EXAMINING THE RISK FACTORS FOR HEPATITIS B INFECTION AMONG PREGNANT WOMEN ATTENDING ANTENATAL CARE IN KUNENE REGION,

A CASE CONTROL STUDY

A THESIS SUBMITTED IN PARTIAL FULLFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF SCIENCE IN APPLIED FIELD EPIDEMIOLOGY

AT

THE UNIVERSITY OF NAMIBIA

BY

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APRIL 2018

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ABSTRACT

The Hepatitis B is a viral infection caused by Hepatitis B virus (HBV) which is a double stranded DNA virus, a member of the Hepadnavidae family of viruses. World Health Organization estimates that about 257 million people are living with Hepatitis B virus infection. Namibia has a high prevalence of Hepatitis B infection (9%) among pregnant women and Kunene region prevalence of 8%.

The researcher conducted an un-matched 2:1 case-control study to determine the associated risk factors for Hepatitis B infection among pregnant women in Kunene region. Cases were study subjects with reactive results for HBsAg or HBeAg and controls were study subjects with negative for both HBV markers. A total of 115 cases and 230 controls were interviewed. Mean age among the cases was 29 years range 16 – 45 (SD = 6.6), controls the mean was 26 years range 13 – 45 years (SD = 6.8). Bi-variate analysis was conducted to determine the odds ratios at 95% confidence level. Significant risk factors at p-value less than 0.05 were retained in multiple logistic regression models to determine significant associations.

The multivariate analysis found that polygamous marriages (AO: 3.45; CI: 1.25 – 9.57; p= 0.02). Body piercing and scarification (AOR: 4.34; CI: 2.30 – 8.17; p= 0.00), body tattoos (AOR: 2.95; CI: 1.09 - 7.99; p = 0.03), history of abortion (AOR: 2.91; CI: 1.38 – 6.16; p= 0.00), STI’s (AOR: 3.34; 95%CI: 1.92 – 5.80; p= 0.00) and previous history tooth extraction or any dental procedures (AOR: 2.03; 95% CI: 1.17 – 3.54; p = 0.01) was significantly associated with Hepatitis B infection. Gravidity, parity, HIV positive status and history of blood transfusion were not associated risk factor in multivariate model (p = >0.05). The Ministry of Health and Social Services in Kunene region should implement preventative strategies such as Hepatitis B screening, treatment, health education, infection control and hepatitis B vaccination for the general population.
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<tbody>
<tr>
<td>ALT</td>
<td>Alanine Amino-Transferase</td>
</tr>
<tr>
<td>ANC</td>
<td>Antenatal Care</td>
</tr>
<tr>
<td>AOR</td>
<td>Adjusted Odds Ratio</td>
</tr>
<tr>
<td>ART</td>
<td>Anti-Retroviral Treatment</td>
</tr>
<tr>
<td>CccDNA</td>
<td>Covalently Closed Circular DNA</td>
</tr>
<tr>
<td>CDC</td>
<td>Center of Disease Control</td>
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<tr>
<td>CHB</td>
<td>Chronic Hepatitis B</td>
</tr>
<tr>
<td>COR</td>
<td>Crude Odds Ratios</td>
</tr>
<tr>
<td>DNA</td>
<td>Deoxyribonucleic Acid</td>
</tr>
<tr>
<td>ELISA</td>
<td>Enzyme Linked Immunosorbent assay</td>
</tr>
<tr>
<td>HCC</td>
<td>Hepatocellular Carcinoma</td>
</tr>
<tr>
<td>HBV</td>
<td>Hepatitis B Virus</td>
</tr>
<tr>
<td>HCV</td>
<td>Hepatitis C Virus</td>
</tr>
<tr>
<td>HBsAg</td>
<td>Hepatitis B Surface Antigen</td>
</tr>
<tr>
<td>HBeAg</td>
<td>Hepatitis B Envelope Antigen</td>
</tr>
<tr>
<td>HBcAg</td>
<td>Hepatitis B Core Antigen</td>
</tr>
<tr>
<td>HBIG</td>
<td>Hepatitis B Immunoglobulin</td>
</tr>
<tr>
<td>mRNA</td>
<td>Messenger RNA</td>
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<tr>
<td>MCH</td>
<td>Mother Child Health</td>
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<tr>
<td>PEG-IFN</td>
<td>Pegylated Interferon</td>
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<tr>
<td>rcDNA</td>
<td>Relaxed Circular HBV DNA</td>
</tr>
<tr>
<td>RNA</td>
<td>Ribonucleic Acid</td>
</tr>
<tr>
<td>Acronym</td>
<td>Description</td>
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<tr>
<td>---------</td>
<td>------------------------------------</td>
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<tr>
<td>STI</td>
<td>Sexually Transmitted Infections</td>
</tr>
<tr>
<td>WHO</td>
<td>World Health Organization</td>
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</table>
ACKNOWLEDGEMENT

Firstly I would like to express my gratitude to my Heavenly Father who brought me this far through His grace and strength.

I wish to express my sincere gratitude to the following people and institutions:

The Center for Disease Control and Prevention (CDC) and Global Fund Namibia in collaboration with the Ministry of Health and Social Services (MoHSS) for funding this thesis and all other costs including my tuition fees for two years. I greatly appreciate the support from Kunene Regional Health Directorate for allowing me to carry out my research in the selected health facilities.

I wish to extend my deepest gratitude to my supervisor, Dr. Jacob Sheehama and co-supervisor, Mrs. Ester Mulenga for their sustained guidance and critical review throughout the process and execution of this research. I humbly thank the management team and administrators at the University of Namibia for allowing me to study at their institution.

My sincere gratitude goes to Dr. Mensah Kofi Nyarko (Namibia FELTP advisor) for laying a good foundation for Field Epidemiology and Laboratory Training program in Namibia, for his patient guidance, advice and encouragement throughout the my studies.

Special thanks to Mr. Suleiman Souwadogo (CDC Technical Advisor for Laboratory Services) for guiding and mentoring me meticulously throughout my research even during his busy schedule. I would also like to give special thanks to Drew Baughman (CDC, Biostatistician) for his statistical assistance.

Sincere gratitude goes to the Epidemiology department (MoHSS) for their continuous support during my studies. Thanks to the Kunene regional director Mr. Thomas Shapumba and Regional Management Team for their support and good hospitality during our field placement.
To all health workers who assisted me during the data collection procedure, I thank you all and this project will not have succeeded without your effort.

May the Almighty God bless you all.
DEDICATIONS

This study is dedicated to my dear parents; Billy Mwaningange and Ndmonoghenda Shikongeni Mwaningange who gave me solid foundation, support and wisdom. I would also want to dedicate this project to my lovely daughter Loide Tuhafeni Haimbodi. Let this study be the source of inspiration for all your academic and future career endeavors.

Lastly, I dedicate this study to the late Governor of Kunene Region, Ms. Angelika Muharukua who sadly passed on towards the end of this project. Let this study contribute towards her legacy of improving maternal and child health care in Kunene region.
DECLARATION

I, Iyaloo Wilkka Mwaningange, hereby declare that this study is a true reflection of my own research, and that this work, or part thereof has not been submitted for a degree at any other institution of higher education. No part of this thesis may be reproduced, stored in a retrieval system, or transmitted in any form, or by any means (e.g. electronic, mechanical, photocopying, recording, or otherwise) without prior permission of the author, or the University of Namibia on her behalf.

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Iyaloo Wilika Mwaningange                          Date
CHAPTER ONE

1. INRODUCTION

1.1 BACKGROUND OF THE STUDY

Hepatitis B Virus (HBV) is a double-stranded DNA virus belonging to the Hepadnavidae family which is a potentially life-threatening liver disease (1). The acute form of Hepatitis B is detected by serological positive Hepatitis B Surface Antigen marker (HBsAg) while chronic hepatitis have positive serology test for Hepatitis Envelope Antigen marker (HBeAg) (2). The World Health Organization estimates that about 257 million people are living with Hepatitis B virus infection, and these are positive for hepatitis B surface antigen (3). Furthermore in 2016, Hepatitis B resulted in 887 000 deaths, mostly from complications such as cirrhosis and hepatocellular carcinoma (4).

In terms of geographical distribution, the Western Pacific has 95 million and Africa has 75 million cases and these geographical areas have large number of people who live with chronic hepatitis B infection (3). The HBV regional prevalence variations are categorized as low prevalence (less than 2%) regions which include North America, New Zealand and Western Europe. The intermediate endemic regions include such areas as Eastern Mediterranean, Middle East and Eastern Europe which have a prevalence of 2 – 4.99 percent. The higher intermediate endemic regions have prevalence of 5 - 7.99 percent and encompass North African and Central Asian countries. The high endemic region includes Sub-Saharan Africa, China and Western Pacific Region that have prevalence of more than 8 percent (5). A study by Adam on Hepatitis B
infection among pregnant women in Africa revealed the prevalence rates to be as low as 3.3% in Zimbabwe and as high as 13% in Nigeria and Malawi (6).

Perinatal and early childhood transmission are common routes of HBV(7). The World Health Organization estimates that About 5% of women worldwide are carriers of chronic HBV infection (4). The unpublished study by Mhata and colleagues on distribution of Hepatitis B in Namibia estimated a 11.8% prevalence rate of Hepatitis B infection in Namibian general population(8). The same study found out that Kunene Region ranked among top five (12%) on prevalence of Hepatitis B in the general population (8).

Perinatal and early childhood transmission are common routes of HBV infection in high and intermediate endemic areas while in low endemic areas, unprotected sexual intercourse and use of contaminated needles were found to be common route of transmission (7). In Namibia, Hepatitis B prevalence in pregnant women was found to be 9%, and Kunene region had 8 percent prevalence among pregnant women (8),(9). Although neighboring South Africa has similar prevalence of 8% in pregnant women (10), due to the variations in population characteristics, generalizing the findings of this study to Kunene regions may not be representative enough. In addition, the predominant Himba nomadic indigenous tribe that lives in Kunene region, has a unique population dynamics and ancient traditional practices such as child and polygamous marriages which may have influence on transmission of various communicable diseases (11). It is against this background that this study was undertaken to establish the significant risk factors for Hepatitis B infection in pregnant women by reviewing the literature on the subject matter and critical analysis of the collected data.
1.2 PROBLEM STATEMENT

The first problem is that Namibia has a high prevalence (9%) of Hepatitis B infection among pregnant women and Kunene region where the study was undertaken has an estimated hepatitis B infection prevalence of 8% among pregnant women. Hepatitis B infection is associated with unfavorable pregnancy outcomes and complications such as preterm delivery, gestational diabetes mellitus, placenta praevia etc. and Namibia has a high number of infant maternal deaths and these maybe due to these high hepatitis B infection ratios. The second problem is that the majority of the population in Kunene region is Himba and these by virtue of their nomadic and strong cultural ties have traditions and practices that put pregnant women at high risk of acquiring Hepatitis B infection. Practices as child and polygamous marriages as well as lack of education, access to health care and modern civilization are all at risk factors that predispose pregnant women to Hepatitis B infection. Therefore one has to understand and identify significant risk factors for Hepatitis B infection in pregnant women and develop the targeted control strategies.

1.3 THE PURPOSE OF THE STUDY

The purpose of the study entailed examining the risk factors associated with acute (HBsAg) and chronic (HBeAg) HBV infection among pregnant women attending antenatal care in Kunene region.

1.4 OBJECTIVES

This study aimed at achieving following objectives:

- To determine socio demographic factors which are associated with HBV infection in pregnant women attending ANC in Kunene Region.
- To identify the potential risk factors which are associated with HBV infection in pregnant women attending ANC in Kunene Region?

### 1.5 STUDY HYPOTHESIS

#### Table 1.5 Hypotheses

<table>
<thead>
<tr>
<th>Research objectives</th>
<th>Null hypotheses [H0]</th>
<th>Alternative hypotheses [HA]</th>
</tr>
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<tbody>
<tr>
<td>To determine the socio demographic and potential risk factors for Hepatitis B</td>
<td>There will be no observed statistical significance</td>
<td>There will be observed statistical significance</td>
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<tr>
<td>infection in pregnant women attending antenatal care</td>
<td></td>
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### 1.6 SIGNIFICANCE OF THE STUDY

The epidemiological data that was derived from this study may be essential to program managers, health planners, and clinicians for resource mobilization, priority setting and utilization in management of Hepatitis B infection. The generated information may be relevant for monitoring the impact of recently introduced Hepatitis B vaccination and screening packages in antenatal care clinics in Kunene Region and country at large. This study is an immense contributor of additional knowledge on the epidemiology and control of HBV infection in Namibia. The scarcity of recent studies on this subject in the Namibian context is a pointer of current knowledge gap.
1.7 STUDY LIMITATIONS

The limited logistical challenges could not permit the collection of blood specimen for HBV for the purpose of a study. The study enrolled the subjects based on the verified documented laboratory results for Hepatitis B from the client’s ANC records instead. Consequently, pregnant women who did not attend ANC during the study period were given opportunity to participate in the study. Although the initial proposal included Omusati region, the proposed population could not be included in this study as Hepatitis screening at ANC clinics in that region is not routinely done.

1.8 DELIMITATION

The study focused on the risk of Hepatitis B infection among pregnant women attending Antenatal Care in nine health facilities in Kunene region. This region is home to marginalize Ovahimba, Ovatwe and Ovadhemba ethnic groups. Due to the unique traditional practices among these ethnic groups, the findings of this study are likely to be different from the regions which do not have these communities. In addition, the study population was limited to pregnant women that were screened for viral hepatitis B infection during pregnancy as it is closely related to high risks of maternal complications (12). Therefore by limiting the study for Hepatitis B in pregnant women in this region, targeted public health interventions becomes possible.

1.9 SUMMARY

This chapter covered the general overview of Hepatitis B infection and the global distribution of disease. The section also covered the global, regional and national prevalence of Hepatitis B infection in pregnant women. The implication of Hepatitis B on child and maternal health was emphasized in the problem statements. The research objectives and hypothesis were also
discussed in this chapter. The significance of the study, its limitations and delimitations were explained in this chapter.
CHAPTER TWO

2. LITERATURE REVIEW

2.1 INTRODUCTION

This chapter is dedicated to avail literature on Hepatitis B infection among pregnant women. The section aims to dissect the available information on this subject matter. Literature review provides an overview of the research in the graduate student’s area of interest and it often highlight recent findings and gaps in the literature (13). The chapter covers the scientific and epidemic background of hepatitis B in pregnant women. This chapter covers the epidemiology, pathogenesis, risk factors and interventions for hepatitis B infection in pregnant women.

