SCHOOL OF PUBLIC HEALTH
COLLEGE OF HEALTH SCIENCES
UNIVERSITY OF GHANA

MATERNAL FACTORS ASSOCIATED WITH HEPATITIS B TRANSMISSION TO NEONATES; A STUDY SELECTED ANTENATAL CLINICS IN THE SEKONDI TAKORADI METROPOLIS AND EFFIA KWESIMINTSIM MUNICIPALITY, 2019.

BY
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THIS DISSERTATION IS SUBMITTED TO THE UNIVERSITY OF GHANA, LEGON IN PARTIAL FULFILMENT OF THE REQUIREMENT FOR THE AWARD OF MASTER OF PHILOSOPHY IN APPLIED EPIDEMIOLOGY AND DISEASE CONTROL DEGREE.

JULY, 2019
DECLARATION

I CHRISTABEL AYEPAH hereby declare that with the exception of the references made to other peoples' work which I have duly acknowledged, this proposal which is my original work has neither in whole nor in part been presented to the University or elsewhere for another degree.

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(SUPERVISOR)
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ABSTRACT

**Background:** Neonatal Hepatitis B viral (HBV) infection is a public health concern worldwide. Prevalence of chronic HBV infection in Ghana is in excess of 8.0%, and was 2.3 per 100,000 in Sekondi Takoradi in 2017. The purpose of the study was to determine maternal factors associated with transmission of Hepatitis B to newborn babies.

**Methods:** A cross-sectional study involving pregnant women was carried out at antenatal units of selected hospitals in the Sekondi Takoradi Metropolis. Hepatitis B positive pregnant women of 34-37 weeks gestation were recruited into the study. Demographic information was collected from the mothers and a review of their antenatal record books performed to extract data on their medical and obstetric history. Maternal venous blood was collected and Hepatitis B viral load and e-antigen level assessed. Cord blood samples were collected and tested for Hepatitis B virus infection in the babies.

**Results:** Eighty-nine Hepatitis B positive mothers had their babies screened for HBV. Prevalence of neonatal HBV was found to be 17.98% (16/89). Birthweight of infected babies was significantly higher than the uninfected babies (3.33kg vs. 3.12kg, p-value=0.043). Viral load (935.18 IU/mL) of mothers whose babies were not infected was lower compared to viral load (4178.63 IU/mL) of mothers of the infected babies (p-value <0.001). Prevalence of neonatal infection of hepatitis B e-Antigen positive mothers was higher (81.82% vs. 31.82%, p-value 0.007). There was no significant association between mode of delivery of the baby and its infection status.

**Conclusion:** Mother-to-child transmission of (HBV) in this study occurred in babies with higher birthweight, and whose mothers had high Hepatitis B viral DNA and were HBV e-
Antigen positive. Hence, HBV DNA levels, HBV profile and use of antivirals, be incorporated into routine Antenatal services in Sekondi Takoradi.
# TABLE OF CONTENTS

DECLARATION ........................................................................................................................ i
ACKNOWLEDGEMENT .............................................................................................................. ii
ABSTRACT ................................................................................................................................ iii
LIST OF TABLES ........................................................................................................................ viii
LIST OF FIGURES .................................................................................................................... ix
LIST OF ABBREVIATIONS ....................................................................................................... x

CHAPTER ONE ......................................................................................................................... 1
INTRODUCTION ........................................................................................................................... 1
  1.1 Background ....................................................................................................................... 1
  1.2 Problem Statement .......................................................................................................... 3
  1.3 Conceptual Framework ................................................................................................. 5
    1.3.1 Narrative of Conceptual Framework ........................................................................ 5
  1.4 Justification .................................................................................................................... 6
  1.5 Research Questions ........................................................................................................ 7
  1.6 Objectives ....................................................................................................................... 8
    1.6.1 General Objective: .................................................................................................... 8
    1.6.2 Specific Objectives: ............................................................................................... 8

CHAPTER TWO ............................................................................................................................... 9
LITERATURE REVIEW ................................................................................................................... 9
  2.1 Etiology and Pathology ............................................................................................... 9
  2.2 Virological Characterization ........................................................................................ 11
  2.3 Risk Factors .................................................................................................................. 12
  2.4 Transmission ................................................................................................................ 13
    2.4.1 Prenatal Transmission ............................................................................................ 14
    2.4.2 Natal Transmission ................................................................................................ 17
    2.4.3 Postnatal Transmission ......................................................................................... 17
  2.5 Diagnosis and Staging ................................................................................................... 19
  2.6 Prevention ...................................................................................................................... 20
    2.6.1 Screening ............................................................................................................... 21
    2.6.2 Prevention through Vaccination ............................................................................. 22
2.7 Natural History of Chronic Hepatitis B ................................................................. 23
2.7.1 Phases of Chronic Hepatitis B ........................................................................... 23
CHAPTER THREE ..................................................................................................... 26
METHODS ................................................................................................................ 26
3.1 Study Design ......................................................................................................... 26
3.2 Study Area ............................................................................................................. 26
3.3 Variables ............................................................................................................... 29
3.3.1 Outcome variable .............................................................................................. 29
3.3.2 Independent Variables (Table 1) ..................................................................... 30
3.4 Sampling ............................................................................................................... 32
3.4.1 Study Population ............................................................................................... 32
3.4.2 Sample Size Determination ............................................................................. 32
3.4.3 Eligibility Criteria ............................................................................................. 33
3.4.4 Sampling Procedure ........................................................................................ 34
3.5 Data Collection Technique, Tools and Instruments ............................................. 35
3.5.1 Training of research Assistants ....................................................................... 36
3.5.2 Hepatitis B Surface Antigen Screening Procedure in Neonates ..................... 37
3.6 Data Quality Control ............................................................................................ 39
3.7 Data Analysis ......................................................................................................... 41
3.8 Ethical Considerations ........................................................................................... 41
3.8.1 Voluntary consent/ Withdrawal .................................................................... 42
3.8.2 Possible Risk and Discomfort ........................................................................ 42
3.8.3 Possible Benefits ............................................................................................... 43
3.8.4 Confidentiality .................................................................................................. 43
3.8.5 Compensation .................................................................................................... 43
3.8.6 Choice of Participation ...................................................................................... 44
CHAPTER FOUR ...................................................................................................... 45
RESULTS ............................................................................................................... 45
4.1 Demographic Characteristics of Mothers in the Study ....................................... 45
4.2 Other Medical Conditions of the Mothers ............................................................ 46
4.3 Background Characteristics of Babies Delivered by the Hepatitis B Positive Mothers ... 46
4.4 Association Between the Infection Status of Babies and Mother’s Demographic Characteristics

4.5 Association Between Obstetric and Family Characteristics of Hepatitis B Positive Mothers and the Infection Status of their Babies

4.6 Differences in Means of Continuous Variables by the Infection Status of the Baby

4.7 Equality of Means of Viral Load of Mothers and the Infection Status of their Babies

4.8 Association Between Infection Status of Baby and the Hepatitis B Envelope-Antigen Status of Mother and Mode of Delivery of Baby

4.9 Binary Logistic Regression Analysis of Mode of Delivery and Hepatitis B e-Antigen on Positive Infection Status of Baby

CHAPTER FIVE

DISCUSSION

5.1 Prevalence of infections among Babies born to Hepatitis B positive mothers

5.2 Association between Obstetric and family characteristics of Hepatitis B positive mothers and the infection status of their babies

5.3 Association between Maternal Viral Load and Babies Born to Hepatitis B-Positive Mothers

5.4 Association between Maternal Hepatitis B e-Antigen Positive and Babies Born to Hepatitis B-Positive Mothers

5.5 Association between Mode of Delivery and Babies Born to Hepatitis B-Positive Mothers

5.6 Limitations

CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusion

6.2 Recommendations

REFERENCES

APPENDICES

Appendix I: Questionnaire

Appendix II: Information on The Study

Appendix III: Certificate of Consent
LIST OF TABLES

Table 3.1: Independent Variables ..........................................................................................30
Table 3.2: Operational Definition of Independent Variables ..............................................30
Table 4.1: Descriptive Characteristics of Study Participants ..............................................45
Table 4.2: Background Characteristics of Babies Delivered by the Hepatitis B Positive Mothers .................................................................................................................................48
Table 4.3: Association between demographic characteristics of Hepatitis B positive mothers and the infection status of their babies .....................................................................50
Table 4.4: Association Between Obstetric and Family Characteristics of Hepatitis B Positive Mothers and The Infection Status of Their Babies ................................................................52
Table 4.5: Differences in Means of Continuous Variables by The Infection Status of the Baby .................................................................................................................................54
Table 4.6: Equality of Means of Viral Load Of Mothers by the HBsAg Status of Their Baby55
Table 4.7: Equality of Means of Viral Load of Mothers by Their Demographic Characteristics .................................................................................................................................56
Table 4.8: Association Between Infection Status of Baby and the Hepatitis B e-Antigen Status of Mother and Mode of Delivery of Baby ......................................................................57
Table 4.9: Binary Logistic Regression Analysis of Mode of Delivery and Hepatitis B e-Antigen on Positive Infection Status of Baby .........................................................................................58
LIST OF FIGURES

Figure 1.1: Relationship between the (Neonatal Hepatitis B Infection) and the independent
variables. .................................................................................................................................................. 5

Figure 3.1: Geographical location of Sekondi Takoradi Metropolis, Ghana................................. 29

Figure 4.1: Other medical conditions of Hepatitis B positive mothers............................................. 46
# LIST OF ABBREVIATIONS

