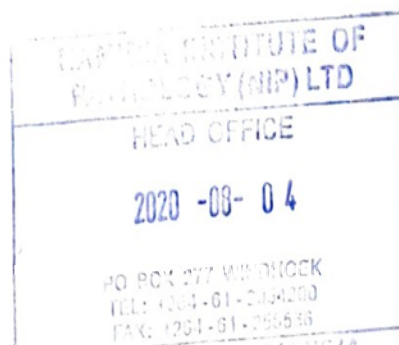
Dear Ms Shaduka

RE: AN INVESTIGATION INTO THE AETIOLOGY OF ANAEMIA IN PREGNANT WOMEN IN NAMIBIA

1. The above mentioned research proposal has been perused and found beneficial to the country.
2. After the review, it is a pleasure to inform you that approval was granted for you to proceed with the research on condition that the following be complied with:
3. Consultation with the relevant Heads of the Business Units starting upon with your research.
4. For the required data, a request in writing should be sent to the ICT department, and you shall expect to receive a response in two weeks.
5. Observe and adhere to all ethical considerations and confidentiality to protect your clientele information.
6. Report to the office of the Senior Manager: Specialised Services upon starting your research.
7. Final report to be shared with the Namibia Institute of Pathology Limited (NIP) and the Ministry of Health and Social Services.

Yours Sincerely

Boniface Makumbi
Acting Chief Operations Officer



APPENDIX D: DATA COLLECTION TOOL

QUESTIONNAIRE

Study Title: An Investigation into the Aetiology of Anaemia in Pregnant women in Namibia

Date:

Requisition No:

NB: Please **tick** in the appropriate **shaded** column.

SOCIO DEMOGRAPHIC INFORMATION

AGE:

> 25	
25-35	
< 35	
Single	
Married	
Primary and below	
Secondary and above	

MARITAL STATUS:

EDUCATIONAL STATUS:

OBTETRIC INFORMATION

GESTATIONAL STATUS:

1 ST Trimester	
2 nd Trimester	
3 rd Trimester	

NUTRITIONAL INFORMATION

EATING ANIMAL PRODUCTS:

Daily	
Weekly	
Occasionally	

EATING GREEN VEGATABLES:

Daily	
Weekly	
Occasionally	

APPENDIX E: CONSENT FORM

Research Title: An Investigation into the Aetiology of Anaemia in Pregnant women in Namibia

Investigator: Emma N Shaduka

Institution: Namibia University of Science and Technology (NUST)

Email: shadukae@gmail.com

Purpose

This study is being carried out with the purpose of acquiring knowledge on the aetiology associated with anaemia in pregnancy, to identify the prevalent anaemia and the socio-demographics linked to its development at the time of pregnancy.

Procedure

1. The study will be conducted over a period of 1 year.
2. Participants will include pregnant women aged 18 and above, within their first trimester.
3. A questionnaire will be used to collect sociodemographic, obstetric and nutritional information.
4. Participants who consent to partake in the study, will be required to complete a consent form.
5. A laboratory requisition number will be allocated to each individual participant to protect their identity.
6. All data will be stored onto a protected personal device.

Risks and discomforts

There are no foreseeable risks for partaking in this study. However, if participants feel uncomfortable to continue with the study, they are free to withdraw.

Benefits

The study will contribute valuable knowledge to aid in the management of anaemia in order to improve maternal and fetal health outcomes.

Consent

I have read and understood the provided information and had the opportunity to ask questions for clarity. I voluntarily agree to participate in this research.

Participant's signature

Date.....

Investigator's signature

Date.....