2.2 OVERVIEW ON HEPATITIS B INFECTION

Hepatitis B is a viral infection is caused by Hepatitis B virus (HBV) which is a double stranded DNA virus, a species of the genus Orthohepadnavirus and a member of the Hepadnavidae family of viruses (14). The virus was first identified in 1965 among the Aboriginal people of Australia by Bloomberg (2). The virus is transmitted by contact of infected body fluids such as saliva, blood, semen and vaginal fluid (7). The common routes of transmission are vertical transmission from infected mother to the newborn child, unprotected sexual intercourse, and contact with contaminated instruments and perinatal transmission is responsible for 60 – 90% of all routes of transmissions (15). In pregnancy hepatitis B infection can have negative impact on maternal and child health. In order to understand the cause of hepatitis B infection in pregnancy, one has to identify and analyze the risk factors. This chapter examines the theoretical framework as well as the literature on risk factors for hepatitis B infection in pregnant women.
2.2.1 Epidemiology

Hepatitis B infection is a leading cause of morbidity and mortality of liver related ailments (7). The World Health Organization estimated that about 2 billion people are living with hepatitis B infection and about 360 million people die from liver cirrhosis which is caused by hepatitis B infection (16). The global prevalence of chronic hepatitis B (CHB) in 2015 was estimated to be at 3.7%, of which 3.9% was among males and 3.5% among females (17). In contrast by 1990, the total number of HBsAg positive individuals was estimated at 223 million and the prevalence was 4.2%(18). The HBsAg endemicity is categorized as low (<2%), low-intermediate (2–4.9%), high intermediate (5–7.9%) and high (>8%). Western Sub-Saharan Africa region is the area with the highest HBV endemicity while South, Central and East sub-Saharan Africa, Central and East Asia represent regions of high-intermediate endemicity (5).

In terms of disease mortality, the annual number of deaths from HBV is estimated 786,000, compared to 499,000 due to HCV (total viral hepatitis 1.29 million), 1.47 million due to HIV, 1.17 million due to malaria and 1.2 million due to tuberculosis (19). Based on these figures, Hepatitis B was ranked 15th and HCV 25th among the total causes of death. Viral hepatitis (A, B, C, D, and E) resulted in 1.44 million deaths and was ranked 8th as a cause of human mortality, while HIV ranked sixth. The deaths from cirrhosis and HCC were estimated to be at 1.03 million and 750,000 respectively (19).

The prevalence of Hepatitis B infection among pregnant women has been studied in intermediate and high endemic countries. The study in Ethiopia and Kenya revealed that Hepatitis B prevalence was above 14% among pregnant women, while the study in Iran found out a 2% HBV prevalence among pregnant women (20), (21). The study by the World Health Organization estimates that about 5% of pregnant women in high endemic areas are carriers of Hepatitis B
infection (22). In low endemic countries where Hepatitis prevalence is less than 2%, the infection is more prevalent in other key populations such as intravenous drug users rather than among pregnant women. According to Centre for Diseases Control, Hepatitis B infection among pregnant women in United State of America was 1.5%, however the prevalence among pregnant immigrants of nationality from high endemic countries ranged between 7 – 10 percent respectively (23). This is the indication that even in low endemic countries, there are key populations with pregnant women who are having higher proportions of Hepatitis B infection.

World Health Organization estimates that 90% of pregnant women with hepatitis B infection live in Sub- Saharan Africa, China and South Pacific(16). The prevalence of Hepatitis B in Sub-Saharan Africa ranges from 3% in Zimbabwe to 13% in Nigeria and Mozambique (6). The study conducted in Cameroon revealed an 8% prevalence among pregnant women attending antenatal care and these findings concurs with the findings of the Nigerian and Kenyan studies (24). Southern African countries like Zimbabwe and South Africa have intermediate prevalence of three and six percent respectively among pregnant women (6). According to a situational analysis by Mhata and colleagues, the prevalence of Hepatitis B infection among pregnant women in Namibia was found to be 9.2 percent (8) and of interest is Kunene region with an estimated prevalence between 6 – 8% (9). Additional studies are required in order to understand the cause of high prevalence of Hepatitis B infection among pregnant women in both regions and Namibia at large.

2.2.2 Genotyping

In terms of molecular epidemiology five HBV genotypes are more frequently detected in Africa, A, B, C, D and E (25). Zampino and colleagues suggest that genotype A is predominant in southern and eastern Africa, genotype D in northern Africa and genotype E in the vast region
from Senegal to Namibia and eastward to the Central African Republic (25). According to global estimation of Hepatitis B, Namibia has estimated prevalence 8.6% which indicates that it is high endemic area (17).

2.2.3 Morphology of Hepatitis B Virus
Hepatitis B Virus Hepatitis B virus (HBV) is a double-stranded Deoxyribonucleic Acid(DNA) virus belonging to the Hepadnavidae family that can develop into a potentially life-threatening liver diseases (7).

![Graphical Image of Hepatitis B Virus](image)

The figure 2.2.3 is illustrating the physical structure of HBV. The infectious Hepatitis B virion, known as the Dane particle, is approximately 42 nm in size and is composed of an outer lipid envelope containing viral glycoproteins as well as an inner nucleocapsid (15). The viral glycoproteins within the lipid envelope of the virion constitute the Hepatitis B surface antigen (HBsAg). The nucleocapsid is composed of hepatitis B core antigen (HBcAg) and encloses a copy of double-stranded circular HBV DNA and the HBV DNA polymerase (26).

Figure 2.2.3 Graphical Image of Hepatitis B Virus Source:(2)
2.2.4 Life Cycle of Hepatitis B Virus

The HBV life cycle begins with binding of the hepatitis B virion to hepatocytes through interactions between cell surface receptors and viral envelope proteins, including the HBsAg (Figure 2) (26). This process is facilitated by the binding of HBV envelope proteins and cellular receptors. This is followed by entry of the hepatitis B virion into the hepatocyte through endocytosis and the subsequent release of the nucleocapsid into the cytoplasm (27). The nucleocapsid is uncoated at the nuclear membrane and relaxed circular HBV DNA (rcDNA) is released into the nucleus. The rcDNA is converted into a covalently closed circular double-stranded DNA (cccDNA) molecule that serves as a template for transcription of four viral mRNAs. As transcription of the HBV cccDNA occurs within the hepatocyte nucleus, random integration of the HBV genome into the host chromosomes can also occur. Translation of viral RNA into the various HBV proteins involves four overlapping open reading frames, namely: S (surface envelope), C (core), P (polymerase), and X. The pre-S domain of the large HBsAg protein appears to play a key role in binding of HBV to surface receptors on hepatocytes while the pre-core protein ultimately undergoes proteolysis and becomes hepatitis B e antigen (HBeAg) in the endoplasmic reticulum (28). The largest mRNA, (which is longer than the viral genome), is used to make the new copies of the genome and to make the capsid core protein and the viral RNA-dependent-DNA-polymerase (29). In the assembly phase, these four viral transcripts undergo additional processing to form progeny virions which are released from the cell or returned to the nucleus and re-cycled to produce additional copies (26). Release begins when the long mRNA is transported back to the cytoplasm where the virions P protein synthesizes DNA via its reverse transcriptase activity. The final replication step is the assembly and release of HBV Dane Particles (30).
2.2.5 Pathogenesis of Hepatitis B infection

Hepatitis B virus has the similar structure to retroviruses since it has the ability to replicates through a ribonucleic Acid (RNA) intermediate and can integrate into the host cell genome. In this way, these unique features of the HBV replication cycle ensures persistence of HBV infection in hepatocyte (15). After the susceptible person is exposed, the virus is transported by bloodstream to the liver, which is the primary site for hepatitis B replication (31). The incubation period ranges between 2 to 6 months with an average of 6 months period (23). Hepatitis B infection is detected with serological HBsAg and HBeAg markers (20). Center for Disease Control suggests that hepatitis B virus can produce either asymptomatic or symptomatic infection with clinical manifestation occurring between 6 weeks to 6 months after exposure (23). Hepatitis B virus is regarded as highly infectious organism and it was discovered to be 100 times more infectious than Human Immunodeficiency Virus(HIV) as it can survive on contaminated open surface up to seven days (2).

The age at which a person is infected with the virus determines the disease outcome. About 90% of those who acquire HBV perinatally or in early childhood will develop Chronic Hepatitis B, since their immune system cannot destroy and clear infected hepatocytes (32). In adults, 90% of infections are acute and only 5-10% develop into Chronic Hepatitis B leading to acute liver failure among 1% of acute HBV infections (29),(15). According to Larrubia, 1-2% of people with CHB may lose the HBV surface antigen, which is considered to be a definitive recovery however, the virus can reactivate if they become immunosuppressed (33).
2.2.5.1 The four stages of Chronic Hepatitis B infection

Chronic Hepatitis B (CHB) has four distinct phases with different durations and outcomes, and these phases are linked to the degree of HBV replication and the response of the immune system (29)(34)(35).

1. Immune-tolerant phase: The duration of this phase is normally 10-30 years in cases of HBV that is acquired perinatally; however it can be of shorter duration in HBV infections acquired during childhood. During this phase the hepatitis B viral protein, HBeAg is present (29). During this stage, there is usually minimal liver damage as the immune system tolerates the virus (34).

2. Immune clearance phase (immuno-reactive): This phase is likely to occur in individuals who acquire HBV in late childhood, adolescence or adulthood. In this phase, immunotolerance is lost, and as such the immune system attacks infected hepatocytes. (34). In the first 5 years of this phase, about 50% of cases in this group loses HBeAg and forms Antibodies to HBeAg, a process called seroconversion and this figure rises to 70% by 10 years. (29).

3. Inactive carrier phase: This phase consisting of HBeAg negative HBV with low or undetectable HBV DNA levels, a normal ALT and no damage to the liver (35).

4. Occasional surface antigen loss occurs. In this phase, HBeAg seroconversion to anti-HBe occurs and is associated with minimal HBV replication and less activity in the liver. Patients in this phase form the largest group with HBV (35).

5. Reactivation phase: In this phase the patients reconvert to HBeAg positive spontaneously or due to immunosuppression. Usually most of the patients are HBeAg
negative with detectable DNA levels, high ALT levels, moderate to severe necro-inflammation and liver biopsy shows variable amounts of fibrosis. (34).

2.2.6 Diagnosis of Hepatitis B infection

The diagnosis of HBV infection and its associated disease is based on a constellation of clinical, biochemical, histological, and serologic findings (15). The standard testing for hepatitis infection is conducted by testing the blood serum for a variety of viral antigens or antibody markers (21). A number of viral antigens and their respective antibodies can be detected in serum after infection with HBV, and proper interpretation of the results is essential for the correct diagnosis of the various clinical forms of HBV infection.

Diagnosis of acute hepatitis infection is done based on the following laboratory tests: HBsAg, HBsAb and HBcAb IgM. If HBsAg is negative then acute HBV infection is ruled out, and if it’s positive the patient is infected with HBV and a repeat test after six months will determine has resolved or is chronic (2). The Hepatitis B surface antigen (HBsAg), is a protein on the surface of the virus which can be detected in high levels in the blood during acute or chronic Hepatitis B infection (30). The HBsAg is the first serologic marker to appear in a new acute infection, which can be detected as early as 1 week and as late as 9 weeks, with an average of one month after exposure to the hepatitis B virus (30).

The test is done by Namibia Institute of Pathology (NIP) using serology ELIZA test. Hepatitis screen profile is defined as being made up of the following assays: Hepatitis B Surface Antigen (HBsAg) +/- hep B S AG confirmation test, Anti-Hepatitis B Core antigen IgM, Anti-Hepatitis B Core (Total IgM and IgG), Hepatitis B Surface antibody HAVAb-IgM antibody and Hepatitis C
antibody. HBsAg and other markers for laboratory diagnosis are done on an Architect i2000 or i2000 immunochemistry analyzer (36).

In terms of quality control, before patient samples were analyzed on the architect system, a set of positive and negative controls were analyzed first, and only if the expected results were obtained upon measurement, the patient specimens will be tested. However if expected control results were not obtained upon control testing, no patient specimen were analyzed.

2.2.7 Transmission of Hepatitis B

Hepatitis B Virus is transmitted through activities that involve percutaneous (i.e., puncture through the skin) or mucosal contact with infectious blood or body fluids such as semen or saliva (15). The common routes of transmission includes perinatal, early childhood infection, tribal tattooing and scarification, sexual contact, blood transfusions, unsafe injection practices including injecting drug use and occupational exposure of health care workers (5).

2.2.7.1 Sexual transmission

Seeing that Hepatitis B can be present in semen and genital fluids, sexual intercourse is considered as one the common route of transmission in low endemic area. The unprotected sexual encounter among unvaccinated population with multiple sex partners is regarded as the leading cause of HBV transmission (27). With Infection in adults accounting for 5% of chronic hepatitis B cases, one of the recommended strategies for prevention of sexual HBV transmission is the vaccination of the at risk populations and promotion of safer sex practices (37). This intervention is likely to reduce the prevalence of hepatitis B among pregnant women and subsequently lead to possible decline in perinatal transmission.
2.2.7.2 Percutaneous inoculation

In absence of vaccinations and post- exposure prophylaxis, there is 30% likelihood to acquire Hepatitis B infection through percutaneous inoculation (4). The prominent percutaneous transmission in low and middle income countries is sharing of contaminated instruments. Usually individuals get exposed to infected body fluids during traditional practices such as body scarifications, home deliveries and circumcisions. In high income countries, the common percutaneous routes of transmission is sharing syringes and needles by intravenous drug users (IVDU) (37). Public health education and the use of disposable needles or equipment are considered as effective methods of prevention.

2.2.7.3 Horizontal transmission

Horizontal transmission is acquired via minor breaks in the skin or mucous membranes in the households with child to parents contact being the most notable cause particularly in none vaccinated populations (31). Although HBV DNA has been detected in various body secretions of hepatitis B carriers, there is no firm evidence of HBV transmission via body fluids other than blood (27). However, numerous studies have established that prevalence of Hepatitis B infection seems to be higher in the household with family infected with hepatitis B virus (38)(39). Promotion of personal hygiene, vaccinations and routine screening of the risk population are some of the strategies that are likely to prevent horizontal transmission of hepatitis B infection.

2.2.7.4 Blood transfusion

The risk of acquiring post-transfusion Hepatitis B depends on factors like prevalence and donor testing strategies. In low prevalence areas it is estimated to be one to four per million blood
components transfused (40) while in high endemic areas, the prevalence translates to 1 in 20,000 (41). The higher prevalence among the blood donors in low and middle-income countries creates additional burden in terms of blood safety as resources are limited. However, despite the resource constraints, different strategies are used such as HBsAg screening of donors in African countries. In Namibia, both HBsAg and anti-HBc is used to screen donors (42).