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Full Form</th>
</tr>
</thead>
<tbody>
<tr>
<td>AHB</td>
<td>Acute Hepatitis B</td>
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<tr>
<td>ALT</td>
<td>Alanine Aminotransferase</td>
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<td>ANC</td>
<td>Antenatal Clinic</td>
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<td>ANOVA</td>
<td>Analysis of Variance</td>
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<td>Anti-HBc</td>
<td>Hepatitis B Core Antibody</td>
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<td>Anti-HBe</td>
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<td>Anti-HBs</td>
<td>Hepatitis B Surface Antibody</td>
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<td>aOR</td>
<td>Adjusted Odds Ratio</td>
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<td>APRI</td>
<td>AST to Platelet Ratio Index</td>
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<td>ART</td>
<td>Antiretroviral Therapy</td>
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<td>AST</td>
<td>Aspartate Aminotransferase</td>
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<td>CHB</td>
<td>Chronic Hepatitis B</td>
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<td>cOR</td>
<td>Crude Odds Ratio</td>
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<td>DHIMS</td>
<td>District Health Information Management System</td>
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<td>DNA</td>
<td>Deoxyribonucleic Acid</td>
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<td>Elective Cesarean Section</td>
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<td>Fluorescein Isothiocyanate</td>
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<td>Ghana Ports and Harbours Authority</td>
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<td>HBeAg</td>
<td>Hepatitis B e-Antigen</td>
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<td>HBIG</td>
<td>Hepatitis B Immunoglobulin</td>
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<tr>
<td>Abbreviation</td>
<td>Full Form</td>
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<tr>
<td>HBsAg</td>
<td>Hepatitis B Surface Antigen</td>
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<td>LMICs</td>
<td>Lower Middle-Income Countries</td>
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<td>MTCT</td>
<td>Mother to Child Transmission</td>
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<td>Normal Human Serum</td>
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<td>Polymerase Chain Reaction</td>
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<td>PWID</td>
<td>People Who Inject Drugs</td>
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<tr>
<td>RR</td>
<td>Relative Risk</td>
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<tr>
<td>SGA</td>
<td>Small for Gestational Age</td>
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<tr>
<td>UQ</td>
<td>Ultra Quality</td>
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<tr>
<td>USA</td>
<td>United States of America</td>
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<tr>
<td>VRA</td>
<td>Volta River Authority</td>
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<td>WHO</td>
<td>World Health Organisation</td>
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CHAPTER ONE

INTRODUCTION

1.1 Background

Hepatitis B viral (HBV) infection causes a serious infectious disease of the liver, and which results in acute and chronic diseases such as cirrhosis of the liver and hepatocellular carcinoma (WHO, 2002).

Globally, nearly 350, 000, 000 – 400,000, 000 individuals have the chronic hepatitis B (CHB) status, and the hepatitis B virus with approximately, 500,000 to 1, 200, 000 people deaths every year from chronic hepatitis, liver cirrhosis and hepatocellular carcinoma (Liu et al., 2015). In 2015, WHO estimated that 257 million people were chronically infected with HBV, and 887,000 died from complications of HBV related liver diseases (Nelson & CDC, 2017).

The high rate of vertical transmission is as a result of HBV infection during pregnancy, consequently leading to hepatitis B infection in fetuses and neonates (Belopolskaya et al., 2015); (World Health Organization, 2015a).

Transmission of HBV is mainly by contact after cutaneous or mucosal infection of blood and other bodily fluids, such as saliva, menstrual, vaginal, and seminal fluids (World Health Organization, 2015a); (Sarin et al., 2016). Sexual transmission occurs predominantly in both unvaccinated homosexual and heterosexual persons, as well as individuals who have several sex partners or through contact with commercial sex workers (Chernet, Yesuf, & Alagaw, 2017). Chronic hepatitis status is attained in only 5% of cases after infection with HBV as an adult (World Health Organization, 2015a). Intrauterine transmission of HBV has recently been
implicated as the major route of maternally-acquired HBV infection behind the failure of vaccine to prevent neonatal hepatitis B infection (Navabakhsh et al., 2011); (Awuku & Yeboah-Afihene, 2018). A greater proportion of babies of Hepatitis B-infected mothers become end up with the hepatitis B virus despite both primary vaccination HBV vaccination and Hepatitis B immunoglobulin (HBIG) prophylaxis and rates of transmission correlate directly with levels of maternal HBV viraemia (World Health Organization, 2015). There is a 10% or more risk of MTCT with excessively high maternal concentrations of HBV-DNA, observed in HBeAg-positive women, despite HBIG and vaccine prophylaxis (World Health Organization, 2015a) (World Health Organization, 2015a). (Gentile & G, 2014); (Chen et al., 2013). Commencement of prompt and appropriate immunoprophylaxis therapy immediately after birth and continued as scheduled in breast fed infants adds little extra risk for the transmission of HBV even in the face of the presence of HBV DNA in the breast milk of HBV infected mothers (Navabakhsh et al., 2011). Hence, breastfeeding need not be delayed until all doses of HBV vaccine have been administered to the infant (Navabakhsh et al., 2011) (SA Maternal & Neonatal Community of Practice, 2016).

In Africa, viral hepatitis, and HBV infection is an endemic health condition of public health concern, with levels comparable to the major communicable diseases, including Human Immuno-deficiency Virus (HIV), tuberculosis and malaria (WHO, 2002). HBV from infected mothers to their fetuses or newborns remains a major source of perpetuating the reservoir of chronically infected individuals (Rafat & Hakim, 2016). Sub-Saharan Africa has high prevalence up to 20%. Acute HBV infection results in chronic infection in approximately 90% of perinatally-infected individuals as pertains in Sub-Saharan Africa (Archampong, 2016); (Archampong, 2016b); (Adegbesan-omilabu et al. 2015).
In 2013 the prevalence of chronic hepatitis B virus infection in Ghana was 12.92% (Archampong, 2016); (Awuku & Yeboah-Afihene, 2018); (Adjei et al., 2016). However, this was derived from an analysis of only 12 studies (Spearman et al., 2017); (Keane, Funk, & Shimakawa, 2016) but the prevalence rate of HBV in Ghana from other studies was 10–15% with prevalence of HBV infection ranging from 12-15% from blood donors (Andersson et al., 2013). There are currently no other thoroughly conducted reviews specifically summarizing data on prevalence of HBV in Ghana (Awuku & Yeboah-Afihene, 2018); (World Health Organisation, 2016). This observation points out that although there may have been significant research into understanding the burden of HBV in Ghana, the evidence available remains fragmented (Ofori-Asenso & Agyeman, 2016). In Ghana, HBV is of significant public health importance and a disease that requires greater attention.

1.2 Problem Statement

Globally, the World Health Organisation (WHO) estimates that two billion people are infected with the HBV, with over 240 million chronic carriers (4%-6% of world population) (Chernet et al., 2017); (Isah et al., 2016). Over 600,000 people die every year due to the consequences of HBV infection (Rafat & Hakim, 2016).

In 2014, the WHO set five key service coverage targets of 90% coverage of hepatitis B childhood vaccination, 90% coverage of birth-dose vaccination or other means to prevent mother-to-child transmission, 100% of blood donations screened in a quality-assured manner, 90% of injections given with safety-engineered devices, and distribution of at least three hundred sterile needles and syringes/people who inject drugs (PWID)/year (Sonderup et al., 2017) (World Health Organization, 2016). Reaching these five service coverage targets by 2030
would reduce the incidence of chronic infections by 90% and mortality by 65% (as compared to 2015 levels), which would eliminate hepatitis B as a public health threat (World Health Organization, 2016). Hence, improving these services to eliminate hepatitis will strengthen health systems (Navabakhsh et al., 2011).

In 2014, coverage in the African Region remained low (10%). Ultimately, newer methods to prevent mother-to-child transmission of HBV, such as the administration of antivirals to Hepatitis B-positive pregnant women to reduce the Hepatitis B viral DNA load (HBV-DNA), will be needed to achieve the proposed targets (WHO Afro, 2017).

In the Sekondi Takoradi Metropolis hepatitis B, annual prevalence for 2017 was 2.3 per 100,000, with no available data on neonatal incidence (District Health Information Management System [DHIMS] 2 Database). Currently, the health system in Ghana does not capture data on Hepatitis B, hence, there was no source data to validate this figure.

This current study is to establish the maternal factors responsible for the transmission of hepatitis B in neonates born to infected mothers in selected health facilities in the Sekondi Takoradi Metropolis, to provide data needed to confirm these findings and to determine the significant factors for transmission in Hepatitis B-exposed neonates in the country.
1.3 Conceptual Framework

Figure 1.1: Relationship between the (Neonatal Hepatitis B Infection) and the independent variables.

1.3.1 Narrative of Conceptual Framework

High maternal hepatitis B viral deoxyribonucleic acid (DNA) levels is a major risk factor of hepatitis B viral (HBV) infection. Mothers with hepatitis B Viral (HBV) DNA level of in excess of 8 log10 copies/mL is related to immunoprophylaxis failure in pregnancy. Latest studies report that vaccine breakthrough has been reported in infants whose mothers are Hepatitis B e-Antigen positive have HBV DNA levels higher than 1,000,000 copies/mL. Pregnant mothers with an HBV DNA titer above 107-108 copies/mL may lead to immunoprophylaxis failure.

There are higher rates of HBV infection in children born to HBeAg-positive mothers with positive associations between both HBsAg and HBeAg. HBeAg is able to pass through the placenta from mother to fetus. There is resultant T-cell tolerance in the uterus with HBeAg positivity having a positive correlation with increasing MTCT and high maternal viral load.
Hence, HBeAg-positive mothers tend to have significantly higher HBV viral loads than in the HBeAg-negative mothers.

Vaginal delivery is another risk factor associated with MTCT of HBV with Cesarean section being superior to vaginal delivery in preventing MTCT. Cesarean section results in the lowest rate of mother-to-fetus micro-transfusion among four delivery modes, including Cesarean section, normal spontaneous vaginal delivery, vacuum or forceps delivery and emergency Cesarean after labor. Cesarean section reduces the rate of immunoprophylaxis failure to 2.1% at 7-12 months after birth, compared with 5.9% associated with vaginal delivery.

1.4 Justification
Despite the high prevalence of Hepatitis B viral (HBV) infection, there is little attention dedicated to it. Currently 25% of persons with Chronic Hepatitis B (CHB) are persons infected as infants or young children, and they die prematurely from cirrhosis of the liver and hepatocellular carcinoma. These individuals remain asymptomatic until end-stage liver disease is detected later in life. The most important and the main transmission route is the perinatal route through which infection occurs frequently if the mother with Hepatitis B virus (HBV) is HBeAg-positive and has a high level of serum HBV-DNA. If infants born to HBsAg-positive mother do not receive perinatal hepatitis B vaccine and immunoglobulin prophylaxis, infection rates in infants born to HBsAg positive and negative mother are 70 to 90% and 10 to 20%, respectively, of which 90% will progress to chronic infection. This phenomenon makes prevention of maternally-acquired HBV infection a crucial step in eradicating HBV infection.

High maternal viral load and the presence of maternal Hepatitis B e-Antigen confer major risk of mother-to-child-transmission to their babies and data showed that about 10%-23% of infants
from HBsAg positive mothers with combined immunoprophylaxis displayed HBV DNA positivity or HBsAg positivity at birth. Prolonged exposure of the fetus to vaginal secretions during delivery also leads to high transmission rates as compared to Cesarean mode of delivery.

There is scanty information available on the current rate of neonatal hepatitis B viral infection and factors associated with transmission from among babies of infected mothers in Ghana. Additionally, high cost of testing, and presentations of cases in the advance stage of the disease account for the high mortality rate as result of HBV.

This emphasizes the need to conduct a study on the maternal risk factors associated with Hepatitis B viral transmission among their babies. Previous studies have shown seven to ten percent (7-10 %) of infants born to infected mothers develop CHB even after administration of Hepatitis B vaccine and immunoglobulin after delivery.

This study seeks to provide the necessary baseline information on the current prevalence of neonatal HBV infection. The findings from this study will inform both health specialists and policy makers of the need to focus on prevention of in utero and intra partum transmission, together with active and passive immunization, which is the practical method for settings with no universal national HBV control policies like Ghana.

1.5 Research Questions

1. What is the association between viral load of mothers who are HBsAg positive and infection in their babies?

2. What is the association between the Hepatitis B e-Antigen status of hepatitis B positive mothers and transmission of the virus to their babies?
3. What is the association between the mode of delivery of babies born to mothers who are HBsAg positive and the infection status of their babies?

1.6 Objectives

1.6.1 General Objective:

To determine maternal factors associated with transmission of hepatitis B viral infection to their babies of infected mothers

1.6.2 Specific Objectives:

1. To determine association between viral load of mothers who are HBsAg positive and infection in babies

2. To determine association between the Hepatitis B e-Antigen status of mothers who are HBsAg positive and transmission of infection to their babies

3. To determine association between the mode of delivery of babies born to mothers who are HBsAg positive and the infection status of their babies
CHAPTER TWO

LITERATURE REVIEW

2.1 Etiology and Pathology
Chronic hepatitis B (CHB) is a serious infectious disease of the liver with the hepatitis B virus (HBV) being the aetiologic agent. The HBV is an enveloped virus containing a double stranded circular DNA genome and belongs to the hepadnavirus family (WHO, 2002). Hepatitis B Virus infection is deemed persistent following the presence of hepatitis B surface antigen in the blood or serum after six months, with or without active viral replication and indication of hepatocellular injury and inflammation (World Health Organization, 2015a). The age of acquiring the HBV infection is the main determinant of the risk of chronic HBV infection (World Health Organization, 2015a) (Dyson et al., 2014). HBV infection of newborns is associated with greater risks of chronic liver disease and serious subsequent complications (Chen et al., 2013).

An estimated proportion of 15-40% of patients will develop serious complications as a result of HBV infection with related liver failure and hepatocellular carcinoma without therapy (Dyson et al., 2014). It is projected that there are over 620,000 HBV-related deaths each year worldwide with 5–10% of liver transplants resulting from chronic HBV infections in both lower-middle income countries (LMICs) and developed countries (Dyson et al., 2014); (World Health Organization, 2015a); (Xiaosong et al., 2018). Acute disease and fulminant liver diseases have been implicated in all five hepatitis viruses, with the highest numbers of deaths resulting from liver cancer and cirrhosis, which occur after decades of chronic hepatitis B or C infection (WHO Afro, 2017); (Sonderup et al., 2017).
Earlier studies have demonstrated that the risk of developing cirrhosis occurs in 8–20% in untreated persons with CHB over five years (World Health Organization, 2015a). The authors further explain that hepatic decompensation occurs in approximately 20% of those with cirrhosis, and an extremely high and has been implicated as a leading risk factor of hepatitis B-related hepatocellular carcinoma, in 1% to 5% of cases. They also illustrated that without treatment, persons with advanced, symptomatic cirrhosis as a result of HBV infection, have a 15–40% survival five years after infection. This study reported that HBV infection individuals results in hepatitis B “e-antigen” (HBeAg)-positive or -negative variant of the disease.

The clinical features, natural history and consequently, the observed immune response of HBV infection, occur as a result of interactions between the virus and host (Cui et al., 2016). This inflammatory response by the host cells is, therefore, crucial to the further spread of virus as well as causing liver damage (Tan, Koh, & Bertolletti, 2015). The clinical course of CHB is usually influenced by impaired immunity from various causes (Dionne-Odom, Tita, & Silverman, 2016); (Sarin et al., 2016); (Han et al., 2014). During pregnancy, the maternal immune system is altered to tolerate the genetically different fetus, and hormonal factors may also play a significant role in altering immune regulation or viral replication (Han et al., 2014); (Hellard, Chou, & Easterbrook, 2017).

The T cell response to HBsAg was weaker in pregnant women than in adults males and non-pregnant females with resultant immunological response to acute HBV infection, and consequently the clinical course of Acute Hepatitis B (AHB) in pregnant women being different from those in the general population (Han et al., 2014); (Liu et al., 2014); (Liu, 2015).

The extent of adverse pregnancy outcomes is directly associated with hepatitis B virus DNA load as evident by the fact that individuals with HBeAg positive chronic infection usually have
high amounts of hepatitis B viral DNA (Cui et al., 2016). There is high hepatitis B virus DNA load even with concurrent administration of standard passive-active immunoprophylaxis together with the hepatitis B virus vaccine. Furthermore, due to the high failure rate of the hepatitis B virus immune globulin (HBIG) in neonates the rate of intrauterine infections is approximately 15% (Cui et al., 2016). Evidence from other clinical trials have indicated that antiviral treatment in women with a high hepatitis B virus load in the third trimester will effectively reduce incidence of mother-to-child transmission of HBV infection (Cui et al., 2016) (Liu et al., 2014); (Sarin et al., 2016). Routine hepatitis B vaccination is given is to infants soon after delivery in many countries worldwide (World Health Organization, 2015a). The effectiveness of this intervention on end-stage liver disease or hepatocellular carcinoma is still significantly low 20–40 years after the introduction of universal infant immunization (Akinyemiju, Mcdonald, & Lantz, 2015).

2.2 Virological Characterization

The hepatitis B virus (HBV) of the hepadnavirus family, is one of the viruses with the smallest sizes known to infect humans (World Health Organization, 2015a). The HBV has a high affinity for hepatic cells, with inflammation of hepatocytes occurring through destruction of infected liver cells (World Health Organization, 2015a). HBV is a leading carcinogenic virus with its infection resulting in extremely higher risk of hepatocellular carcinoma (World Health Organization, 2015a). The viral genome encodes the HBsAg, hepatitis B core antigen (HBcAg), the viral polymerase and the HBx protein which is a 154 amino acid that interferes with transcription, signal transduction and cell cycle progress, protein degradation, apoptosis and chromosomal stability in the host (World Health Organization, 2015a).
The virus is a 42 nm double-shelled particle found in infected blood, and is composed of an outer covering of HBsAg and a nucleocapsid component of hepatitis B core antigen (HBcAg) inside its structure. Detection of HBV DNA is detected in serum and is a marker of viral replication and the risk of infectivity of an infected individual. The hepatitis B envelope antigen (HBeAg), presents as a soluble protein in serum, unlike the HBsAg and HBcAg, which are not particulate (World Health Organization, 2015a).

A minimum of nine genotypes of HBV (Hepatitis A to I) are recognized worldwide differentiated by their genome sequences (World Health Organization, 2015b). Higher rates of hepatocellular carcinoma have been found in persons infected with genotypes E and F (in comparison with genotypes B or D), as well as in those individuals infected with particular subtypes of genotype A specific to southern Africa, although aflatoxin exposure plays a key role in the sub-Saharan African region (World Health Organization, 2015b).

Both antiviral therapy and the HBV vaccine have been proven to be equally effective in protecting against all HBV genotypes (World Health Organization, 2015a). Several naturally occurring mutations in the pre-core region of the HBV (pre-core mutants) have been identified in HBeAg-negative persons with CHB and these are known to prevent HBeAg synthesis (World Health Organization, 2015a). Numerous genotypes in the HBV influences the prevalence of pre-core mutations, but the functional role of this mutation in liver disease is unclear (World Health Organization, 2015a).

2.3 Risk Factors
Age is a major risk factor for mother-to-child-transmission (MTCT) of hepatitis B viral (HBV) infection. The risk of advancement to chronic HBV infection is inversely proportional to the age at which the infection was acquired (Nelson & CDC, 2017).
A greater proportion of neonates, up to 34%, are known to become infected with HBV after birth due to close contact with their infected mothers (Gentile & G, 2014). Some host and viral factors, significant among them being coinfections with HIV, hepatitis C virus (HCV) and hepatitis D virus (HDV), together with other cofactors such as alcohol use, also increase the rate of disease progression and risk of developing HCC (World Health Organization, 2015a).

Age, sex, and host immunity status are important factors influencing the clinical manifestation and outcome of acute HBV infection (Han et al., 2014) (Cui et al., 2016) (Xiaosong, 2018). Hepatitis B virus load, together with the HBeAg, represents a key factor in determining disease prognosis, from immune tolerance, to chronic inflammation, to life-threatening complications, through induction of inappropriate immunological reactions (Xiaosong, 2018); (Huang & Zhong, 2017).