2.2.7.5 Mother to Child Transmission

Perinatal transmission is the common route of transmission of Hepatitis B virus in high endemic areas as one third of HBV infections occur perinatally (43). The opportunities for Hepatitis B exposure from infected mother can occur during prenatal stage which is intrauterine transmission, during delivery and in postpartum (31). In the absence of immunoprophylaxis 10 – 20% Hepatitis B seropositive mothers transmit the virus to their neonates (44).

The study by Chen et al., found out that maternal first-degree family history of HBV infection, intrahepatic cholestasis, and premature rupture of membranes were risk factors for perinatal transmission of HBV (45). It is estimated that intrauterine transmission accounts for 5% mother child transmission (46). This could be attributed to blood-borne infections which spread mainly via the placenta and placental leakages (43). Wang and Zhu suggest that HBV can be integrated into placental tissues leading to infections. This argument supports the theory that intrauterine transmission may occur due to Toxoplasmosis, Other Agents, Rubella, Cytomegalovirus, Herpes Simplex (TORCH) infections, which may result in placental cracks, or placental barrier damage, increases the risk of neonatal HBV infection (47). In addition, Xu and colleagues argue that HIV infection will also increase the risk of HBV infection (14).
Furthermore, prenatal transmission is associated with chronic Hepatitis B infection in pregnancy as presence of HBeAg usually denotes a high Hep B viral load which increases the likelihood for intrauterine transmission (48). However, Petrova and Kamburov argue that the risk of intrauterine infection is relatively low because the fetus is protected from HBV by the placenta. Reported vaccination failures imply in utero transmission especially in high-risk groups (49). The intrapartum transmission, which occurs during delivery, accounts for 90% - 95% of hepatitis B infection from infected mothers to the newborns (7). Labor transmission occurs mainly through the HBV contaminated maternal blood, amniotic fluid, and vaginal secretions, which are either swallowed by the fetus or get into the fetal blood circulation by placental rupture (35). In addition, premature rupture of membranes was associated with an increased risk of maternal-neonatal transmission (35).

Various studies have demonstrated that vertical transmission can be minimized by immediate administration Hepatitis B immunoglobulin and hepatitis B vaccination. However, the efficacy of these interventions is still being disputed (50), (51). Although little is known about transmission in infancy, a small proportion infants will acquire the hepatitis infection during postpartum stage through breastfeeding and other forms of contacts with contaminated body fluids such as saliva (43). In addition, studies established that mothers who are positive for HBsAg, HBeAg, and anti-HBc, have detectable HBV-DNA in their breast milk, but if only HBsAg is positive, HBV-DNA can be detected in only 46% of the subjects (37), (52).

The World Health Organization estimates that about 10% of exposed infants will be infected with hepatitis B infection despite the vaccination and provision of immunoprophylaxis (53). In Gambia the vaccination rate has reached 90%, and this has reduced Gambia’s HBsAg-positive rate to below 0.3% (54). However, Jonas argues that provision of joint neonatal HBV vaccine
and HBIG still have an immunization failure rate of 20%-30% in infants born to HBsAg-positive mothers (46).

Numerous studies have established that children infected with hepatitis B in infancy can be carriers of hepatitis B later in adulthood and subsequently develop life threatening liver related disease such as hepatic carcinoma and liver cirrhosis (52),(55),(56). The study by Ioannis et.al reflects that the presence of HBV-DNA in cord blood is significantly associated with spontaneous preterm birth in chronic HBV-infected pregnant women (48).

Considering that mother to child transmission of hepatitis B is the common route transmission and is associated with poor outcome of labor such as preterm delivery, it is relevant that primary infection in pregnant women is prevented by understanding and mitigating the potential associated risk factors.

2.3 RISK FACTORS FOR HEPATITIS B INFECTION

The risk of hepatitis B infection in pregnancy is similar with general female population as most of hepatitis infections are acquired prior to current pregnancy (46). The study by Ibrahim and colleagues revealed that there is no significant differences in prevalence between pregnant and none pregnant women however, there are risk factors more common and specific to pregnant women (57). This subsection reviews the literature on the identified risk factors in the study.

2.3.1 Socio demograhic factors

Study by Lao and colleagues demonstrates that age is significantly associated with high hepatitis B prevalence where women younger than 25 years were found to have a significant higher prevalence of hepatitis B infection (58). These findings were found to be consistent with study in Uganda (59). Lao and colleagues argue that this trend could be attributed to horizontal
transmission in early adolescent period (58). However, in agreement with numerous studies, age was not found to be as significant risk factor (57),(60).

In relation to geographical distribution, pregnant women living in urban areas have significantly higher burden of hepatitis B as compared to rural pregnant women. However, numerous studies have shown that geographical distribution to be insignificant in the acquiring of Hepatitis B infection (24),(1),(61). Lem and colleagues argue that higher hepatitis B rates among pregnant women living in urban areas is attributed to high risk lifestyle which prevails in urban areas (24). The study in Cameroon by Abongwa and colleagues concurs well with the notion that significantly higher prevalence rates of HBV infection occur among the urban pregnant women than rural pregnant women, this however was not found to be significant (24). These results contradict the findings of Mohebbi and colleagues were no difference in prevalence of hepatitis B was observed between urban and rural pregnant women (20).

Educational status was found to be associated with hepatitis B infection among pregnant women in numerous studies (57)(1). This argument is consistent with the Cameroonian study were HBV infection prevalence was found to be 4.2% among pregnant women with tertiary education compared to 23% among women who never attended school (24). These findings were attributed to high level of educational awareness on risky behavior and prevention among the educated pregnant women (62). However the numerous studies indicate that educational level is not associated with hepatitis B infection in pregnant women (44),(20),(63).

Marital status is another important socio demographic factor which might be associated with hepatitis B infection in pregnant women. The study done by Yakasai and colleagues demonstrated no association between marital status and seroprevalence for hepatitis B virus
The results of this study further revealed that pregnant women married in polygamous unions are significantly at risk of acquiring hepatitis B infection because multiple partnerships poses a risk of sexually transmitted diseases including hepatitis B virus (57). These findings are consistent with studies in Kenya and Uganda (59) (64). Pregnant women living in cohabitating relationships in Cameroon carried a significant higher burden of hepatitis B infection (24). On the other hand various studies conducted across the globe have contradicting findings, as marital status was not found to be associated with hepatitis B infection among pregnant women (61)(59).

Numerous studies have attempted to link the ethnicity of pregnant women with prevalence of hepatitis B infection (65). Cultural and social norms of specific ethnic groups were found to be risk factors to prevalence of communicable disease including hepatitis B infection (66). In Namibia, Ovahimba and Ovadhemba pastoral ethnic group which resides in Kunene and some part of Omusati region have similar ancient nomadic and cultural practices as Masai tribes in Kenya (67). The Maasai people in Tanzania and Kenya, and Ovahimba living in Namibia are often characterized by atypical pastoral lifestyle which is based primary or exclusively on livestock herding (68). According to United Nation’s report on State of Indigenous People, the modernization of homelands of indigenous ethnic tribes made the inhabitants susceptible to infections such as HIV/AIDS and viral hepatitis (11). The studies done in Nigeria found no association between ethnicity and Hepatitis B infection (57)(65).

Type of occupation was also identified as a significant socio-demographic risk factor which can be associated with hepatitis infection in pregnant women. These findings confirms with study by Eke and colleagues were occupational status for health worker was found to be a significant risk factors (65). These findings could be attributed to possible exposure to contaminated body fluids
in the clinical settings. The findings of Yakasai and colleagues demonstrated a higher prevalence of hepatitis B infection among professional pregnant women, however this association was found to be insignificant in other studies (57). In contrast, the study by Umare and colleagues found out that unemployed pregnant women are associated with higher prevalence of hepatitis B infection but was not considered as significant exposure factor (61).

2.3.2 Obstetric/ Gynecological history

Obstetric and gynecological history of women can reveal a number of exposure factors which can have influence on acquiring hepatitis B infection. The study in Deder Hospital, Ethiopia could not establish the association between parity and hepatitis B infection in pregnant women attending antenatal care (61). Similar results were found in South Nigeria and in Kenya where hepatitis B prevalence was significantly higher in nulliparous women (63),(69) Although one would expect the prevalence to be significantly higher in multiparous women due to frequent exposure to contaminated surfaces and instruments during delivery (62), Alegbeleye and colleagues argue that nulliparous pregnant women may have terminated pregnancies and numerous sexual partners which could have exposed them to hepatitis B virus as opposed to multiparous women (63). Despite these arguments, various studies failed to establish the association between hepatitis B infection and parity (24),(61),(60).

Another prominent risk factor which is found to be significantly associated with hepatitis B infection in pregnant women is history of abortion (57). These findings are well eluded in the Ethiopian, and Nigerian study that found out that history of abortion is a significant risk factor (44)(61),(63). Instruments and procedures used during abortions are likely to be a source exposure as number of abortions are conducted by unskilled personnel in unsterile environments (44). This finding is in agreement with study in Nairobi Kenya where the
researchers found out that the prevalence of HBV infection was significantly higher among pregnant women who had history of abortion (69). Ezechi and colleagues argue that instrumentation during abortion and related activities may serve as sources of exposure and subsequently may increase the likelihood of acquiring the infection (70). The World Health Organization estimates that on average, 56 million induced abortions occurred worldwide each year which translates to 35 induced abortions per 1000 women aged between 15–44 years of which 25% of all pregnancies ended in an induced abortion (71). The results from a study in Iran contradicts this argument as history of abortion was found to be insignificant risk factor (1).

Women with a history of home delivery, have higher risk of acquiring hepatitis B infection due to absence of infection control (72). On contrary, the study in Ebony State, Nigeria found out an insignificant difference between women who delivered at home and those who delivered in the hospitals (60). As hepatitis B virus is able to survive outside the hosts for up to seven days, nosocomial infections during hospital deliveries is likely to take place if universal precautions are not followed (15).

### 2.3.3 Potential Risk Factors

Numerous studies agree that body piercing, tattooing and scarifications are associated with hepatitis B infection in pregnant women (24), (61), (65). The use of contaminated instruments is identified as a driver for hepatitis B transmission in the general population including pregnant women (73). Inconsistent results were found by Bawazir (74), Nazzal (38) and Bayo (59) where no significant association between scarifications and hepatitis B infection were observed. Other researchers documented that significant risk factor for hepatitis B infection is history of blood transfusion (41).
This high prevalence of Hepatitis B infection is associated with blood transfusion as illustrated by transfusion risk models where risk exceeds that of HIV yet receives comparatively little attention(75). In agreement with other studies, history of blood transfusion was found to be strongly associated with hepatitis B infection in pregnant women in Yemen, Ethiopia and Cameroon.(74)(61)(76). Although there have been medical breakthrough that led to the improvement in screening of blood and blood products, the studies in resource constrained countries reflects a significant association of hepatitis B infection in pregnant women with history of blood transfusion (60),(1)(74). The study on blood donors in Namibia, revealed the prevalence of 0.6%, however all donors and blood products go through extensive screening before being used by the recipients (42). Numerous studies have found out that history of blood transfusion is an insignificant risk factor (38),(59). Bloch and colleagues argue that availability of the HBV vaccine and increase in immunization coverage in Africa may mitigate prevalence and transfusion risk in the future (75).

History of tooth extraction or dental procedure and history of surgery were found to be significantly associated with hepatitis B infection in pregnant women (77)(38). A study by Ugbebor colleagues shows that history of tooth extraction was a significant predictor for hepatitis B infection due to non-adherence of infection control measures in dental clinic and surgical procedure rooms (78). Studies in Iran that were conducted in the pre-vaccination era showed that HBV infection of dentists was approximately three to six times greater than in the general population, and dentists had the highest rate of HBV infection among all health workers (79). Alavian and colleagues argue that percutaneous injuries in dental students were more frequent than in all other health care students (79). However, the study in Ethiopia and North-
Eastern Nigeria showed that there was no association between history of dental procedure and hepatitis B infection (61),(80).

History of hospital admission was found to be significant risk factor for nosocomial hepatitis B infection in pregnant women (38). Possible factors that might predispose this group of patients to develop HBV infection includes the following: severe immune-suppression secondary to the disease, prolonged hospital admission, repeated venipunctures and an increased need for invasive procedures (81). On the other hand, the study done by Umare in Ethiopia concluded that history of hospital admissions was not associated with hepatitis B infection among pregnant women (61).

Nosocomial infection can occur from patient to patient, from patient to health care worker and other way around. Hepatitis B virus is considered the most commonly transmitted blood-borne virus in the healthcare setting (5). Despite a number of prevention strategies such as use of disposable needles consumables, sterilization of surgical instruments and vaccination of healthcare workers documented cases of nosocomial of hepatitis B infection do occur (82). However, the exact risk of nosocomial infection is unknown. Seeing that hepatitis B infection can be asymptomatic, the number of infected patients reported in the literature is likely to be underestimated as many infected patients may be asymptomatic and only a fraction of exposed patients are detected and recalled (81). Vaccination of health workers and adherence to universal precautions during medical procedures are some of the effective strategies for prevention of hepatitis B from health workers.

History of sexual transmitted infection (STI’s) can have a significant association for hepatitis B infection in this population as STI’s and HBV share the common route of transmission (83). The study in Ethiopia demonstrates that history of sexually transmitted infections is a significant
predictor for hepatitis B infection (44). These findings are consistent with study in Lagos, Nigeria where history of sexually transmitted infections was a significant risk factor (84). These findings could be attributed to the fact that hepatitis B is present in blood, semen and other body fluids where sexual contacts with multiple partners serves as a vehicle for transmission of infection in pregnant women. However the similar studies done by Ethiopia and Nigeria seems to confirm otherwise (44), (65).

2.3.3.1 Human Immunodeficiency Virus (HIV) Status

According to Center for Disease Control (CDC), immunosuppressed patients including HIV infected are at risk of developing chronic hepatitis B infection (23). The vast majority of people living with HIV/AIDS are in low- and middle-income countries with Sub-Saharan Africa being the most affected region, with an estimated 25.6 million people living with HIV in 2015 and 66% of new estimated HIV infections in 2015 occurred in sub-Saharan Africa (85).

There has been an increase in access to antiretroviral therapy (ART) in sub-Saharan Africa, which has improved the life expectancy among the HIV-infected individuals. Consequently as people live longer, HBV related morbidity and mortality in HIV co-infected patients become more prominent (86). HIV apart from being identified as a risk factor, the co-infection with hepatitis B Virus can accelerate the progression of both infections. The study by Ladep and colleagues concluded that low immunity levels among HIV patients who are co-infected with hepatitis B as opposed to HIV mono-infected patients leading to accelerated HIV disease progression which can negatively impact on maternal and fetal outcomes (87). Furthermore, Matthews and colleagues suggest that there chronic HIV/HBV co-infection is associated with long-term morbidity and mortality that exceeds the impact of infection with either one of these viruses alone in African populations (88). This argument is in agreement with Nikolopoulos et
al., who suggest that HIV-HBV infection seems to increase the infectivity of HBV, the rate of HBV reactivation and the risk of cirrhosis (89).