2.4 Transmission
There is potential viral transmission from mother to newborn despite at-birth prophylaxis with immunoglobulin and vaccine (Rafat & Hakim, 2016).

The major route of spread of HBV is through the skin or mucosal exposure to infected blood and various body fluids, including saliva, menstrual, vaginal, and seminal fluids, and these are the known vehicles of human transmission (World Health Organization, 2015a).

Horizontal transmission of hepatitis B occurs as a sexually transmitted infection in unvaccinated men who have sex with men and heterosexual individuals with multiple sex partners or who have contact with sex workers. Only less than 5% of persons infected as adults leads to chronic hepatitis B infection. Additionally, accidental inoculation of the virus results in transmission of undetectable amounts through infected blood or body fluids as a result of medical, surgical and
dental procedures, or from razor blades and other objects contaminated with infected blood; use of inadequately sterilized syringes and needles; intravenous and percutaneous drug abuse; tattooing; body piercing; and acupuncture (World Health Organization, 2015a); (Navabakhsh et al., 2011).

2.4.1 Prenatal Transmission

Mother-to-child-transmission (MTCT) is the predominant route of hepatitis B virus (HBV) infection in many endemic areas worldwide, and serves as a major source in maintaining the reservoir of the infection in some regions, notably in China and South- East Asia (World Health Organization, 2015a). A large proportion of viraemic mothers, especially those who are seropositive for HBeAg, transmit the infection to their infants just before, or shortly after birth without prophylactic administration of HBV vaccine to neonates (Navabakhsh et al., 2011). According to the World health Organisation (WHO), the risk of perinatal infection is also increased if the mother has acute hepatitis B in the second or third trimester of pregnancy or within two months of delivery. The WHO, however, argues that although in utero transmission occurs in some fetuses, this is an uncommon finding and is generally associated with antepartum haemorrhage and placental tears. There is a 90% risk of developing chronic infection resulting from perinatal infection (up to 6 months of age) but this drastically decreases to 20–60% when infection occurs from the ages of 6 months and 5 years (World Health Organization, 2015a); (Spearman et al., 2017).

Horizontal transmission, including household, intrafamilial and especially child- to- child, is also important (Adjei et al., 2016). Up to 50% of infections in children are as a result of factors other factors apart from mother-to-child-transmission and, in many endemic regions, prior to
the introduction of neonatal vaccination, the prevalence peaked in children 7–14 years of age (World Health Organization, 2015a).

The pre-natal (intrauterine) route of HBV transmission is currently considered the chief culprit behind this failure (Navabakhsh et al., 2011). The various possibilities hypothesized include:

1. A breach in the placental barrier:

Transplacental leakage of HBeAg-positive maternal blood, which is induced by uterine contractions during pregnancy and the disruption of placental barriers (such as threatened preterm labor or spontaneous abortion), is one of the most likely routes to cause HBV intrauterine infection (Gentile & G, 2014).

Amniocentesis inoculates the intrauterine cavity with maternal blood because the needle traverses the abdominal and uterine wall. However, HBV transmission during amniocentesis appears to be rare, particularly in mothers who are HBeAg-negative and when the procedure is done using a 22-gauge needle under continuous guidance (Navabakhsh et al., 2011).

2. Placental infection and trans-placental transmission of HBV:

Placental infection in a fetus with intrauterine HBV infection can either be the route for transmission of HBV from the mother to the fetus or secondary to fetal infection by another route (Gentile & G, 2014). To distinguish between these two possibilities, researchers have measured the gradient of placental infection between the maternal side and the fetal side of the placenta and concluded that in the majority of cases, transplacental infection is the mechanism for HBV intrauterine infection (Isah et al., 2016).
3. Studies have also demonstrated that HBV-DNA exists in oocytes of infected females and sperms of HBV-infected males. Therefore, it is possible for the fetus to become infected with HBV at conception (Wang et al., 2003).

4. Another possibility is the intrauterine transmission of HBV to the fetus, not from maternal blood but ascending from vaginal secretions of the mother that contain the virus (Cui et al., 2016).

According to Abdi, (2015) transmission via the placenta is not as common as previously thought; actually, viral DNA is rarely found in amniotic fluid or cord blood. They observed that most infant infection in the womb is caused by the mother’s blood transfusion to the fetus during uterine contractions or rupture of fetal membranes or by vertical transmission perinatally by exposure to blood or secretions of an infected birth canal of the mother. The study concludes that an estimated 50% of cases of chronic hepatitis B are results of vertical transmission or acquired in early childhood. They countered that the mechanisms through which HBV infections are transmitted in the uterus are controversial and being reviewed (Abdi, 2015). Some of the assumptions this study illustrated include transmission through the placenta, transfer through placental leakage, cracks in the placental barrier, mononuclear cells in peripheral blood and transmission through the father. Intrauterine transmission of HBV infection occurs via two pathways:

(1) Blood release (hematogenous) that causes the infection of placental vascular endothelial cells and is probably the main route for infection transmission

(2) Cellular transport through cell by cell.
One of the explained mechanisms of the intracellular transport route is binding of HBsAg-anti-HBsAb with Fc-\(\gamma\) receptor III (Abdi, 2015).

### 2.4.2 Natal Transmission

According to Navabakhsh et al., (2011) transmission of HBV to the infant at the time of birth is believed to be a result of exposure to maternal cervical secretions and maternal blood that contain the virus.

The authors believe that there are still some controversies regarding the effect of delivery mode on MTCT; in current obstetrical guidelines, the mother’s HBsAg positivity does not affect the planned mode of delivery irrespective of her HBeAg status or level of viremia. They additionally cited some articles that recommend cesarean section in case of high maternal HBV-DNA levels, whereas others believe that mode of delivery does not influence the rate of HBV transmission provided that all infants receive HBIG and HBV vaccine at the recommended schedule. According to them, a recent systematic review in 2008 on four randomized controlled trials (RCTS) involving 789 people concluded that cesarean section before labor or before ruptured membranes (elective cesarean section or ECS) appears to be effective in preventing MTCT of HBV. However, the authors point out that the conclusions of this review must be considered with great caution due to high risk of bias in each included study (graded C). RCTS of higher quality are required for assessing the effects of ECS in comparison to vaginal delivery for preventing MTCT of HBV (Navabakhsh et al., 2011).

### 2.4.3 Postnatal Transmission

Although HBV-DNA is present in the breast milk of HBV infected mothers, feeding their infants with this milk poses no additional risk for the transmission of HBV provided that
appropriate immunoprophylaxis is commenced at birth and continued as scheduled. There is no need to delay breastfeeding until the child has received all doses of HBV vaccine (Navabakhsh et al., 2011).

Breastfeeding does not have a negative influence on the immune response to the HBV vaccine and does not increase its failure rate (Navabakhsh et al., 2011). As a general rule, the study recommended to explain to mothers that they should take good care of their nipples while breast-feeding, ensuring proper latch-on and allowing the nipples to dry before covering to avoid cracking or bleeding, having in mind that HBV is commonly passed by blood-to-blood routes. HBV from infected mothers to their fetuses or newborns remains a major source of perpetuating the reservoir of chronically infected individuals (Rafat & Hakim, 2016).

In some instances, infants born to HBsAg-positive mothers become infected with hepatitis B in the face of both HBV vaccination and/or hepatitis B virus immune globulin (HBIG) prophylaxis; (Liu et al., 2014). It is estimated that the risk of transmission, despite HBV vaccination and HBIG, are diverse, but correlate to levels of circulating maternal HBV virus. Extremely high maternal concentrations of HBV DNA, usually observed in HBeAg-positive women, create an additional 10% or more risk of transmission, even after HBIG and vaccine prophylaxis (Dionne-Odom et al., 2016).

World Health Organization, (2015a) recommends the use of Antiretroviral Therapy (ART) initiated during pregnancy in HIV-infected pregnant women which has been proven to considerably reduce the risk of mother-to-child transmission of HIV during pregnancy, labour and delivery, and breastfeeding to less than 1–2%. The WHO-recommended tenofovir containing regimens are equally effective against HBV infection and hence play a key role in reduction of mother-child-transmission of HBV. They also indicated that data from previous
studies recommend maternal treatment with Nucleoside Analogue therapy in the third trimester in addition to the HBV vaccine and HBIG administered to the infant may also reduce HBV transmission in neonates. According to them, defective adherence to the neonatal vaccination schedule may thus be resolved, and rather to focus on the administration of the initial birth dose of vaccine (with or without HBIG) in neonates born to mothers with very high HBV viral DNA copies. Conversely, despite the general practice globally that allow treating highly viraemic pregnant mothers with lamivudine, telbivudine or tenofovir, as an adopted a policy, particularly in Asia, the efficacy of adjuvant maternal treatment with antivirals in the third trimester of pregnancy remains uncertain (World Health Organization, 2015a). Such treatment would be for a limited period for reducing the risk of infection to the baby. If a woman requires treatment based on her own clinical condition then that treatment would be continued through the pregnancy. The authors concluded that lamivudine is the most widely studied agent of those that are active against HIV and HBV; and that there is also a sizeable body of data in women who have received tenofovir as part of an ART regimen.

2.5 Diagnosis and Staging
According to World Health Organization (2015a), regular assessment and follow up of HBsAg-positive persons is required to guide its management and serve as a basis for treatment. They recommended the assessment to include: additional serological markers of HBV infection such as hepatitis B e-Antigen (HBeAg); determining serum aminotransferase levels to help detect inflammation within the liver; quantification of HBV DNA levels; and staging of liver fibrosis by non-invasive tests (NITs) like the aspartate aminotransferase (AST)-to-platelet ratio index (APRI), transient elastography (FibroScan) or FibroTest. They further observed the occurrence of circulating antibodies in an infected case [antibodies to the hepatitis B surface antigen(anti-
HBs) and antibodies to the hepatitis B core antigen (anti-HBc)] indicated past HBV infection of the. Immunity to HBV infection after vaccination is, however, depicted by the occurrence of only the anti-HBs (Sonderup et al., 2017). The authors defined CHB as the presence of serum HBsAg after 6 months. Some studies suggest quantification of serum HBsAg levels to distinguish inactive HBsAg carriers from persons with active disease.

The World Health Organization, (2015a) also concluded that serum HBeAg levels need to be measured establish whether the person is in the HBeAg positive or HBeAg-negative phase of infection, and since this status can change with time, there needs to be lifelong monitoring in both instances. In infected individuals with CHB, a positive HBeAg result usually specifies an active HBV replication and a high infectivity status of that person. Studies have proven that after HBeAg-positive seroconversion (anti-HBe) there is usually spontaneous improvement in the prognosis, with a reduction in HBV replication, and normalization of ALT levels (Isah, 2016). This, seroconversion, therefore, eliminates any need for treatment in such a case. Additionally, HBeAg is used to monitor treatment response because HBeAg (anti-HBe) seroconversion in HBeAg-positive persons with a persistent undetectable HBV DNA viral load is now a potential determiner for completion of antiviral therapy (Sarin et al., 2016). This is, however, a rare occurrence even after administering potent nucleoside/nucleotide analogue (NA) therapy. It is possible for a proportion of HBeAg negative persons to have active HBV replication but even when positive for anti-HBe but fail to produce HBeAg as a result of the presence of HBV variants or pre-core mutants (World Health Organization, 2015a).

2.6 Prevention
HBeAg status is an important simple marker and risk predictor of pregnancy-related outcomes in chronic hepatitis B virus-infected individuals (Chen et al., 2013). Prompt detection of
hepatitis B virus infection prior to pregnancy, monitoring the risk of preterm birth, and providing appropriate medical intervention. There will be resultant improvement of maternal hepatitis viral DNA levels, e-Antigen status, thus reducing the risk of mother-to-child transmission (MTCT), and improving neonatal outcomes (Cui et al., 2016).

Preventive strategies for HBV infection include catch-up vaccination for target groups such as household and sexual contacts of persons who are HBsAg-positive; and persons at risk of acquiring HBV infection, including people who inject drugs (PWID), men who have sex with men (MSM), and persons with multiple sex partners as well as young adolescents; (World Health Organization, 2015a) (Kwon & Lee, 2011).

2.6.1 Screening

It is recommend by internationally-approved protocols for several high-risk groups to be screened for HBsAg, and to provide hepatitis B vaccination for persons at risk and not immune to the HBV (World Health Organization, 2015a). Chief among these are the household and sexual contacts of persons with CHB, HIV-infected persons and other individuals with reduced immunity such as diabetics, persons who inject drugs (PWID), men who have sex with men, sex workers, as well as other groups such as indigenous peoples, persons who are incarcerated, and the transgender (Dionne-Odom et al., 2016). It is also suggested for routing screening of blood and organ donors for HBsAg and other blood-borne pathogens in accordance with WHO recommendations to prevent HBV transmission, especially in LMICs (Sarin et al., 2016). In the United States and Europe, population based screening is also recommended for migrants from endemic (World Health Organization, 2015a). However, screening protocols for HBsAg in Lower-Middle Income Countries (LMICs) are, by far, scarce and limited (Name & Link, 2015).
2.6.2 Prevention through Vaccination

There are widely available and effective vaccines for HBV for over than two decades, and recently, major improvements in antiviral therapies for HBV have been made. (Stanaway et al., 2016). Other hepatitis B (HBV) infection prevention and control strategies include infant and neonatal hepatitis B vaccination and prevention of mother-to-child HBV transmission using antiviral therapy in the third trimester. Reports from the study indicate that utilization of recombinant DNA-derived vaccines against HBV are highly recommended and have been available for more than two decades. The primary hepatitis B immunization series originally comprises three doses of the HBV vaccine. Vaccination of infants and, in particular, injection of hepatitis B vaccine within 24 hours of birth confers a 90–95% protection, and thus enables prevention of infection with HBV, as well as decreasing HBV transmission if followed by at least two other doses (World Health Organization, 2015a). It has been recommended by that all infants receive universal hepatitis B vaccination, with the first dose to be given as soon as possible after birth. The WHO also discussed that that there has been resultant dramatic decline in the prevalence of CHB among babies and children worldwide, particularly in those regions where universal infant vaccination programmes have been implemented due to this strategy. The study explained that an inexplicable proportion of vaccinated children (5–10%) have a poor response to vaccination, and will remain as adults vulnerable with sustained susceptibility to acquisition of HBV infection(World Health Organization, 2015a).

In countries with intermediate or low endemicity, a substantial disease burden may result from acute and chronic infection acquired by older children, adolescents and adults (World Health Organization, 2015a). Target groups for catch-up vaccination as well as other preventive strategies include young adolescents; household and sexual contacts of persons who are
HBsAg-positive; and persons at risk of acquiring HBV infection, such as PWID, men who have sex with men, and persons with multiple sex partners (Kwon & Lee, 2011).

### 2.7 Natural History of Chronic Hepatitis B

Chronic hepatitis B (CHB) viral infection has a dynamic and complex natural history, with non-linear progression through several recognizable phases (World Health Organization, 2015a). The authors described different phases as “immune tolerant”, “immune-active”, “immune-control” and “immune-escape” which do not always relate directly to criteria and indications for antiviral therapy.

#### 2.7.1 Phases of Chronic Hepatitis B

1. The World Health Organization, (2015a) states that the immune-tolerant phase usually occurs in HBsAg-positive children and young adults who acquire the infection during the perinatal or early childhood period. This phase is known to be sustained until young adulthood and may persist 10–30 years after perinatal infection. According to them, serum HBeAg is classically detectable, HBV DNA levels are high (usually more than 200 000 IU/mL), and alanine aminotransferase (ALT) levels are usually normal or only marginally raised. They also determined inflammation of hepatocytes to be negligible at this stage, with minimal or slow progression to fibrosis, and reduced spontaneous HBeAg loss.

2. According to the study, the HBeAg-positive immune-active phase of active inflammatory disease is typically preceded by the immune-tolerant phase. The authors also observed that abnormal or fluctuating serum ALT levels are accompanied by alternating reduction in HBV DNA levels. They also stated that this phase may present with symptoms of impairment hepatic
function together with more severe, histologically evident hepatitis and fibrosis. According to
them the phase persists from several weeks to years, with resultant successful seroconversion
from an HBeAg-positive to an anti-HBe state. They also determined rates of HBeAg
seroconversion are higher in persons whose serum aminotransferases are markedly raised, as
well as those infected with genotypes D, A, F and (in Asia) B.

3. The non-replicative or inactive immune-control phase (previously called the inactive carrier
phase), typically occurs after successful seroconversion from an HBeAg positive to anti-HBe
state, and occurs in an estimated 10–15% of HBeAg-positive persons per year (World Health
Organization, 2015a). The study predicted that remission of hepatitis B infection may occur
after clearance of HBeAg, with delayed progression of hepatic fibrosis, while serum ALT levels
return to normal together with low or undetectable levels of HBV DNA (less than 2000 IU/
mL). A substantially reduced risk of hepatic cirrhosis and liver cancer occur in cases of HBeAg
seroconversion at a young age, before the onset of significant liver disease, and this confers a
good prognosis for such individuals. The authors, however, argued that active viral replication
is known to recur in a vast majority of infected persons.

4. The World Health Organization (2015a) explained that HBeAg-negative (“immune escape-
mutant”) active chronic hepatitis is detected in an estimated 5–15% of HBeAg-negative, anti-
HBe-positive cases in the inactive carrier state together with HBeAg-positive chronic hepatitis.
According to the authors, mutations in the pre-core or basal core promoter region of the hepatitis
B viral genome produce HBV variants with no expression of HBeAg, thus making HBeAg
levels undetectable (and anti-HBe detectable) in such people. This phase typically occurs later
in the progression of the disease, and is observed in older populations, with a fluctuating course
and abnormal or alterations in the levels of serum ALT and HBV DNA, necro-inflammatory changes, and swift progression to cirrhosis (annual rate of 8–20%).

5. Spontaneous HBV reactivation may occur during the next phase, or may be activated by cancer chemotherapy or other immunosuppressive therapy, with resultant fatal acute-on-chronic hepatitis, with preventive nucleos(t)ide analogue (NA) therapy, therefore, chosen as the preferred mode of treatment (World Health Organization, 2015a). The study concluded that persistent HBV DNA in the liver in cases of occult or undetectable hepatitis B surface antigen (HBsAg) in the blood, also becomes reactivated by aggressive chemo- or immunosuppressive therapy. Occult infection, according to the authors, depicts a constant reservoir of new infections in blood transfusion services usually in areas of HBV-endemicity within Lower-Middle income countries (LMICs) with the HBsAg status being used as singular marker of infection among donor populations. The study, thus, illustrated that reactivation is known to occur in individuals who have undergone HBsAg clearance, and who are negative for HBV DNA but anti-HBc positive after receiving potent immunosuppressive drugs.
CHAPTER THREE

METHODS

3.1 Study Design

A descriptive cross-sectional approach was employed to determine maternal risk factors associated with transmission of Hepatitis B virus to their babies. The study was carried out at antenatal units and labour wards of selected public and private hospitals in the Sekondi Takoradi Metropolis and the newly created Effia Kwasimintsim Municipality. All hepatitis B positive pregnant women of 34-37 weeks gestation, who had been screened and tested positive for Hepatitis B surface antigen (HBsAg) and attended hospitals for ANC services at these hospitals were eligible to partake in the study. Maternal antenatal books were reviewed, maternal and neonatal data were collected with the use of interviewer-administered questionnaires. Cord blood samples of babies born to hepatitis B positive mothers were collected and screened for the hepatitis B surface antigen (HBsAg). Venous blood samples of mothers were also collected and tested for hepatitis B viral DNA (HBV-DNA) levels and hepatitis B e-Antigen status.