In Sub – Saharan Africa where hepatitis B infection is endemic, 13% of HIV infected pregnant women are co-infected with hepatitis B virus (46). The study by Barth et al., reflects 12% of HBsAg and 17% of HBeAg prevalence among HIV positive study population (86).

Hepatitis B Virus and Human Immunodeficiency Virus share the common mode of transmission and can have negative health effect on pregnant women and infants. The Ethiopian study demonstrates that HIV infection among pregnant women in Bahir Dar city was significantly associated with hepatitis B infection (90). The findings by Dial et al., confirm the significant association between HIV and hepatitis B infection in pregnant women in Tshwane District in South Africa (10).

Barth and colleagues argue that the association between hepatitis B infection and HIV status is not prominent in Africa as most of the individuals get exposed to HBV before acquiring HIV (86). The study in Uganda seems to confirm this argument as there was no association found between hepatitis B infection and HIV status among pregnant women (59). The study in Western Cape, South Africa reflects higher prevalence of HBsAg seropositivity among HIV positive pregnant women as opposed to HIV uninfected women however, the observed difference was not statistically significant (91). Similar results were obtained in Kwazulu Natal, South Africa (92). Additional studies are required to understand the relationship between Hepatitis B infection and HIV.
2.3.3.2 Vaccinations Status

None vaccination or unknown vaccination status is a potential significant risk factor for hepatitis B infection (38). Recombinant DNA-derived vaccines against HBV have been available for more than two decades which consists of three doses of vaccine (93). The vaccine contains one of the viral envelope proteins, hepatitis B surface antigen (HBsAg). It is produced by yeast cells, into which the genetic code for HBsAg has is inserted and subsequently an antibody to HBsAg known as anti–HBs is established in the bloodstream that leads to and immunity to HBV infection (94).

The Hepatitis B vaccine is an unaudited virus which provides acquired immunity after it is introduced in the body. Vaccination of infants with hepatitis B vaccine within 24 hours of birth was found to be 90–95% effective in preventing infection with HBV as well as decreasing HBV transmission if followed by at least two other doses (37). The vaccine demonstrated 91% decrease in hepatitis B infection after it was introduced in China while the decline in incidence of acute hepatitis B was also observed in Taiwan which was attributed to the universal neonatal hepatitis B vaccinations (53).

The World Health Organization recommends universal hepatitis B vaccination for all infants, and that the first dose should be given as soon as possible after birth as this strategy has resulted in a dramatic decrease in the prevalence of chronic hepatitis B among young children in regions of the world where universal infant vaccination programs have been implemented (95). However, Chen et al., argue that a proportion of vaccinated children (5–10%) has a poor response to vaccination, and will remain susceptible as adults to acquisition of HBV infection (53).
Seeing that countries with intermediate or low endemic transmission, may result from acute and chronic infection acquired by older children, adolescents and adults, target groups for catch-up vaccination as well as other preventive strategies that covers young adolescents; household and sexual contacts of persons who are HBsAg-positive; and other persons at risk of acquiring HBV infection is recommended.

The World Health Organization recommends for member countries to implement hepatitis B vaccination as part of global strategy to decrease disease burden and prevention related life threatening hepatic illnesses (95). In 2015, Namibia has introduced the Hepatitis B vaccine as part routine vaccination for infants.

2.4 INTERVENTIONS FOR HEPATITIS B INFECTION

Improving the identification and public health management of persons with chronic HBV infection can help prevent chronic liver infection that will help to eliminate HBV transmission (23).

The interventions for reducing hepatitis B are implemented using all levels of care such as primary prevention secondary prevention and treatment.

2.4.1 Prevention of Hepatitis B Infection

The WHO guidelines on Hepatitis B is recommending high risk population such health workers, individuals who are incarcerated, intravenous drug users, sex workers, men sleeping with men and pregnant women to be screened for hepatitis B infection (96). Serologic testing for hepatitis B surface antigen (HBsAg) is the primary way to identify persons with chronic HBV infection. The directive on Focused Antenatal Care in Namibia recommends that all pregnant women are screened for hepatitis B infection as part of routine prenatal investigation. The screening of HBV
infection in the perinatal period has become standard care, which can identify newborns that require prophylaxis with hepatitis B vaccine and HBIG as well as pregnant women who require antiviral therapy. Specific hepatitis B immunoglobulin (HBIG) is available for passive protection and is normally used in combination with hepatitis B vaccine to confer immediate passive and active immunity in newborns, which is administered as an effective prophylactic measure to prevent mother to-infant transmission of HBV, however, 5%-10% infants of HBeAg-positive mothers are still infected with HBV despite these interventions (55).

2.4.2 Treatment for Hepatitis B Infection

Although HBV infection can be prevented by vaccination, it is important to treat CHB persons who are at high risk of progressing in order to reduce morbidity. Currently, seven antiviral agents such as lamivudine, adefovir, entecavir, telbivudine, tenofovir, emtricitabine, standard and PEGylated Interferon (PEG-IFN) are approved for the treatment of CHB in high-income countries, and have been shown to delay the progression of cirrhosis, reduce the incidence of HCC and improve long-term survival (4). However in Low income countries, the anti-retroviral (ART) treatment is provided based on affordability and clinical condition of the patient as positive outcomes are not always guaranteed. In addition, the issues some the ART’s are used for HIV treatment and possibility of future drug resistance has to be considered (97).

2.5 SUMMARY

This chapter covered the epidemiology, pathogenesis and natural history of hepatitis B virus. The chapter also covered extensively on mother to child transmission of hepatitis B infection. The risk factors for hepatitis B were explored in detail. The chapter elaborated on recommended interventions which can reduce the morbidity and mortality of hepatitis B infection
CHAPTER THREE

3. RESEARCH METHODOLOGY

3.1 INTRODUCTION

The literature review which included epidemiology, pathology, natural history and risk factors of Hepatitis B infection among pregnant women was covered in the previous chapter. This chapter is outlining the processes, procedures and guidelines involved in the production of this research. This section presents an overview of the research design, study population, sampling, data collection, data analysis and ethical considerations.

3.2 RESEARCH DESIGN

The study design refers to the architecture of the study as the choice of the study design determines the sampling, the data collection and data analysis methods (98). The choice of study design is determined by the research questions and its objectives. In this study questions related to Hepatitis B risk factors were asked to the respondents and observational checklists used to establish the presence or absence of such risk factors in Kunene region. The suitable study design for answering the research questions was analytical approach. In contrast to a descriptive studies, the goal of analytical studies is to determine the cause of or risk exposure for a disease by assessing whether particular exposure is related to a disease or other health outcomes of interest (98). In this study the analysis focused on presence of risk factors among the cases and controls with the aim of establishing the relationship between the identified risk factors and Hepatitis B infection among the pregnant. Subsequently the suitable study design was a facility based on unmatched cases control which examined the risk factors associated with HBV infection among pregnant women in Kunene Region. A case control study is type of observational study in which
the subjects are selected on basis of either being having a particular disease (cases) or not having a disease of interest (controls) (99). Ones cases and controls are selected, the researcher collects exposure measurements and compares them between the two groups.

In this study situation the cases were enrolled from ANC attendees with reactive serology result for HBsAg or HBeAg or reactive to both hepatitis B antigen markers while controls were recruited from ANC attendees with non-reactive serology results for both HBsAg and HBeAg. The ratio for cases to controls was 1:2 respectively. Seeing that this study design was observational, the information was generated without manipulating the variables. The quantitative approach has enabled the quantification of identified risk factors for HBV.

### 3.3 STUDY POPULATION

A study population is a well-defined collection of individuals or objects known to have similar characteristics (100). All individuals or objects within a certain population usually have a common, binding characteristics or traits. Depending on the available resources and study design, the researcher can have target population which includes all subjects that have characteristics of interest (98). Alternatively, one can use the accessible population which is the portion of the population to which the researcher has reasonable access and may be limited to region, state, city, county, or institution (98).

In this study the accessible population of interest was pregnant women who have attended the antenatal care visits and have documented laboratory HBV results in the nine health facilities in Kunene region during the period of January – August 2017. In total nine (9) public health facilities which are providing ANC services were selected in order of frequency in ANC attendance.
3.3.1 Study Setting

The study setting was based in Kunene region which is situated in North Western part of Namibia located along the southwestern part of Angola. Kunene is one of the fourteen administrative regions of Namibia. Kunene's western edge is the shores of the Atlantic Ocean. In the north, the region borderers Angola and has estimated population of 98,844 (101). The area size is 115,293 Square Kilometers where population density is 0.8 per square km with 26% of population living in urban areas while 74% lived in rural areas (101).

The most commonly spoken languages at home are Otjiherero language (47% of households), Nama/Damara (32%) and other Namibian languages 21 percent. The Region is home to nomadic Himba ethnic group, Ovadhemba, Herero, and other communities. The Himba are indigenous peoples with an estimated population of about 58,000 people in the Kunene Region and on the other side of the Kunene River in Angola (101). There are also a few groups left of the Ovatwe, who are also Himba, but are hunter-gatherers. The Himba are a semi-nomadic, pastoralist people, culturally distinguishable from the Herero people in northern Namibia and southern Angola, and speak OtjiHimba, a variety of Herero, which belongs to the Bantu family within Niger–Congo (102). Due to harsh climatic and geographic conditions where they resides and their seclusion from outside influences, the Himba have managed to maintain and preserve much of their traditional lifestyle. One of the common cultural practices is polygamy and girl child. The annual growth rate is 2.3% and fertility rate is 4.9 children per woman (101).

In terms of health care Kunene Regional Health Directorate is demarcated by three health district which is Opuwo Health District, catchment population is 58,480, Outjo Health District with population of 22,876 and Khorixas with population of 13,765(10). The health directorate’s mandate is to provide health services through 3 district hospitals, 3 health centers, 24 clinics and
199 outreach points. The region has 20 Primary Health Care Facilities which are screening pregnant women for Hepatitis B infection.

### 3.3.2 Study Inclusion and Exclusion Criteria

#### Table 3.3.2 Inclusion and exclusion Criteria

<table>
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<td>Not attended ANC first visit during the period under review</td>
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<tr>
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<td>Results for Hepatitis B serology done at ANC and results available</td>
<td>Hepatitis B serology test not done/results unknown</td>
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### 3.4 PROCEDURE

#### 3.4.1. Serologic testing for Hepatitis B Screening at ANC sites

All interviewed cases and controls had laboratory results of Hepatitis B test which is done routinely at first ANC visit. The test is done by Namibia Institute of Pathology (NIP) using serology ELIZA test. HBsAg and other markers for laboratory diagnosis were done on an Architect i2000 or i2000 immunochemistry analyzer (36). The specimens were loaded onto the architect analyzers and run automatically and the results were transferred automatically from the analyzer to the laboratory information system. Results were given as either reactive or non-reactive.
3.4.2 Risk Factors Assessment

Trained health workers interviewed cases and controls using a standardized questionnaire and ANC record data abstraction form. Risk factors were categorized into socio-demographic information, obstetric and gynecological history, general biological potential risk factors. Socio-demographic factors included age, place of residence (rural or urban), marital status, educational level, ethnicity and employment status. The obstetric and gynecological history included gravidity, parity, history of home delivery and hospital delivery, type of delivery (history vaginal and caesarean section) and history of abortion. The general biological exposure risk factors included body piercing or scarifications, presence or history of tattoos, blood transfusion, hospital admission, dental procedure, history of surgical procedures, history of STI’s, HIV status and vaccination status.

3.5 SAMPLING METHOD

Sampling is defined as the process of selecting a group of people, events, behaviors, or other elements which is representative of the research population (103). The purpose of the sampling process is to enable the researcher to observe the characteristics of study subjects which could be representative of the entire population. Furthermore sampling ensures that less resources and time are used as study population is downsized to the identified selected sample. Sample is the selected elements or group of individuals for participation in a study where people are referred to as subjects or participants (100). The sample was derived from the sample frame of ANC clients that were screened for Hepatitis B during the antenatal care visits at the selected health facilities during the period of January to June 2017 (Figure 3.5).
The population size was projected from 1969 ANC clients in Kunene region during the period of January – August 2016. Population size estimation was based on 2016 Health Information System report for ANC first visit attendance for the period of eight months. In this study cases were defined as follows: Pregnant women attending the antenatal care in the sampled health facilities during the period under review with the documented reactive results for HBsAg or HBeAg test or reactive to both Hepatitis B antigen markers. Controls are pregnant women that have attended antenatal care in the sampled health facilities during the period under review with documented non-reactive or negative results for HBsAg and HBeAg markers. Systematic random sampling was used for the selection of ANC clients with non-reactive results for HBsAg/HBeAg (controls). From the sampling frame of 1573 Hepatitis B negative clients, every sixth client with none reactive HBsAg/HBeAg test was selected from the ANC registers and

**Figure 3.5: Flow Chart for Sampling Process**

The population size was projected from 1969 ANC clients in Kunene region during the period of January – August 2016. Population size estimation was based on 2016 Health Information System report for ANC first visit attendance for the period of eight months. In this study cases were defined as follows: Pregnant women attending the antenatal care in the sampled health facilities during the period under review with the documented reactive results for HBsAg or HBeAg test or reactive to both Hepatitis B antigen markers. Controls are pregnant women that have attended antenatal care in the sampled health facilities during the period under review with documented non-reactive or negative results for HBsAg and HBeAg markers. Systematic random sampling was used for the selection of ANC clients with non-reactive results for HBsAg/HBeAg (controls). From the sampling frame of 1573 Hepatitis B negative clients, every sixth client with none reactive HBsAg/HBeAg test was selected from the ANC registers and
enrolled in the study as a control (1573/247 = 6). Due to the limited number clients with reactive HBsAg/HBeAg results, purposeful sampling selected 115 clients with Hepatitis B reactive results as cases. The total study population consisted of 115 cases and 230 controls which generated 345 the study population.

Both cases and controls must have attended ANC during the period of January – August 2017 and consented to participate in the study. The purposive sampling method was applied to for selection of 9 high volume health facilities which attend to 90% of ANC clients in the region. The 25 ANC facilities in the region were ranked by the number of annual first ANC visits of which 9 were selected.

3.5.1 Sample size Criteria

The sample size in this case control studies is determined by precision, power, confidence interval, and the documented odds ratio and frequency of disease of interest.