3.2 Study Area

The Sekondi-Takoradi Metropolis is located at the south-eastern part of the Western Region of Ghana between Latitude: 4° 54' 59.99" North and Longitude: 1° 45' 59.99" West (Fig.1). The Metropolis is bordered to the west by Ahanta West District and to the south by Shama District. To the south of the Metropolis is the Atlantic Ocean and to the Northern part is the Wassa East District.

It has an estimated total population of 555, 548 people. The Metropolis covers a land size of 191.7 km square and is the most urbanized among the 22 districts in the Western Region (Ghana
Statistical Services, 2012). The Metropolis is made up of mainly Ahantas, but all other ethnic
groups are present (Ghana Statistical Services, 2012).

Healthcare zones are divided into four sub metropolitan areas being Effia Kwesimintsim,
Esikado, Sekondi and Takoradi Sub Metropolis (“2013 annual reproductive and child health
report annual report,” 2013). There are a total of 76 health facilities in the metropolis ten
hospitals (four government, one quasi-government and five private), three Health Centres, fifty
four clinics (government and private) and nine Community Health and Planning Services
(CHPS) compounds (Ghana Statistical Services, 2014).

The total number of Antenatal clinics in Sekondi Takoradi is 41 while 33 health facilities
undertake delivery services with the following distribution in the various sub-metropolitan
areas: Esikado 8, Kwesimintsim 15, Sekondi 3 and Takoradi 7 (Ghana Statistical Services,
2014); (World Health Organisation, 2016).

This study was conducted at the Antenatal clinics (ANC) of purposively-selected health
facilities, being public, quasi-government and private, and based on having the highest ANC
attendance and deliveries for 2017 and 2018. These are 1. Effia Nkwanta Regional Hospital, 2.
Takoradi Hospital, 3. Kwesimintsim Hospital, 4. Esikado Hospital, 5. Ghana Ports and
Harbours Authority (GPHA) Hospital, 6. UQ Specialist Hospital, 7. Jemima Crentsil Hospital
and 8. Our Lady’s Clinic. The selected ANCs are the leading health facilities for Antenatal care,
delivery and postnatal care in Sekondi Takoradi. Services provided include: sexual and
reproductive health services, management of individuals with chronic Hepatitis B viral
infection including screening and diagnostic test and hepato-protective medication, prevention and treatment of STIs including family planning, comprehensive palliative and a Home Based care, support for Orphans and Vulnerable Children, Hepatitis B vaccination, and additional areas including health research, training and capacity building of healthcare workers. The health facilities have an average patient turnover of 60-200 patients per day, headed by trained medical doctors who are the Medical Directors and Superintendents, and supported by other well-trained qualified medical doctors and physician assistants. The total staff population comprises medical doctors, Physician assistants trained nurses, biomedical scientists, laboratory technicians, sonographers, radiologists, public health officers, health record staff, and the rest are support staff and volunteers.

The Antenatal clinics are contained within various health institutions and they share some of the services such as laboratory, wards (medical, surgical and labor), theatre, public health unit, HBsAg screening unit. They provide health care for approximately 40 to 100 pregnant women per clinic day including those who are HBsAg positive. Each facility runs an average of three Antenatal clinic days per week. Hepatitis B screening tests are carried out usually during the first antenatal visit. The selected health institutions provide supervised and skilled delivery services including Cesarean sections. An average of 400 deliveries is conducted on a monthly basis in the selected health facilities, but the proportion of neonates who are HBsAg positive is not readily available.
Figure 3. 1: Geographical location of Sekondi Takoradi Metropolis, Ghana.

Source: Google Maps

3.3 Variables

3.3.1 Outcome variable

HBsAg status in neonates
3.3.2 Independent Variables (Table 1)

Table 3.1: Independent Variables

<table>
<thead>
<tr>
<th>Variables</th>
<th>Operational Definition</th>
<th>Measurement/ Possible Values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Information from ANC Booklet</td>
<td>Refers to the demographic characteristics, medical, family and obstetric history obtained from the ANC booklet</td>
<td>Categorical Values</td>
</tr>
<tr>
<td>Number of ANC visit</td>
<td>Refers to the number of times the pregnant woman has visited the ANC clinic</td>
<td>Numerical</td>
</tr>
<tr>
<td>Gestational age at first visit</td>
<td>Refers to the gestational age in weeks of first ANC visit.</td>
<td>Numerical</td>
</tr>
<tr>
<td>Gestational age at current visit</td>
<td>Refers to the gestational age in week of current ANC visit.</td>
<td>Numerical</td>
</tr>
<tr>
<td>HBsAg status</td>
<td>Refers to the HBsAg status of the pregnant woman as reported in the ANC booklet</td>
<td>Categorical Values</td>
</tr>
<tr>
<td>Socio Demographic Characteristics</td>
<td>Refers to the characteristics of the study participants</td>
<td>Categorical Values</td>
</tr>
</tbody>
</table>

Table 3.2: Operational Definition of Independent Variables

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<tr>
<td>Socio Demographic Characteristics</td>
<td>Refers to the characteristics of the study participants</td>
<td>Categorical Values</td>
</tr>
<tr>
<td>Age of participants</td>
<td>Refers to the age in years of the pregnant woman as reported during the interview.</td>
<td>Continuous</td>
</tr>
<tr>
<td>---------------------</td>
<td>---------------------------------------------------------------------------------</td>
<td>------------</td>
</tr>
</tbody>
</table>
| Occupation          | Refers to the work of the pregnant woman as reported during the interview. Employed refers to women who are employed by government or any agency. Self-employed refers to women working for themselves. Unemployed refers to women not working. And others refer to women who are students etc. | Categorical | Values:  
- Government worker  
- Self-employed  
- Unemployed  
- others |
| Educational status  | Refers to the educational status mentioned by the participant during the interview | Categorical | Values:  
- No Formal education  
- Primary/JHS  
- Secondary  
- Tertiary |
| Marital status      | Refers to the marital status reported by the participant during the interview | Categorical | Values:  
- Single  
- Married -cohabitation  
- Divorced/ separated/widowed |
| Gravidity           | Refers to number of times the woman has been pregnant as mentioned by the participant | Numerical   |
| Parity              | Refers to the number of surviving children of participants. | Numerical   |
| Risk Factors        | Refers to factors responsible for MTCT of HBV | Numerical   | -Maternal Viral load  
Categorical values:  
Maternal HBV e-Antigen status  
-Positive  
-Negative |
| Viral Load          | Refers to the maternal viral load levels of affected babies | Numerical   |
| Hepatitis B e-Antigen| Refers to the maternal Hepatitis B e-Antigen status of affected babies | Categorical:  
-Positive  
-Negative |
| Delivery            | Refers to the type method used by patient to give birth to baby | Categorical:  
-Vaginal delivery  
-Caesarean section |
Timely first dose vaccine | Refers to how soon after delivery neonate receives Hepatitis B vaccine | Numerical
---|---|---
HBIG | Refers to whether baby received the Hepatitis B immunoglobulin after delivery | Categorical
Values: Yes/No
Previous Caesarean Section | Refers to whether participant has had Caesarean section prior to this delivery | Categorical
Values: Yes/No

3.4 Sampling

3.4.1 Study Population

All neonates who were born to HBsAg positive mothers, and whose mothers attended Antenatal Clinic at the selected health facilities were eligible to be part of this study. Pregnant women on their routine follow-up visit for Antenatal care who were HBsAg positive, of 34-37 weeks gestation and consented to participate in the study had their new born babies screened for Hepatitis B infection using standardized screening kits.

3.4.2 Sample Size Determination

The Hepatitis B prevalence in neonates born to infected mothers ranged from 7% to 20% (Amidu et al., 2012). This study used a neonatal Hepatitis B viral infection prevalence of 7. The Cochrane formula was used for this study as follows:

Sample size \( n \) = \( \frac{Z^2 \cdot p(1-p)}{d^2} \)

\( n = \frac{Z^2 \cdot p(1-p)}{d^2} \)

\( n \) = minimum sample size

\( Z \) = the critical value associated with the level of significance (1.96 for 95% level of confidence)
p=the neonatal hepatitis b at 7.0 % (0.07)

d=precision/reliability to determine p=6% (0.06)

\[(3.84(0.07))/0.0009\]

Thus n= 70 (minimum sample)

Twenty percentage of this number (14) was added to take care of the attrition in the study population to add up the total number of study participants to be recruited 84.

3.4.3 Eligibility Criteria

Inclusion criteria

1. ANC attendants who were Hepatitis B positive
2. Hepatitis B positive ANC attendants of 34 – 37 weeks gestation,
3. Hepatitis B positive pregnant women who reported at the selected health facilities in labour
4. All eligible ANC attendants who consented to participate

Exclusion Criteria

1. HBsAg positive pregnant women with Human Immunodeficiency Virus (HIV) co-infection
2. Pregnant who were HBsAg positive with co-existing medical disorders, such as Diabetes mellitus, chronic hypertension, chronic renal disease, Haemoglobinopathies, acute hepatitis, acute or chronic symptomatic liver disease
3.4.4 Sampling Procedure

Eight hospitals from the Sekondi Takoradi Metropolis were purposively selected based on being the largest health facilities within the four sub-metropolitan areas and consequently having the highest number of antenatal attendants, having the highest delivery rates, providing both antenatal and delivery services and being equipped with theatres to perform emergency and elective Caesarean sections. The number of pregnant women selected in each facility was proportionate to the size of the estimated pregnant women population. The total number of antenatal attendants per study site in 2018 were as follows: Effia Nkwanta 11,965, Esikado 11,788, GPHA 4,515, Our Lady’s Clinic 1,132, Takoradi 9,352, UQ 1,959, Jemima Crentsil 5,643 and Kwesimintsim 14,757, while the deliveries were: Effia Nkwanta 2,665, Esikado 1,503, GPHA 431, Our Lady’s 187, Takoradi 989, UQ 344, Jemima Crentsil 1,174 and Kwesimintsim 14,757.

All HBsAg positive women of 34-37 weeks gestation attending Antenatal clinics at the selected hospitals were invited to partake in this cross-sectional study. Selection of study participants was done by reviewing antenatal booklets of pregnant women on their routine clinic visits, and selecting those who had tested positive as part of their routine antenatal laboratory tests, and who consented to be part of the study, until the sample size was attained. All pregnant women who met the inclusion criteria were selected until the required sample size was achieved. Pregnant women were recruited on every antenatal attendance day at each study site until the antenatal service ended on that day. All Hepatitis B-positive pregnant woman were eligible to take part in the study only once and women who re-visited the ANC during the study period were not included if they had already participated in the study. To control for double participation, the participant’s identification number was written at the back of the pregnant
woman’s maternal health record booklet. For pregnant women below 18 years who lived at most within a kilometer radius to the study site, their parents (or the marital partner) were traced and their consent sought before the participant was included in the study. All hepatitis B positive pregnant women in labour were also invited to participate in the study. During delivery of their babies, all neonates born to HBsAg positive mothers, were enrolled onto the study.

3.5 Data Collection Technique, Tools and Instruments

Data collection was carried out within a three-month period. The selected Antenatal clinics, labour wards and lying-in wards of the various health facilities were the sites for data collection. The study was explained to the mothers attending antenatal clinics at the study sites including the sample collection procedure, and informed consent form then signed after mothers consented to the study. Study participants were interviewed with the use of interviewer-administered questionnaire directly by the researcher or research assistants in a well-prepared interview room. This contained both closed and open-ended questions about the socio-demographic characteristics, sexual, obstetric, medical and family history. Maternal health record books were reviewed for the following: HBsAg Status, previous Cesarean section, gestational age and estimated date of delivery of current pregnancy during the particular antenatal clinic days. The telephone numbers of consenting mothers were collected, and their preferred place of delivery for current pregnancy noted for follow-up blood sample collection during delivery of their babies.

Neonates whose mothers were HBsAg positive had their cord blood samples taken by midwives at birth for screening for detection of their Hepatitis B Surface Antigen (HBsAg) status with the use of hypodermic needles and transferred into sample collection bottles. Venous blood
samples were collected from mothers of HBsAg-positive babies to test their hepatitis B viral DNA levels and Hepatitis B Envelope Antigen (HBeAg) status. The mothers were escorted to a well-prepared interview room, equipped with an examination bed, adequate light source, instrument tray, cotton swabs, appropriate size cannulas, disposable gloves, tourniquet and sample collection bottles. Mothers whose babies were HBsAg positive after being screened at birth were referred to physicians on duty for follow up of their babies.

3.5.1 Training of research Assistants

Prior to the study two research assistants from each of the selected health facilities, who were trained midwives or laboratory technicians, were recruited. Nurse managers and Senior Biomedical Scientists from each health facility were contacted and the purpose of the study and the roles of research assistants explained to them. Research assistants were subsequently assigned to the research team by their various departmental heads. The research assistants were trained on all aspects of the questionnaire and how to extract relevant information from the mothers as well as from their antenatal booklets. Additionally, they made to re-enact the collection of relevant information required from the maternal health record books of the pregnant women as their questions and concerns were addressed. The midwives were responsible for collection of cord blood of babies delivered at the labour wards for hepatitis B surface antigen (HBsAg) screening at birth, while the laboratory technicians collected venous blood samples from mothers for hepatitis B viral DNA (HBV-DNA) and sHBeAg testing.
3.5.2 Hepatitis B Surface Antigen Screening Procedure in Neonates

HBsAg screening was done at the labour or lying-in wards where neonatal cord blood was taken by either the principal investigator or midwives within 5 minutes of birth to prevent coagulation of the neonatal blood sample.

HBsAg screening was carried out using Biogate Laboratories Ltd® (Burnaby BC Canada V5A 0C4) Rapid Test membrane strips which are pre-coated with mouse monoclonal hepatitis B surface antibody (anti-HBs) to capture antibody on the test band region. This is a one Step HBsAg Whole Blood Test based on the principle of sandwich immunoassay for determination of HBsAg in serum or whole blood.

Monoclonal and polyclonal antibodies are used to identify HBsAg specifically. One or two drops of neonatal cord (whole) blood was pipetted onto test strip after which 1 drop of buffer solution was also dropped onto the buffer-allocated zone on the strip. The mouse monoclonal anti-HBs-colloid gold conjugate and sample consequently moved along the membrane chromatographically to the test region (T) and formed a visible line as the antibody-antigen-antibody gold particle complex formed and was visible to be read as positive or negative within 10 minutes. Both the Test Line and Control Line were not visible before applying any samples.

3.5.3 ELISA Procedure for Determining Hepatitis B e-Antigen

Laboratory technicians tested HBsAg positive mothers whose babies were positive after HBsAg screening post-delivery, for routine hepatitis B serological markers (HBsAg, HBeAg, and antibody hepatitis B e-Antigen [anti-HBe]) using Enzyme-Linked Immuno-Sorbent Assay (ELISA) Kits. Venipuncture was carried out tourniquet applied to puncture sites, and with a
good source of light, cotton swabs with methylated spirit applied to disinfect skin prior to sample collection procedure. Venous blood sample of mothers was collected for HBV DNA and HBV e-antigen testing.

Cell Biolabs’ QuickTiter™ HBeAg ELISA Kit, an enzyme immunoassay developed for detection and quantitation of the Hepatitis B e-Antigen where an anti-HBeAg monoclonal coating antibody had been adsorbed onto a microtiter plate. After specimen was pipetted on to the plate hepatitis B e-antigen present in the sample or standard was bound to the antibodies adsorbed on the plate, and an (fluorescein isothiocyanate) FITC-conjugated mouse anti-HBeAg antibody was added and bound to the antigen captured by the first antibody. Following incubation and wash steps, a horseradish peroxidase (HRP)-conjugated mouse anti-FITC antibody was added which bound to the FITC conjugated anti-HBeAg. Unbound HRP-conjugated mouse anti-FITC antibody was removed during a wash step, and substrate solution reactive with HRP consequently added to the wells. A colored product was formed in proportion to the amount of HBeAg present in the sample. The reaction was terminated by addition of acid and absorbance measured at 450 nm. A standard curve was prepared from recombinant HBeAg and sample concentration is then determined as either positive or negative.

3.5.4 Procedure for Hepatitis B Viral DNA Quantification

Cobas e 411 analyzer (© 2001-2010, Roche Diagnostics GmbH Cobas e411, Germany) was used to perform the COBAS® AmpliPrep/COBAS® TaqMan® HBV Test, v2.0, which is a nucleic acid amplification test for the quantitation of Hepatitis B Virus (HBV) DNA in human plasma and serum. Separation was done to obtain serum for HBV DNA load tests and measurement of HBV serum markers titer (HBsAg, anti-HBs, HBeAg, and anti-HBe). Sample
volume of 200 μL was used for the assay; 1.0 mL and repeat analyses as well as testing was carried out. Specimen were stored in plastic vials and sealed tightly and were stored in labeled 2 mL Nalgene cryovials or equivalent as per the manufacturer’s instructions. A total of 100 μl DNA samples were extracted using polyethylene glycol precipitation and alkaline lysis, to yield a final volume of 225 μl of processed sample.

Polymerase chain reaction (PCR) was accomplished by using 50 μl of the processed patient sample with the PCR reagents provided with the Amplicor kit. Amplification, detection, and quantitation was performed by using a COBAS Amplicor instrument (Roche Molecular Diagnostics). Samples with 200,000 HBV DNA copies/ml in the Amplicor assay was diluted to 1:900, 1:27,000, or 1:810,000, as necessary, in HBV DNA-negative normal human serum (NHS), and the diluted material were reextracted, amplified, and detection of cleaved dual-labeled oligonucleotide detection probe specific to the target to obtain a result within the analytical measurement range of the Amplicor assay. Final results for diluted samples was determined by multiplying the HBV DNA copies/milliliter obtained with the Amplicor assay by the dilution factor for that sample. Results were reported as HBV-DNA copies/milliliter by the COBAS Amplicor instrument.

3.6 Data Quality Control

Pretesting of the questionnaire was carried out at the antenatal clinics of Discove District Hospital in the adjacent Ahanta West District and Volta River Authority (VRA) Hospital in the neighbouring Shama District. Precision of questionnaires was established by clarifying obscure questions to the pregnant women. Repeated pretesting of the questionnaire was carried out to ensure that the tool was clearly understood by the pregnant women. Research assistants were
trained to understand and be well-acquainted with the objectives, interview techniques and data quality of the study and to make the necessary corrections or amendments. In addition, at close of the clinic days, the principal investigator collected and checked the completed forms and corrected errors that arose during the process, for completeness and consistency before data entry was done.

The Control Line used for procedural control during the neonatal HBsAg screening process was consistently checked to appear to ensure the test procedure was performed properly and the reagents of control line were working. To ensure quality, 10% of the negative HBsAg screening, HBsAg profile tests were repeated by different laboratory technician at the Holy Child Hospital in Takoradi.

Serum separation was performed for HBV e-antigen as evidence has shown that co-infection with the later variant confers higher risk of vertical transmissions. To validate each test batch, three calibrator controls, one negative control, as well as a positive control (provided by the manufacturer) were included in each run. An in-house working control known to be positive for HBeAg and a blank well containing only the substrate solution, were also included in each run.

During the HBV DNA amplification and quantification process personal protective equipment were worn, test results were measured most accurately and precisely, all installation and operating conditions were adhered to, as well as use of approved parts for the analyzer. Power interruptions were avoided operating only with an uninterruptible power supply and ensuring that power was switched off while the control unit accesses its memory or were connected to storage device. Required sample volume of 200 μL was used for the assay. Specimen were stored in plastic vials and sealed tightly to prevent desiccation of the sample. Serum plasma
samples were collected aseptically to minimize hemolysis and bacterial contamination. Samples were stored in labeled 2 mL Nalgene cryovials or equivalent.