3.5.1.1 The Level of Precision

The level of precision, sometimes called sampling error, is the range in which the true value of the population is estimated to be. This range is often expressed in percentage points (e.g., ±5 percent) in the same way that results for political campaign polls are reported by the media (104).

Thus, if a researcher finds that 60% of health care workers in the sample have adopted a recommended practice with a precision rate of ±5%, and then he or she can conclude that between 55% and 65 of health care in the population have adopted the practice (105). In this study the level of precision is set ±5 percent.
3.5.1.2 Power

Statistical power is a measure of the likelihood that a researcher will find statistical significance in a sample if the effect exists in the full population (104). The common purpose of conducting a power analysis is to determine the sample size needed for a particular study. In this study power is set 80 percent as it will ensure that type I and II error are eliminated from the study by securing sufficient sample size.

3.5.1.3 Confidence Interval level

The principle behind the confidence interval level is that when a population is repeatedly sampled, the average value of the observed attribute by those samples is equal to the true population value. In a normal distribution, approximately 95% of the sample values are within two standard deviations of the true population value which is the mean value. This means that 95% confidence level will represent, 95 out of 100 samples that will have the true population value within the confidence level limits. However there can be a possibility that the obtained sample will not represent the true population value. In such situations, samples with extreme values are represented by the 5% (2.5% on each side) area under the normal distribution curve. This can be mitigated by reducing for 99% confidence levels and increased for 90% (or lower) confidence levels(105). In this study, the confidence interval is set at 95 percent.

3.5.2 Sample Size calculations

The Epi info version 7.2, Statistical software for unmatched case control studies was used to calculate the sample size based on the following assumptions (table 3.4.2):
Table 3.5.2 Parameters used for sample size sample size calculation

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Specifications</th>
</tr>
</thead>
<tbody>
<tr>
<td>Confidence Interval</td>
<td>95%</td>
</tr>
<tr>
<td>Power</td>
<td>80%</td>
</tr>
<tr>
<td>Ration of controls to cases</td>
<td>2:1</td>
</tr>
<tr>
<td>Percentage of exposed controls</td>
<td>8%</td>
</tr>
<tr>
<td>Minimum detectable odds</td>
<td>2.5</td>
</tr>
</tbody>
</table>

Based on the parameters shown in (table 3.5.2), the sample size of 122 cases and 243 controls (n = 365) was generated. The sample size of 122 cases and 243 controls from pregnant women attending ANC was divided into 9 strata by using a proportionate sampling technique depending on the population size of the stratum. This type of sampling technique ensures that equal representation and improved precision which in return prevent biased information in the study. After the allocation of sample size in the identified strata, systematic random sampling was done using the random numbers with Microsoft excel. Subsequently, the sample size for each stratum was calculated based on the population size of pregnant who attended the ANC first visit during the designated period. (Table 3.5.3)
Table 3.5.3 A proportionated sample size per public health facility (HF) in the Kunene Region N= 365

<table>
<thead>
<tr>
<th>Number</th>
<th>Name of health facility</th>
<th>Population of Pregnant women attending ANC first visit in six months</th>
<th>Proportion of Cases</th>
<th>Proportion of controls</th>
<th>Proportion of sample size per health facility</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Opuwo Clinic</td>
<td>828</td>
<td>60</td>
<td>120</td>
<td>180</td>
</tr>
<tr>
<td>2</td>
<td>Sesfontein Health Centre</td>
<td>52</td>
<td>4</td>
<td>8</td>
<td>12</td>
</tr>
<tr>
<td>3</td>
<td>Okangwati Health Centre</td>
<td>114</td>
<td>8</td>
<td>16</td>
<td>24</td>
</tr>
<tr>
<td>4</td>
<td>Ohandungu Clinic</td>
<td>49</td>
<td>4</td>
<td>7</td>
<td>11</td>
</tr>
<tr>
<td>5</td>
<td>Orumana Clinic</td>
<td>39</td>
<td>2</td>
<td>5</td>
<td>7</td>
</tr>
<tr>
<td>6</td>
<td>Etanga Clinic</td>
<td>77</td>
<td>6</td>
<td>11</td>
<td>17</td>
</tr>
<tr>
<td>7</td>
<td>Outjo Clinic</td>
<td>284</td>
<td>20</td>
<td>40</td>
<td>80</td>
</tr>
<tr>
<td>8</td>
<td>Kamanjab Health Centre</td>
<td>99</td>
<td>7</td>
<td>14</td>
<td>21</td>
</tr>
<tr>
<td>9</td>
<td>Khorixas Clinic</td>
<td>153</td>
<td>11</td>
<td>22</td>
<td>33</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>1695</td>
<td>122</td>
<td>243</td>
<td>36</td>
</tr>
</tbody>
</table>

Total Population (Sampling Frame): 1695

Sample size : 365 (cases and control)
The proportion of sample size was calculated using the following formula: population per health facility divided by total population multiply by total sample size = proportion per health facilities.

\[ \text{E.g. Opuwo clinic: } \frac{828}{1695} \times 122 = 60 \text{ cases}; \frac{828}{1695} \times 243 = 120 \text{ controls} \]

3.6 RESEARCH INSTRUMENTS

A data research instrument is a measuring tool used for collection of information needed to address a research problem or question (100). It is crucial that the research instruments answers the objectives of the studies. Much of the information collected in observational epidemiologic studies is collected in the form of patient/participant self-reports on standardized questionnaires which are either self or interviewer administered in person, by phone, or via mail or the internet. The factors on which information is routinely collected in these studies include socio-demographic characteristics, lifestyle practices, medical history, and use of prescribed and/or over the counter medications (106). The researcher used two types of data collection instruments. For the primary data source, an interviewer-administered questionnaire was used while for secondary data source antenatal care record review form was used.

3.6.1 The structured questionnaire

This instrument was administered to study subjects after the written consent was obtained. The questionnaire consisted variables which aimed at identifying the risk factors. Section A: Demographic Factors - Age, Place of Residence, Marital Status (single, cohabitating, civil, traditional monogamous, traditional polygamous) Educational Level, Occupation and Ethnicity/Home language. Section B: Biological exposure factors – parity, type of previous delivery, history of abortion, history of blood transfusion, previous minor or major surgery, and history of scarifications/tattoos/piercing. Each questionnaire was assigned with unique
identifying numbers which is entered on each page and on the data collection software (appendix B).

3.6.2 Antenatal Care Record Data Abstraction Form

This instrument was used for collecting the secondary information from antenatal registers and individual patient’s documents. This instrument was aimed to collect the client related records such as results for HBsAg or HBeAg, HIV results and vaccination status (see appendix C).

3.7 DATA COLLECTION METHODS

This subsection emphasizes on the data procedures and process. Data collection refers to the process of gathering and measuring information on the variables of interest in an established systematic fashion that enables the researcher to answer the stated research questions and evaluate the outcomes (107). During this stage, the researcher takes note of what type of information is required, data collection method, who will collect the data, where the data will be collected and when the data will be collected (103).

In this study the data was collected using structured questionnaires for individual clients and retrospective review of antenatal care records for the period 1st January - 31st August 2017. After a written consent was obtained from each study subjects, information concerning socio-demographic and other potential risk factors for HBV infection was collected through face to face interview using a pre-tested standard questionnaire. A pretested ANC data review form from client’s records was used.

The questionnaires and ANC record review form was assessed for completeness and accuracy. Incomplete and inaccurate data collection instruments were manually cleaned by the investigator
refilled when possible or exclude from the study. The completed questionnaires and ANC record review forms were entered on Epinfo 7.2 version.

3.7.1 Validity and reliability

The questionnaire was pre-tested to ANC clients from designated pilot heath facility (Ombombo Clinic, in Kunene region) to assess for relevance, sensitivity and acceptability of the questions. The participating health facility for the pilot testing were excluded from the study while the participating subjects were assigned with unique identifiers in the ANC passport which will be used as identification for exclusion from the official study. The questionnaires were translated from English to three common local vernacular languages spoken in Kunene and translated back to English. The instrument was edited after the outcome of pre – testing exercise.

3.8 DATA ANALYSIS METHODS

Data analysis is the process of developing answers to questions through the examination and interpretation of data (107). The basic steps in the analytic process consist of identifying issues, determining the availability of suitable data, deciding on which methods are appropriate for answering the questions of interest, applying the methods and evaluating, summarizing and communicating the results (103).

The researcher entered data in a computer, after it was cleaned, coded and edited for inconsistencies and was analyzed using Epi Info 7.2 and Microsoft excel. Descriptive analysis in the form of frequencies, proportions and means were used to describe the distribution of socio-demographic characteristics and prevalence of risk factors in cases and controls. We determined the differences among each group of socio-demographic factors using chi square for trend.
Bivariate analysis was done to generate the odds ratio to determine the association between each risk factor and Hepatitis B infection. We set confidence interval at level 95% to test for statistical significance. Multiple logistic regressions were done by including all risk factors that were found to be significant in the bivariate model. The P-value of < 0.05 which was generated from multiple logistic regressions was regarded as a true association.

3.9 ETHICAL CONSIDERATIONS

The study commenced after the approval from the University of Namibia Post Graduate Studies Committee, and ethical clearance from the Publication and Research Committee was received. In addition permission to conduct the study was obtained from the National Health Research Unit of the Ministry of Health and Social Services. The Kunene Regional Health Directorate was informed prior to the commencement of the study. Ethical principles of autonomy and confidentiality, beneficence, non-maleficence and justice were maintained throughout the study.

3.9.1 Autonomy and Confidentiality

Prior to interviews, informed signed consent from all participants was required. The aim of the study was explained to the study subjects. The right and dignity of the study subjects was respected throughout the research period. Participation in the study was voluntary and study subjects could withdraw from the study at any stage without being coerced or intimidated.

Questionnaires and data abstraction tools were anonymous and were assigned with unique identification numbers. The electronic data base was locked on the restricted computer which was allocated with a password and paper based data was stored in a restricted lockable cabinet.
3.9.2 Non – Maleficence

The study subjects did not undergo procedures or processes which might have caused physical or emotional discomfort. In order to avoid unnecessary physical discomfort, blood samples were not collected for study purposes as results were from routine ANC screening were made available. For emotional comfort and assurance, clients were individually interviewed in consulting rooms by one interviewer.

3.9.3 Beneficence

The study served as a benchmark for implementation HBV control strategies in Kunene region and country at large. Study subjects with HBsAg and HBeAg positive were given opportunity to access the treatment for themselves and their infants.

3.9.4 Justice

All pregnant women from different social, geographical and cultural backgrounds and attending ANC in the public health facilities were given equal opportunity to be enrolling for the study. All health districts in Kunene region participated in the study.

3.10 SUMMARY

This section focused on describing the study design with detailed emphasis on study population and sampling methods. The section also deliberated on data collection and data analysis methods. Ethical consideration for this study was covered. The next chapter is dedicated to the study findings.
CHAPTER 4

4. RESULTS OF THE STUDY

4.1 INTRODUCTION

This chapter is dedicated to data analysis and findings of the study. The main findings of the study are associated risk factors among pregnant mothers and peculiar to Opuwo region. The results are presented as descriptive and analytical statistics in tables, pie charts and graphs according to objectives of the study.

![Flow chart describing the enrollment process]

**Figure 4.1:** Flow chart describing the enrollment process
The study initially selected 122 cases and 247 controls, bringing the total population to 369 study subjects. Because of the 7% nonresponse rate the total number of study subjects ended at 345. From 122 enrolled ANC clients with reactive HBsAg results, 7 were lost to follow up and the total number of clients enrolled were 115, resulting in reduction of number of controls (HBsAg/HBeAg) to 230 is instead of 247. The ratio of controls to cases remained at 2:1 respectively.

4.2 SOCIO – DEMOGRAPHIC CHARACTERISTICS

The researcher interviewed 345 pregnant women that have attended ANC 1st January – 31st August 2017. Table 4.2 below presents the findings of this study on sociodemographic factors associated with HBV infection.

Table 4.2: Socio-demographic characteristics of study population

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>N = 345</th>
<th>Cases n = 115</th>
<th>Controls n = 230</th>
<th>Chi square</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td></td>
<td></td>
<td></td>
<td>7.4147</td>
<td>*0.02</td>
</tr>
<tr>
<td>&lt; 20</td>
<td>45 (13%)</td>
<td>7 (6%)</td>
<td>38 (17%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>20 - 35</td>
<td>252 (73%)</td>
<td>90 (78%)</td>
<td>162 (70%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>36 - 45</td>
<td>48 (14%)</td>
<td>18 (16%)</td>
<td>30 (13%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Place of residence</td>
<td></td>
<td></td>
<td></td>
<td>0.1568</td>
<td>0.7</td>
</tr>
<tr>
<td>Rural</td>
<td>220 (64%)</td>
<td>75 (65%)</td>
<td>145 (63%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Marital Status</td>
<td></td>
<td></td>
<td></td>
<td>1035</td>
<td>*0.00</td>
</tr>
<tr>
<td>-------------------------</td>
<td>-------</td>
<td>-------</td>
<td>-------</td>
<td>------</td>
<td>-------</td>
</tr>
<tr>
<td>Single</td>
<td>195</td>
<td>57 (50%)</td>
<td>138 (60%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(57%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cohabitating</td>
<td>50 (14%)</td>
<td>16 (14%)</td>
<td>34 (15%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Married Monogamous</td>
<td>56 (16%)</td>
<td>17 (15%)</td>
<td>39 (17%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Married Polygamous</td>
<td>44 (13%)</td>
<td>25 (21%)</td>
<td>19 (8%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Education</td>
<td>5.3296</td>
<td>0.1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No formal Education</td>
<td>119</td>
<td>48 (42%)</td>
<td>71 (31%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(35%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Primary</td>
<td>44 (13%)</td>
<td>12 (10%)</td>
<td>32 (14%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Secondary</td>
<td>174</td>
<td>54 (47%)</td>
<td>120 (52%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(50%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tertiary</td>
<td>8 (2%)</td>
<td>1 (0.8%)</td>
<td>7 (3%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ethnicity</td>
<td>9.3117</td>
<td>0.1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Himba</td>
<td>126</td>
<td>46 (40%)</td>
<td>80 (35%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(37%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dhemba</td>
<td>23 (7%)</td>
<td>8 (7%)</td>
<td>15 (7%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Herero</td>
<td>105</td>
<td>36 (31%)</td>
<td>69 (30%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(30%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Oshiwambo</td>
<td>35 (10%)</td>
<td>15 (13%)</td>
<td>20 (9%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Damara</td>
<td>51 (15%)</td>
<td>8 (7%)</td>
<td>43 (18%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>----------</td>
<td>------------------</td>
<td>-------</td>
<td>-------</td>
<td>-------</td>
<td></td>
</tr>
<tr>
<td><strong>Others</strong></td>
<td>5 (1%)</td>
<td>2 (2%)</td>
<td>3 (1%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Employment Status</strong></td>
<td></td>
<td></td>
<td></td>
<td><strong>0.7994</strong> 0.7</td>
<td></td>
</tr>
<tr>
<td>Formal Employment</td>
<td>37 (11%)</td>
<td>10 (9%)</td>
<td>27 (12%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Self employed</td>
<td>42 (12%)</td>
<td>15 (13%)</td>
<td>27 (12%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unemployed</td>
<td>266 (77%)</td>
<td>90 (79%)</td>
<td>176 (76%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Gravidity</strong></td>
<td></td>
<td></td>
<td></td>
<td><strong>4.9993</strong> *0.03</td>
<td></td>
</tr>
<tr>
<td>Primigravida</td>
<td>82 (24%)</td>
<td>19 (17%)</td>
<td>63 (27%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Multigravida (&gt;1 pregnancy)</td>
<td>263 (76%)</td>
<td>96 (83%)</td>
<td>167 (73%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Parity</strong></td>
<td></td>
<td></td>
<td></td>
<td><strong>9.8845</strong> *0.00</td>
<td></td>
</tr>
<tr>
<td>Nulliparous/ at least one live birth</td>
<td>152 (44%)</td>
<td>37 (24%)</td>
<td>115 (50%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Multipara (2 or more live births)</td>
<td>193 (56%)</td>
<td>78 (76%)</td>
<td>115 (50%)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
4.2.1 Age Distribution of Study Population

Figure 4.2.1: Age Distribution of cases and control, ANC attendees January – August 2017

Figure 4.2.1 shows that the majority of hepatitis B infections among the cases were 90(78%) and belonged to the 20-35 years age group, whilst the least number of Hepatitis B infection among the cases were 7(6%) and belonged to the less than 20 years age group. The age group of < 20 years had lowest prevalence of Hepatitis B infection 7% (7). The age group 36-45 years had 16% (18) cases of Hepatitis B infection. The majority, 70 %(162) pregnant women among the control group belonged to the 20-35 years age group, whilst the least, 13% (30) pregnant women among the control group belonged to the 36-45 years age group. The less than 20 year age group had 17% (38) pregnant women among the control group. Mean age among the cases was 29 years ranging from 16 – 45 (SD = 6.6), whereas in controls the mean was 26 years ranged from 13 – 45 years (SD = 6.8). Age was not found to be associated risk factor for Hepatitis B infection (P > 0.05).
4.2.2 Place of Residence

The majority of the study population 64% (220) resided in the rural areas whilst 36% (125) resided in urban areas. The majority 65% (75) of the case group resided in the rural areas whereas 35(40) of the cases resided in the urban areas. Among the control group, 63% (145) pregnant women resided in the urban areas. There was no association found between place of residence and hepatitis B infection (P= > 0.05).

4.2.3 Marital status and Hepatitis B infection

![Marital Status Among Cases and Controls](image)

**Figure 4.2.3: Marital Status among cases and controls, ANC attendees in Kunene Region January – August 2017**

Table 4.2.3 shows that largest number of study population, 57% (195) were single whilst the least 13% (44) were in polygamous marriages. Among the case group, half 50% (57) of the pregnant women were single, 21% (25) were married in polygamous relationships, 17% (17) were married in monogamous relationships and the least 14 % (16) number of pregnant women were cohabitating. As of the control group, the majority 60% (130) of the pregnant women were
single, 17 % (39) were in monogamous marriages, 15 % (34) were cohabitating and 8% (19) were in polygamous relationships. Marital status was not associated with Hepatitis B infection, however study subjects that were married in polygamous unions had significantly higher proportion of cases (p=0.01)

4.2.4 Educational level and hepatitis infection

About 50% (174) of the study population had secondary education whilst 2% (8) pregnant woman had tertiary education. Surprisingly 35% (119) of the study population had no formal education at all. Among the case group, 47% (54) pregnant women had secondary education, 42% (48) had no formal education, 10% (12) had primary education and almost 1% (1) had tertiary education. On the contrary, 52% (120) of the control group had secondary education, 31% (71) had no formal education, 14% (32) had primary education, and 3% (7) had tertiary education. There was no observed association between HBsAg positivity among different educational levels (P=0.1)

4.2.5 Ethnicity and Hepatitis infection

![Ethnicity among Cases and Controls](image)

Figure 4.2.5: Distribution of cases and controls by ethnic group among ANC attendees in Kunene region, January – August 2017
Figure 4.2.5 illustrates that the study population consisted of 37% (126) Himba, 30% (105) Herero, 15% (51) Damara, 35% (Oshiwambo), 7% (23) Dhemba and 1% (5) others. The majority 40% (46) of the case group were Himba, 31% (36) were Herero, 13% (15) were Oshiwambo, the Dhemba and the Damara were 7% (8) apiece while 2% (2) consisted of others. In the control group, the Herero constituted the majority being 35% (80), 30% (69) were Herero, 18% (43) were Damara, 9% (20) were Oshiwambo, 7% (15) were Dhemba and 1% (3) was made up of other ethnic groups. It is also evident that among cases and control group, women from Himba ethnic tribe accounted for 50%, however no statistical association was observed (P=0.1).

**4.2.6 Employment Status and Hepatitis B infection**

The majority, 266 (77%) of the study population were unemployed, 12% (42) were self-employed and 11% (37) were in formal employment. Among the case group, 79% (90) were unemployed, 13% (15) were self-employed whilst 9% (10) were in formal employment. About 76% (176) of the control group were unemployed and 12% (27) were either self-employed or formally employed. Employment status was not associated with Hepatitis B infection in this study.

**4.2.7 Gravidity and Hepatitis B infection**

About 76% (263) of the study population were multigravida and 24% (82) were prim gravida. Among the case group, 83% (96) were multigravida and 17% (19) were prim gravida, on the other hand 73% (167) of the control group were multigravida and 27% (63) were prim gravida. In the preliminary bivariate model, hepatitis B infection between primigravida and multigravida was statistically significant (P = 0.03).
4.2.8 Parity and Hepatitis B infection

As reflected in figure 4.2.8, the majority 56% (193) of the study population were multiparous whilst 44% (152) were nulliparous. Among the case group, 76% (78) were multiparous and 24% (37) were nulliparous, and on the contrary 50% apiece of control group were either nulliparous or multiparous. Among the cases 78% were multiparous women compared to 24% prevalence in nulliparous women (P = 0.00).

4.3 PREVALENCE OF POTENTIAL RISK FACTORS

The potential risk factors were present in both cases and control group. The illustration depicted in figures 4.3 reflects the distribution and frequency of risk factors both in cases and controls.
As reflected in table 4.3, from the interviewed 115 ANC clients with Hepatitis B infection (cases), the most prevalent risk factors were hospital admission 77% (89), tooth extraction or dental procedures 67% (77) and history of STI’s 54% (62). History of at least one home delivery and always delivering at hospital accounted for 42% (48) and 42 (48) prevalence of risk factor. History of abortion 24%, body piercing or scarification 17% (20), HIV positive status 12% (14) and presence of tattoos (17%) had moderate prevalence in the cases. The least prominent risk factors were cesarean section 2% (2), history of blood transfusion 3% (3), and history surgery 10% (12).

Among the 230 controls, history of hospital admission 65% (149), body piercing 50% (115), only delivering at hospital 43% (101) and tooth extraction 40% (92) were the major exposures. Cesarean section 3% (7), tattoos 3% (7) blood transfusion 1% (3), history of surgery 7% (15), HIV positive status 7% (16) were least prominent risk factors.
4.4 BIVARIATE ANALYSIS

The table 4.4 depicts the crude ratio for socio-demographic and other potential risk factors. In all eight risk factors were identified to be associated with Hepatitis B infection in pregnant women and two risk factors were found to be protective.

Table 4.4: Crude and adjusted correlates of HBV infection among pregnant women attending antenatal care in the Kunene Region, January - August 2017

<table>
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<th>Variables</th>
<th>COR</th>
<th>95% CI</th>
<th>P value</th>
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<td>0.14 - 0.76</td>
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<td>1.17 - 3.54</td>
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Table 4.4 shows that in the bivariate analysis age group below 20 years (COR: 0.33; CI: 0.14 – 0.76; p = 0.01), women married in polygamous marriages (COR: 3.08; CI: 1.61 -5.98; p = 0.00), Damara ethnicity (COR: 0.35; CI: 0.14 – 0.71; P= 0.00), body piercing/scarifications (COR: 4.75; CI: 2.74 - 8.20; p = 0.00), history of abortion (COR: 2.52; CI:1.40 - 4.55; p = 0.00), history of STI’s (COR:4.55; CI: 2.79 - 7.41; p= 0.00), history of hospital admission (COR: 1.86; CI:1.11 - 3.11; p= 0.01), multiparous women (COR: 2.10; CI:1.32- 3.37; p = 0.00), presence with tattoos ( COR:6.70; CI:2.74 - 16.40; p = 0.00) and tooth extractions/dental procedures (COR:3.04; CI:1.90 - 4.86; p = 0.00) were risk factors that were found to be statistically significant. However
the pregnant women of age group of under 20 years and women of Damara ethnicity was associated with protective COR’s. Other risk factors such as place residence (urban or rural), cesarean section, educational status, Hepatitis B vaccination, HIV status, home and hospital deliveries, history of surgical procedure and blood transfusion had p > 0.05.

4.5 MULTIVARIATE ANALYSIS (LOGISTIC REGRESSION)

Table 4.2 depicts associations of risk factors that were generated from multivariate analysis. Out of 10 risk factors from univariate analysis, 6 were found were found to be significant predictors of HBV infection (p < 0.05). In this model, statistical association was observed in women in polygamous marriages (AO: 3.45; CI: 1.25 – 9.57; p = 0.02). Body piercing and scarification (AOR: 4.34; CI: 2.30 – 8.17; p = 0.00) and presence of body tattoos (AOR: 2.95; CI: 1.09 - 7.99; p = 0.03) remained the significant predictors for Hepatitis B infection. History of abortion (AOR: 2.91; CI: 1.38 – 6.16; p = 0.00) and STI’s (AOR: 3.34; 95%CI: 1.92 – 5.80; p = 0.00) were found to be significant risk factors. Statistically significant association was observed between previous history tooth extraction or any dental procedure and HBV infection (AOR: 2.03; 95% CI: 1.17 – 3.54; p = 0.01).

Other four risk factors in the multivariate analysis such as under 20 years of age, Damara ethnicity, history of hospital admission and being multiparous was found not be significant in the final model.

4.6 SUMMARY

This chapter covered the presentation of study results. The presentation particularly put emphasis on the identification of significant risk factors for Hepatitis B infection attending ANC in
Kunene region (n=345). This chapter also examined the prevalence of risk factors among the cases and control group. The next chapter discusses the study findings in detail and its relationship with other study findings.
5. DISCUSSION, CONCLUSION AND RECOMMENDATIONS AND LIMITATIONS

5.1 INTRODUCTION

This chapter will look into the interpretation of the main findings of the study. In this section, the findings of the current study will be compared with studies previously done in other countries. In this chapter, research findings are summarized and concluded in line with the study objectives. Based on the concluding points, recommendations will be generated and limitations of the study will be highlighted in this section.

5.2 DISCUSSIONS ON THE FINDINGS

5.2.1 Socio-demographic Factors of Hepatitis B infection

The study showed significant difference in the distribution of cases between in different age groups (P=0.02) in the bivariate analysis. However, after adjusting with other risk a factor in the multivariate model, no associated was observed (P >0.05). The age group with the highest frequency of Hepatitis B infection (HBsAg) was among the cases was observed in the age group 35 – 36 years. Similar findings were observed in Ethiopia study where the age group 25 – 35 years was identified as associated risk factor for Hepatitis B infection (61). The high frequency of hepatitis B could be attributed to the fact that the 20-35 aged group constituted 78% of the entire study population. Furthermore the Kunene Regional Health Information System data shows that age group of 20 – 35 year constitute about 75% of all ANC first visits. In bivariate analysis, the age group of below 20 years had significant lowest prevalence of Hepatitis B infection (14%) and was found to be protective from Hepatitis B infection (P=0.002) but in multivariate analysis, there was no association found between this age group and Hepatitis B
infection (P >0.05). Although in Kunene region early sexual debus are common due to customary practices, it seems the exposure to Hepatitis B infection from potential sex partners are minimal in this age groups. In contrast the study in Uganda appear to demonstrate that the age group below 20 years was associated with Hepatitis B infection among pregnant women (P = 0.00) (59).

In terms of marital status, this study found out that there is association (p=0.00), between HBV infection and polygamous marriages. Since the polygamous marriages involve multiple sexual partners, exposure to Hepatitis B infection is likely. Concurring findings were observed in Kano State in Nigeria where polygamous marriages were identified as significant predictors for Hepatitis B infection among pregnant women ( P = 0.001) (57). On the contrary, another study in Kenya found no significant association between polygamous marriages and Hepatitis B infection(21).

Despite the fact that Himba, ethnic tribe accounted 67% of all HBsAg positive cases, there was no association observed between the two ( P= 0.3). The high frequency of Hepatitis B among Himba women could be attributed to the fact 75% of regional ANC clients were from Opuwo District which is a home to Himba community (108). In the bivariate analysis there were significant differences observed in different ethnicity (P = 0.00), however after adjusting with other variables in the multivariate model, there was no association observed between ethnicity and hepatitis B infection (P >0.5). The study by Getahun and colleagues concluded that tribal ethnicity does not have significant influence on Hepatitis B transmission in the community source (109). Nevertheless, the study in Kano State show significant association between pregnant women of Igbo tribe and Hepatitis B infection (0.017) (57). It is worthy to mention that
pregnant women of Damara ethnicity were found to be protective in the bivariate model (P = 0.00), however in the multivariate model no significant association was observed.

Living in rural or urban area, educational and employment status were not associated with Hepatitis B infection among this study population. These findings are comparable with a study in Ethiopia and Cameroon (72), (110). This is the indication that pregnant women from either rural or urban areas with different educational backgrounds, may have equal risk of exposure to Hepatitis B infection.

5.2.2 Prevalence of Potential Risk Factors among the Study Population

The results of the study indicated that the prevalence of Hepatitis B infection among the study population ranges between 2% - 77% among cases and 1% – 65% among the controls (thought controls were negative for HBV). These findings are an indication that pregnant women in this study irrespective of HBsAg status were exposed to number of potential risk factors for Hepatitis B infection. The prominent risk factors in both cases and controls were history of hospital admission, tooth extractions, and hospital deliveries. However, risk factors such as history of abortion, home deliveries and history of STI’s were more prevalent among pregnant women with seropositive (HBsAg reactive) results for Hepatitis B. These results are corresponding with various studies which identified dental procedures, STI’s, abortion, hospital admissions, and parity as known potential risk factors (57),(59),(61). Yakasai and colleagues argue that pregnancy in itself is not a risk factor for Hepatitis B as all women being pregnant or not have equal exposure to the risk factors (57).

The prevalence of some of the potential risk factors can be reduced by infection control in hospital setting and homes and Hepatitis B vaccination for the general population. In order to
mitigate these exposures, WHO guidelines recommend for universal coverage of Hepatitis B prevention strategies such vaccination, routine screening of blood donors, infection control in high risk settings and use disposable body puncturing equipment in hospital setting (96). Although these findings are reflecting high prevalence of some of the known potential risk factors, the study went further to determine the significance of associations between identified exposures and Hepatitis B infection.

5.2.3 Associated Risk Factors

Several risk factors for contracting hepatitis were evaluated during the study period. In bivariate analysis the age group below 20 years, women married in polygamous marriages, Damara ethnicity, body piercing/scarifications, history of abortion, history of STI’s, history of hospital admission, multiparous women, presence with tattoos and tooth extractions/dental procedures were found to be statistically associated with Hepatitis infection (p<0.05), however when these factors were put in logistic regression model and adjusted for other factors, only six factors including polygamous marriage, body piercing/scarification, history of abortion, history of STI’s, body tattoos and dental procedure were found to be associated risk factors for HBV infection among the study population. The results of this study agrees with numerous studies carried out where abortion, STI’s, body scarification and dental procedures were found to be significant risk factors for Hepatitis infection (90),(70),(111)(77). The study by Nyamusi and colleagues had opposing findings where risk factors such as body scarification, tattoos and history of STI’s were not associated with Hepatitis B infection (112).

The adjusted odd ratio in multi-variate analysis revealed that pregnant women who had body piercing or scarification were 4 times more likely to be positive for HBsAg (AOR: 2.3; 95% CI:(2.30-8.17); P= 0.00). In Kunene region, body scarification practice is routinely done as
traditional ritual among Himba and Dhemba ethnic tribes. Body piercing is done for cosmetic and occasionally for traditional purposes. The significance of this risk factor can be explained by the fact that many of these procedures are done in communities under unsterile environment, living room for horizontal transmission of Hepatitis B infection. The body scarification and piercing is widely practiced throughout Africa. The study in Nnewi, Nigeria confirms these findings where tribal scarifications among the pregnant women was associated with Hepatitis B infection (P =0.001) (65). Similar study results were found in Moshi Municipal Clinic, Tanzania (P= 0.006) (113). Although the study found significant association between Hepatitis B infection and this scarification practice, the study by Mohammed seems to suggest otherwise where body scarification and piercing was found to be insignificant predictor for Hepatitis B infection. (P > 0.05). In this study, all the subjects were female, which may be biased, as females were more likely to be pierced than men.

In the multi-variate model, pregnant women with history of abortion were 3 times more likely to be positive for HBsAg (AOR: 2.91; 95% CI : (138-6.16); P-value: 0.00). These findings agree with study in Ethiopia which identified abortion as predictor for Hepatitis B infection among the pregnant women attending ANC ( P = 0.017) (61). Yakasai and colleagues indicated that the association of HBV infection and abortion could be related to the fact that abortion is directly related to sexual activity and sexually active women have a higher chance of getting the infection (57). The probability of acquiring Hepatitis B infection increases with unsterile procedures associated with induced abortion and management of post abortion complication. On the other hand, Frambo and colleagues demonstrated in their study that history of abortion is not an associated risk factor of Hepatitis B infection (114). According to Alegbeleye and colleagues,
policies aimed at reducing the incidence of unsafe abortions and promotion of barrier contraception in the environment may assist in reducing the incidence of Hepatitis infection (63). In this study the adjusted odds ratio revealed that pregnant women with history of STI were 3 times more likely to have reactive results for HBsAg (AOR: 3.34; 95% (1.92 – 5.50); P = 0.00). These findings correspond with studies in North Sudan where STI was observed as significant predictor for Hepatitis B infection (P = 009) (115). Since STI is associated with unsafe sex, the probability of acquiring Hepatitis B infection through unsafe sexual contact seems also to be increased. The study in Rwanda have contradicting results as STI was not found to be significantly associated with Hepatitis B infection (P = >0.05) (112).

Despite the low prevalence of tattoos among the study population, the multivariate model reflects that pregnant women with tattoos were 3 times likely to have Hepatitis B infection (AOR: 2.95; 95%CI: 1.09 – 7.99; P = 0.03).Similar findings were observed in China and in Cameroon (116),(110).The tribal tattooing which is practiced in many part of Africa including Kunene region in Namibia is commonly associated with sharing of unsterilized equipment which creates opportunities for horizontal transmission of Hepatitis B infection. Despite these observations, the study in Kano State, Nigeria and in Palestine found opposing results where tattoos were found not be associated with Hepatitis B infection. (57), (38). The risk of acquiring Hepatitis B infection through tattoo procedures is likely to be reduced with single use of sterile piercing equipment.

After adjusting with other risk factors, history of tooth extractions or any other dental procedures were found to be significant predictors for Hepatitis B infection among pregnant women in this study (AOR: 2.03;95%CI:1.17 – 3.5;P = 0.01). The high prevalence of tooth
extraction among the study population was attributed to traditional removal of left and right mandibular central incisors which is practiced by Himba, Herero, Dhemba and other minority ethnic groups that live in Kunene region. This practice is conducted in the community under unhygienic conditions and sharing of equipment is common. The study by Molla and colleagues revealed a significant association between tooth extraction and Hepatitis B infection among the pregnant women (72). Mahbobbi and colleagues argue that contaminated sharp instruments, such as needles, lancets, scalpels, broken glass, specimen tubes and other instruments, can transmit blood-borne pathogens such as HBV(117). Opposing results were observed by Ayele and colleagues where dental procedures where not associated with Hepatitis B infection (118).

Surprisingly this study revealed that some of the risk factors which are known to be potential predictors for Hepatitis B infection were found to be insignificant. The risk factors such as HIV+ status and blood transfusion were not identified as risk factor.

An HIV positive status was not associated with Hepatitis B infection in this current study (COR: 1.85; 95%CI: 0.87 – 3.94; P = 0.01). This observation was comparable with a study in Uganda where HIV status was not associated with HBsAg seropositivity (P= >0.05). Similar results were obtained in Kwazulu-Natal in South Africa(92). However, the study in Dar es Salam in Tanzania observed the significant association between HIV positive status and Hepatitis B infection(119). According to 2016 HIV Namibia ANC Sentinel Survey report, HIV prevalence among pregnant women in Kunene region was 12% while ART coverage was 60% percent(120). Despite such high prevalence, HIV positive status seems to not have influence on HBsAg positivity among pregnant women in Kunene Region.
Blood transfusion was not found to be associated with Hepatitis B infection in this study (P = >0.05). Similar results were obtained in Brazil and Tanzania (39),(116). Nevertheless, the results obtained from studies in Yemen and Cameroon seems to disagree as blood transfusion was found to be associated with Hepatitis B infection in both studies (74),(76). In addition, quality standards for blood safety put which in place by Namibia Blood Transfusion Services minimizes the risks (42). Despite this observation, the results from this study are likely to be biased as only 1.7% of the study population was exposed to blood transfusion.

5.3 CONCLUSION

In conclusion, it is worth noting that various known Hepatitis B risk factors were identified in this study. Under Sociodemographic Hepatitis B risk factors in Kunene region, the bivariate analysis shows that age group below 20 years and pregnant women of Damara ethnicity were found to be protective. However these associations were found to be insignificant in the final bivariate model.

Based on the study results, the significant risk factors (p<0.05) for Hepatitis B infection among pregnant women attending antenatal care in Kunene region were polygamous marriages, body piercing/scarification, history of abortions, history of STI’s, body tattoos and tooth extraction or any other dental procedures. These results indicates abortion (P = 0.00) and sexually transmitted infection (P = 0.00) to be the predominant predictors for Hepatitis B among pregnant women attending Antenatal Care in Kunene region.
5.4 STUDY LIMITATIONS

This study suffered from several limitations as described below:

- The study was health facility based and only covered pregnant women who have attended ANC in public health facilities in Kunene Region.
- The study design was unmatched case control. As such, cases were not matched with controls by age, residential area or other variables.
- Although the initial proposal included Omusati region, the proposed population could not be included in this study as Hepatitis screening at ANC clinics in that region is not routinely done. (Appendix F)
- Hepatitis B results were not available among some potential study subjects, resulting in exclusion from the study.

5.5 RECOMMENDATIONS

From the above presented conclusions in section 5.3 various recommendations are presented in accordance with the objectives and findings of the study.

Seeing that women married in polygamous unions are more at risk of acquiring hepatitis B infection particularly in Himba ethnic group, the Ministry of Health and Social Services in Kunene region can design targeted preventative strategies such as Hepatitis B family based screening, treatment care and support.

The Regional Health Promotion Division in Kunene region should promote single use sterilized body piercing and tattoo equipment and discourage people to undergo these procedures under unsafe environment.
The Community Liaison Officers can hold massive health promotion within the communities on hepatitis and safe infection control procedures.

The government may also consider provision of sterile body piercing and tattoo equipment.

Exposure from infected dental health practitioners or contaminated equipment can be mitigated with routine hepatitis B screening, provision of Hepatitis B vaccination and adherence to infection control standard operating procedures in the health facilities.

Ministry of Health and Social Services have to conduct the community awareness and health education on promotion of Reproductive Health which aims at preventing unwanted pregnancies that might lead to abortion and subsequent exposure to HBV.

The sexually acquired Hepatitis B infection can be minimized if local health facilities personnel promote of safer sexual intercourse and provide screening and treatment in the general population.

5.6 SUMMARY

This chapter presented the interpretation of the main findings of the study based on the specific objectives. The study revealed that women married in polygamous unions, body piercing or scarifications, history of abortion, history of sexually transmitted infections, tattoos and tooth extractions were strongly associated with Hepatitis B infection. These findings provide justifications for Ministry of Health in Kunene region to implement preventative strategies which targets the identified significant risk factors. This chapter further discussed the conclusion, limitations as well as the recommendations.
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APPENDIXES

Appendix A: Participant Information Sheet and Consent Form

My name is…………………………I am working as a data collector for the study being conducted in this hospital by Ms. Iyaloo Mwaningange who is studying Masters in Applied Field Epidemiology with University of Namibia. I would kindly ask you to spare some time and pay attention while I am explaining about the study.

The study title is “Risk Factors Associated with Hepatitis B Infection among Pregnant Women Attending Antenatal Care in Kunene Region”.

The aim of the study is determine the risk factors which could be responsible for transmission of Hepatitis B infection in Pregnant Women in Kunene Region. The study will provide information to the health care providers on the associated risk factors for Hepatitis B infection among pregnant women who have attended ANC visits in the selected health facilities. The outcome of this study will alert the health care providers and managers about the importance of routine HBsAg screening and improve in the provision care for infected mothers and their children.

You were randomly selected to participate in this study. Once you have agreed to enroll in the study, I will be interviewing you using a questionnaire to provide me with information that is helpful for the study and your ANC records will be used to provide additional information including the results for Hepatitis B and HIV results.
You will not be required to undergo any medical examination or procedure for this purpose. There would not be any payment given to you however by participating in this study, you are contributing to the wellbeing of expectant mothers and their children.

Your name and patient identity number will not be written, disclosed and will never be linked with any of the information you are going to provide.

If you happen to be uncomfortable with answering some questions, you are at freedom not to answer and you may withdraw from this study at any time you want to. If you permitting to be interviewed, you can sign or stamp in the area indicated below.

If you have any question please you can contact me by phone mobile …… or contact your nearest health facility or you can call Research Unit in the Ministry of health and Social Services, Tel: 0612039111

Declaration of informed voluntary consent:

I have read/ was read to me the participant information sheet. I have clearly understood the purpose of the research, the procedures, and the benefits, issues of confidentiality, the rights of participating and the contact address for any queries. I have been given the opportunity to ask questions for things that were not clear. I was informed of my rights such as: I have the right to withdraw from the study at any time and I am not obliged to answer any question that I find to be uncomfortable to answer. Therefore, I hereby give consent to participate in this study with my initials (signature) as indicated below.

Signature of participant: _______________  Signature of data collector _______________
Transferred Consent forms

**Otjiherero Language**

**OMASURIRO UEHURI**

Orutuu o A. Orutuu Ruondjivisiro Jomunarupa nauina Orutuu Ruomajandjerero.

Ena randje ouami ...............mbiungura otjomuoronganise kondononeno ndjimaikaendisisua morukondua ruoje ijo Omuozikua Iyaloo Mwaningange ngurihonga o Masters Matlabjuru Uomitjise ponganda jomerihongero uokombanda ndji ja Namibia. Mekuningire kutja undjipe ombango ngunda Ami amehandjaura ohunga nongondononeno ndji.

Ongondononeno jahakua nai “ Ovinenge Mbyakutua Komuahuangero Uomasuriro Uehuri nu tjinene Kovakaendu Ovatumba Mbejenda Kotuveze Tuomatarero mo Rukondua rua Kunene nauina Omusati.”

Ondando jongondononeno okuzikamisa oviune mbiri oumba mbimavituara komahuangero Uomasuriro Uehuri B Movakaendu Ovatumba motukondua tua Kunene nauina Omusati. Ondjiri, ongondononeno ndjimaijdandja ondjvisiro kovahunge koumba mbuakutua kovinenge oumba mokati kovakaendu ovatumba mbaja Komatarero kozo Klinika zetu. Oumune uongondononeno ndji maujandja ondjvisiro kovaungure uouveruke nauina ovanane ounandengu uomatarero uo HBsAq nauina okujandjera omaunguriro oomaa kovakaendu mbeno omutjise nauina ovanatje vao.
Imbui omuano ove mbuuatoor eru a kutja ukare norupa mongondononeno ndji. Ove tjiuai kumu ekuritjangi sa mongondononeno, Ami mekara nomapuiriro kove meungurisa orutuu ruomapuiriro kutja undjipe ondjivisiro ndjimajuungirisiro ua kongondononeno nauina omatjangua uomatarero maejandja ondjivisiro oueziua mumuna ondjivisiro ko Masuriro Uehuri B nauina omatarero uozonongo Kondui Jehinga.

Konakuhepa okukatareru a ouveruke uoje poo okukaenda momirari kondando ndji. Kapena otjimariva tjiomatjijandjua kove mokukara norupa mongondononeno ndji, nguari mojandja ohambuarakana kongaro ombua jovakaendu ovatumba novanatje vao.

Ena roje karina kutjangua omuano mburiri nauina karina kuungurisiua mukangamua ondjivisiro ndjimondjiraere. Una oujara okuhina kuzira nguamua epuriro ndiuhi kuvanga okuzira. Ndovazu uerimunu kutja konakurimuna naua ngunda oua orupa mongondononeno mojenene okurinanunununa tjamua oruvez e tjiuavanga nao.

Tjiuna epuriro poo ongendo ohunga nongondononeno, mohakaene naami konomora jokaendjezeua nga ……….poo hakaena noruze ruouveruke nduri popezu naove poo motono Poruvez Ruongondononeno mo Ministry Jouveruke nauina Ondunino Jotjiuana, ponomora 0612039111.

Omeriraisiro uomurijandjere:

Mbalese/ mbalese nauina erizuvisa

Omunue uomuna rupa: ________________ Omunue uomuuonge uondjivisiro ________________
Damara Language

ǁKHOIKHOSA !HARODI ǂGAO-IOB TAMAS KA IO HEBATITIS B-ǂAEB DIDI HĪA
ǁORAS AI!ā HĀ !KHOǃHOMIS DIDE ǂA!NĂS KUNENE TSĪ OMUSATI ǁKHRIKHA
!NĂS ǁHOBA !KHODANAS DI ǂANSA HARES

ǁHao-a-o-i di ǂAnt'ans ǂKhanib tsī Mâ-ams ǂHaweb

Ti ǂons ge a…………………..Tita ge ǂandi hare-aose ne hares di saogub hīa sa
ǁkharib !nā, !Gōahesas Iyaloo Mwaningange’s, hīa ǁGawi ǀkhalkhasens di ǂhaweb,
Epidemiology timi ǂansaba Namibia’b di ǁGawi ǀKhalkhasens-ǃkhaib tawa ra ǁKhalkhasens
xa ra !khodanaheba ra sīsenba. Tita ge kaise !gam!gamsenxase ra ǂgantsi ǂgao, īts ne hares
di sīsen-!]gauba ta ra mī!a soab !nā ǂorisase !gā.
Ne ǂansa hares di ǂgaiǂams ge “ǁKholkhosa !Harodi, hīa ǂGao-lob tamas ka io Hepatitis B-
ǂaeb, ǂGamǀkha Tare-khoedi hīa ǁOras ai!ā !Khoǃhomisa Kunene tsī Omusati ǁKharikha !nā
ra !Khoǃoadi !nāu hādi ǂkha !gælaresa.” timi hā.
Ne ǂansa hares ge ǀkholkhosa !harodi ai a !am-māïsa, hīa Hepatitis B’s di taniǃkharus,
ǂGamǀkha Tare-khoedi !nā-us, Kunene tsī Omusati ǀkharikha !nās di !reamxasib ase a
ilkhasa. ǀNas ǀkhas xases ge ne ǂansa haresa !gasasiba ǂurusib !koǃhomí-aona nira ǀlkha-
kaiba, ǀkholkhosasiba uhā !harodi, ǂgamǀkha tare-khoedi, hīa ANCǃkoǃhomide, sida di
ǂurusib !khain tawa saris ǂkha ge a !khoǃoadi !nā-u hāse. Ne ǂansa hares di !nurib ge
ǂurusib !koǃhomí-aon tsī ǂgælguí-aon tsīna nira ǂan-kai, HBsAg’n di sisen-]gaub !kholna-
|gaub di nâhâsasib tama tsî nâhâbasa !khoîhomisa, lnâti-i |aeba uihâ lgûdi tsî lîdi di |gôarona nira mâ lkha.

|Nas ge a !aroma, sadu tâgâ!nàgurase ge a lhwûïtuihesa, îdu ne tansa hares di saogub !nâ lhao. Sats ne tansa hares di saogub !nâ xoalâgasensa ni mî|guis ka, o ta ge satsa lgamlaresa nira dîdi |gâu!nâ-u nira uhâ-u, tita ne tansa hares di saoguba hûib ase iba lkha !gasasiba nira ho lkhaše, tsîb ge sa loras ai!â hâ !khoîhomis di !nuriba, ha|arora !gasasib, hîa Habatitis B’s tsî HIV’s ôa!nâ-khaib tsîra di !nuriba uhâba nira ilkha-kaiba.

Sats ge |guis khemi i !âi-i tamas ka io kaxu!nâ sîsen-[gau-i !nâ ne soab !nâ lhaosa i|ganhe tite. Nausa i ge |guis khemi matare-e satsa mâhe tite, ne tansa hares di saogub !nâ lhaos !aroma, sats ge ñas !nâ, |gam|kha lgûdi tsî lîdi di |gôaron di ûrusib tsî !gâise hâs tawa ra |aro.

Sa |ons ge xoa-mâïhe tite tsî tatses tsîna |guis khemi i xu-i !aroma, lnats ta mâte !gasasib soab !nâ sîsen-uhe tite. Sats ge lkhâiti a !norasa, !eream tâgo tamats ka i di-e !eream tama isa. Sats !norasase, ne tansa hares di saogub soab !nâ ni tsâ tama is ka, ots ge mâ-i ka a lae hîats ta !gao-i ai, o a !gae-oasen lkha.

Sats mâ-i ga a di tamas ka io !âi-thânsen-au-e ni uhâs ka, ots ge saora ti tânîmâhera lâlawa!gôas, hîa a ……….tawa a lkhamibate lkha tamas ka io sa |guse mâ ûrusib !khai-e a lkhami lkhami lkha tamas ka io ôa!nàdi !âb, û Unidos tsî |Hûhâsib !Oabadi Ministeries !nâ, lâlawa!gôas 0612039111 tawa a lkhami lkha.

!Gara!a, !làn!ansa mãsenxa mâ-ams dis:

Tita ge khom-ai tsî go a !lànlanhe

Tita ge go khom-ai tamas ka io go a khom-aîbahe, ne lhaoaon di ñkhaniba. Tita ge jôasa |nàu!asa ne ôa!nâs di tâïbasens ai uhâ, sîsen-[gaugu tsî domdoredi, sâusasib di màsib,
Hao-ao i di xoa!gaos: ______________  Ans Hare-ao-i di xoa!gaos: ______________

Appendix B: ôa!nâs Di Dî-ai !Haweb

Dî-ai !haweb di !Gôas (!Haris di !gôas!/Khaib di !gôas/sîsenlare-ao-i di !gôas 3 !gôadi |kha______

|Gamlares di Tses ____________

|Gamlare-ao i di |Ons____________
Appendix B: Research Questionnaire

Questionnaire Number 3 digits:

Date of interview _____________

Interviewer Name _____________

Health Facility: _____________

Case/ Control _____________

A: Socio-demographic information

1. How are you old now (in years)? _______________

2. Where are you living? (Tick only one)  □ Urban (1)  □ Rural (0)

3. What is your marital status? (Tick only one)  □ Single (1)  □ Cohabitating (2)

□ Married Monogamous (3)  □ Traditional Polygamous (4)

4. What is your educational Level (Tick only one)

□ No formal education at all (1)

□ Primary education (1-7 grades) (2)

□ Secondary education (8-12 grades) (3)
5. What is your ethnicity? (Tick only one)

☐ Himba (1) ☐ Omudhemba (2)
☐ Herero (3) ☐ Oshiwambo (4)
☐ Damara (5) ☐ Others______________ (6)

6. What is your occupational status? (Tick)

☐ Employed in formal sector (1) ☐ None - formal employment (2)
☐ Unemployed (3)

B: Gynecological /Obstetric history (Write or Tick)

7. How many times you have been pregnant (including current pregnancy)? _____

8. Question number 8.1 and 8.2

8.1 How many life births do you have? _____

8.2 How many still births did you have? _____

9. Have you ever delivered at home by traditional birth attendant? ☐ NO (0) ☐ YES (1)
10. Have ever delivered in the hospital? [ ] NO (0) [ ] YES (1)

11. Have you ever delivered vaginally [ ] NO (0) [ ] YES (1)

12. Have you ever delivered through caesarean section [ ] NO (0) [ ] YES (1)

13. Do you have a history of abortion? [ ] NO (0) [ ] YES (1)

<table>
<thead>
<tr>
<th>C: General Risks for Hepatitis B virus infection (Please tick only one)</th>
</tr>
</thead>
<tbody>
<tr>
<td>14. Did you have body piercing practice/Scarification? [ ] NO [ ] YES</td>
</tr>
<tr>
<td>15. Did you have body tattoos? [ ] NO [ ] YES</td>
</tr>
<tr>
<td>16. Did you ever received blood in the hospital? [ ] NO [ ] YES</td>
</tr>
<tr>
<td>17. Where you ever hospitalized? [ ] NO [ ] YES</td>
</tr>
<tr>
<td>18. Do you have history of any dental procedure? [ ] NO [ ] YES</td>
</tr>
<tr>
<td>19. Do you have history of surgical procedure [ ] NO [ ] YES?</td>
</tr>
<tr>
<td>20. Do you have previous history of sexually transmitted infection? [ ] NO [ ] YES</td>
</tr>
</tbody>
</table>
Appendix C: ANC Record Data Abstraction Form

Questionnaire Number (District number/Facility number/client number with digits):

_____________

Date of data collection: ___________

Name of the Data collector: ___________

______________________________________________________________________

1. HBsAg results: 
   - Reactive
   - None-Reactive

2. Presence of HBeAg and results: 
   - Reactive
   - Not reported/Detected

3. HIV test results: 
   - Reactive
   - None-Reactive
   - Unknown

4. Hepatitis B vaccination status: 
   - Vaccinated
   - Not vaccinated
   - Unknown vaccination status
CENTRE FOR POSTGRADUATE STUDIES
University of Namibia, Private Bag 13301, Windhoek, Namibia
340 Mandume Namuthiya Avenue, Pionwies Park,
Tel: +264 61 206 3275/4652; Fax +264 61 206 3290; URL: http://www.unam.edu.na

RESEARCH PERMISSION LETTER

Student Name: Iyakoo Wilka Mwaningange

Student number: 9419691

Programme: Masters of Science in Applied Field Epidemiology

Approved research title: Examining the risk factors of hepatitis B infection among antenatal care attendees in Kunene and Omusati regions, a case control study

TO WHOM IT MAY CONCERN

I hereby confirm that the above mentioned student is registered at the University of Namibia for the programme indicated. The proposed study met all the requirements as stipulated in the University guidelines and has been approved by the relevant committees.

The proposal adheres to ethical principles as per attached Ethical Clearance Certificate. Permission is hereby granted to carry out the research as described in the approved proposal.

Best Regards

[Signature]

Name: Dr Marius Hedimbri
Director: Centre for Postgraduate Studies
Tel: +264 61 2063275
E-mail: directorpgs@unam.na

23/06/17

Date
Appendix E: Permission Letter from Ministry of Health and Social Services

REPUBLIC OF NAMIBIA

Ministry of Health and Social Services

Private Bag 13198
Windhoek Namibia

Ministerial Building
Harvey Street
Windhoek

Tel: 061 – 2032150
Fax: 061 – 222358
Email: shinemhipangelwa71@gmail.com

OFFICE OF THE PERMANENT SECRETARY

Ref: 17/3/31M
Enquiries: Mr. J. Nghipangelwa

Date: 01 August 2017

Ms. Iyaloo Mwaningange
P.O. Box 238
Mariental
Namibia

Dear Ms. Mwaningange

Re: Examination of risk the factors of hepatitis B infection amongst antenatal care attendees in Kunene and Omusati Region.

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 purposes;
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;

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3.6 Final report to be submitted upon completion of the study;
3.7 Separate permission should be sought from the Ministry of Health and Social Services for the
publication of the findings.

Yours sincerely,

[Signature]

Andreas Mwoombola (Dr)
Permanent Secretary

"Health for All"
APPENDIX F: LETTERS FROM KUNENE AND OMUSATI REGIONAL HEALTH DIRECTORS

Republic Of Namibia

MINISTRY OF HEALTH AND SOCIAL SERVICES

KUNENE REGIONAL DIRECTORATE
MIS/ Disease Surveillance and Response unit

Private Bag 3003
Opuno
Namibia

Tel: 065-272846
Fax: 065 273022

Enquiries: Mr. M.S. Shikongo

Office of the Director

All: Senior Medical Officers

Opuno District
Outjo District
Khorixas District

Attn: All Primary Health Care Supervisors

Ms. H. Shikwambi - Opuno District
Ms. R. Amunyela - Outjo District
Ms. A. Day - Khorixas

CC: All District NIP Laboratories

Authorization for the Upcoming Study on Hepatitis B Infection Among Pregnant Women Attending Antenatal Care in Kunene Region During the Period of January 2016 - August 2017

Approval has been granted to Ms. Iyalo Mwaningange a Field Epidemiology resident to conduct the above-mentioned study in the health facilities in Kunene Region. This study aims to examining the risk factors which might be associated with acute (HBsAg) and chronic (HBeAg) HBV infection among pregnant women attending antenatal care in Kunene region.

It is in this vein that we are requesting your good office to render necessary support by permitting her to access the ANC records and interview the clients in the designated PHC facilities and maternity wards. The study is expected to enroll 122 pregnant women with positive results for Hepatitis B infection (Reactive to HBsAg/HBeAg) and 243 pregnant women with negative results for Hepatitis B.
Please take note that actual data collection will commence from 14th August to 30th September 2017. Supervisors are hereby requested to bring this directive to attention of health care providers working in the designated health facilities. See the attached list of the sampled health facilities and the authorization letter from the Permanent Secretary. For detailed information do not hesitate to contact Ms. Iyaloo Mwaningange at mobile: 0818044479 or by email: iyaloowilhka@gmail.com

Your efforts are highly appreciated.

Yours sincerely

Mr. T. Shapumba
Regional Director
Kunene Regional Health Directorate
REPUBLIC OF NAMIBIA
Ministry of Health and Social Services

Private Bag 504  
Outapi  
NAMIBIA  

Enquiries: Ms L. Kornelius  

Directorate: Omusati Region  
Tel: 065-251820  
Fax: 065-251071

Ms Iyaloo Mwaningange  
Student: Masters of Science Applied Field Epidemiology  
University of Namibia

03 October 2017


1. Your letter on the above subject matter is hereby acknowledged.
2. We wish to inform you that Omusati Health Directorate has only introduced Hepatitis B screening among pregnant women as from the beginning of July 2017, such that it is not advisable to include them in your study population.
3. Please accept our humble apology and all the best in your endeavors.

Thank you very much for understanding.

Sincerely yours,

ALFONS AMOOM (MR)  
REGIONAL DIRECTOR

"Health for All"