3.7 Data Analysis
Data were entered, filtered and cleaned in Microsoft excel and later imported to STATA version 15 for analysis. Double entry verification of the questionnaires was done to ensure the validity of the entry. Graphical and tabular techniques included relative frequencies, percentages and proportions. P-value of <0.05 was considered as statistically significant at 95% confidence level. Descriptive statistics, (mean, standard deviation, percentages and frequency distribution) were used to summarize the quantitative data and to determine and proportions. HBsAg and HBeAg results were recorded as either positive or negative and HBV load results were recorded numerically. Simple percent was used to compare categorical variables, Pearson chi square to determine significant association and was presented as contingency table with p-values. The fisher’s exact test was used for variables with less than 5 frequencies and logistic regression models used for continuous variables to test for significance between neonatal hepatitis B transmission and the associated risk factors. The findings were presented as frequencies, percentages, crude odds ratio, adjusted odds ratio, 95% confidence interval and p-values.

3.8 Ethical Considerations
The study proposal was reviewed and approved by the Ghana Health Service Ethics Review Committee (GHS-ERC) with GHS-ERC Number GHS-ERC023/01/19. Permission was sought from the Western Regional Health Directorate, Sekondi Takoradi Metropolitan Health Directorate and Effia Kwesimintsim Municipal Health Directorate, and the community leaders before the beginning of the study that determined the maternal risk factors associated with
transmission of Hepatitis B virus among babies in the Sekondi Takoradi Metropolis and Effia Kwesimintsim Municipality.

Written informed consent was obtained from all participants. Participants were informed about the study and its importance was explained to enable them make informed decisions. The research team explained to the participants that the study was for research purposes and would provide information to help improve the health of the community.

3.8.1 Voluntary consent/ Withdrawal

The participants were made aware that participating in the study was voluntary and they had the right to participate, refuse to participate or withdraw from the study at any point in time without explanation. They were made aware of the fact that there was no direct benefits or risks involved in this study except for the fact that some of the questions may be a bit discomforting. The importance of the study was explained to the participants and assured of confidentiality throughout the study. The results obtained would be used only for the set objectives of this study. Participants were identified by code numbers and data stored both electronically and in hard copies with access given only to the research team.

3.8.2 Possible Risk and Discomfort

In participating in this study, participants were asked some questions, and blood samples were taken from them, and their newborn babies in the labour ward. Study participants were reassured that there would be no direct harm to them except for possible minor uneasiness when answering certain questions. Participants were informed that they might be uncomfortable about answering certain questions, and they and their babes would experience some temporary pain at the site of the needle puncture. They had the right to refuse to answer any question if it
made them uncomfortable, and to prevent us from taking blood samples from their babies. Participants were informed of the right to exit the study at any time that they wished. Assurance was given to study participants of proper sterilization that sites of venipuncture prior to blood sample collection, and puncture sites properly covered after the procedure, and all infectious waste disposed of in approved waste bins to ensure a sterile procedure. Participants were given the opportunity at the end of the interview to review their responses, and they could ask to change any responses that they wanted without giving us any reason for withdrawing.

3.8.3 Possible Benefits

Participants were made aware of the fact that there would be no direct benefits and were also given health education on viral hepatitis and benefits of screening and vaccination after the interview. Infected mothers were encouraged to get members of their household tested for early diagnosis and management of Hepatitis B. The findings would benefit the District Health Management Team in planning for health care delivery in the district.

3.8.4 Confidentiality

Participants were assured of confidentiality and that it would be maintained throughout the study and names of participants would be captured on questionnaires. The information would be securely stored without names and would only be accessible to the research team. Results of this study will be disseminated in such a way that no information will be linked to the identity of the participants.

3.8.5 Compensation

Study participants were made aware that being a part of this study was voluntary and there was no monetary compensation to the participants for accepting to be part of this study. Hepatitis B positive babies were referred to pediatricians for proper management to prevent liver disease.
Mothers of hepatitis B positive babies were reassured that early detection in their babies and rolling them onto care will ensure their babies’ well-being. Mothers were urged to bring all close family contacts for screening and vaccination.

3.8.6 Choice of Participation
Participants did not have to participate in this study if they did not wish to and they were informed that their refusal to participate would not attract any penalty. If they agreed to participate, they could reverse their consent and exit the study at any time. This would not affect them in any way.
CHAPTER FOUR

RESULTS

4.1 Demographic Characteristics of Mothers in the Study

A total of 89 Hepatitis B positive mothers participated in the study. The mean age of the mothers was 29.1 years (range: 17-39 years; SD = 29.12 ± 5.85) with seven (7.87%) of them within the age range of 17-19 years. The highest proportion (46.07%) of the study participants were within the age group of 20-29 years and 30-39 years. Majority (86.52%, 77/89) of them were married, 34.83% with basic education, and 42.70% (38/89) with secondary level education. Majority (87.64%) of the mothers were employed, while 12.36% were unemployed (Table 4.1).

Table 4.1: Descriptive Characteristics of Study Participants

<table>
<thead>
<tr>
<th>Variables</th>
<th>Frequency (n= 89)</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age group</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17-19</td>
<td>7</td>
<td>7.87</td>
</tr>
<tr>
<td>20-29</td>
<td>41</td>
<td>46.07</td>
</tr>
<tr>
<td>30-39</td>
<td>41</td>
<td>46.07</td>
</tr>
<tr>
<td><strong>Marital Status</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unmarried</td>
<td>12</td>
<td>13.48</td>
</tr>
<tr>
<td>Married</td>
<td>77</td>
<td>86.52</td>
</tr>
<tr>
<td><strong>Highest Educational Level</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Basic</td>
<td>31</td>
<td>34.83</td>
</tr>
<tr>
<td>Secondary</td>
<td>38</td>
<td>42.7</td>
</tr>
<tr>
<td>Tertiary</td>
<td>20</td>
<td>22.47</td>
</tr>
<tr>
<td><strong>Occupational Status</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Employed</td>
<td>78</td>
<td>87.64</td>
</tr>
<tr>
<td>Unemployed</td>
<td>11</td>
<td>12.36</td>
</tr>
</tbody>
</table>

SD: standard deviation.
4.2 Other Medical Conditions of the Mothers
Six of the mothers (6.7%) had other medical conditions. These included diabetes (2.2%, 2/89) G6PD (2) whilst one person each had hypertension, asthmatic, sickle cell and epilepsy (Figure 4.1).

![Figure 4.1: Other medical conditions of Hepatitis B positive mothers](http://ugspace.ug.edu.gh)

4.3 Background Characteristics of Babies Delivered by the Hepatitis B Positive Mothers
A total of sixteen babies (17.98%) of 89 mothers delivered by the eighty-nine hepatitis B positive mothers were found to be positive for hepatitis B virus infection. The mean birth weight of the babies was 3.1 kg (range: 2.5 – 4.5 Kg; SD = 3.1 + 0.4) with seven (7.87%) of them within the birth weight range of 4.0 – 4.5 Kg. Most of the babies were males, (56.2%, 56/89), 72.4% of them were delivered by spontaneous vaginal delivery, with all 89 babies (100%) delivered at term gestation. The mean length of babies at the time of delivery was 57.1 cm (range: 54-59 cm; SD = 57.1 + 0.9) with only one (7.87%) baby being 54 cm. Majority (44.3%)
of the study participants had a birth length of 57 cm. The babies’ mean head circumference at
birth was 37.0 (range: 35-39 cm; SD = 37.0 + 2.4). The mean duration of delivery of the babies
was 8.6 hours (range: 0.5-18; SD = 8.6 + 4.6), with a high proportion of babies (82.0%, 78/89)
receiving hepatitis B vaccination at the time of delivery. None of the babies had any observed
physical abnormalities at birth, and all babies (89, 100%) were breastfed (Table 4.2).
Table 4.2: Background Characteristics of Babies Delivered by the Hepatitis B Positive Mothers

<table>
<thead>
<tr>
<th>Variables</th>
<th>Frequency</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Weight of Baby at Birth</strong></td>
<td>3.07 + 0.40</td>
<td></td>
</tr>
<tr>
<td>2.5-2.9Kg</td>
<td>43</td>
<td>48.3</td>
</tr>
<tr>
<td>3.0-3.9Kg</td>
<td>39</td>
<td>43.8</td>
</tr>
<tr>
<td>4.0-2.5kg</td>
<td>7</td>
<td>7.9</td>
</tr>
<tr>
<td><strong>Sex of child</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>39</td>
<td>43.8</td>
</tr>
<tr>
<td>Male</td>
<td>50</td>
<td>56.12</td>
</tr>
<tr>
<td><strong>Gestation at Birth of Baby</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Term</td>
<td>89</td>
<td>100</td>
</tr>
<tr>
<td><strong>Mode of Delivery of Baby</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Caesarean</td>
<td>26</td>
<td>28.0</td>
</tr>
<tr>
<td>Vaginal</td>
<td>63</td>
<td>72.4</td>
</tr>
<tr>
<td><strong>Length of baby at birth (cm)</strong></td>
<td>57.09 + 0.91</td>
<td></td>
</tr>
<tr>
<td>54</td>
<td>1</td>
<td>1.14</td>
</tr>
<tr>
<td>56</td>
<td>22</td>
<td>23.86</td>
</tr>
<tr>
<td>57</td>
<td>39</td>
<td>44.32</td>
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<td>58</td>
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<td>25</td>
</tr>
<tr>
<td>59</td>
<td>5</td>
<td>5.7</td>
</tr>
<tr>
<td><strong>Head Circumference of Baby at Birth (cm)</strong></td>
<td>37.02 + 2.41</td>
<td></td>
</tr>
<tr>
<td>35</td>
<td>2</td>
<td>2.3</td>
</tr>
<tr>
<td>36</td>
<td>35</td>
<td>38.2</td>
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<td>37</td>
<td>37</td>
<td>41.6</td>
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<tr>
<td>38</td>
<td>11</td>
<td>12.4</td>
</tr>
<tr>
<td>39</td>
<td>4</td>
<td>4.5</td>
</tr>
<tr>
<td><strong>Duration of Birth of Baby</strong></td>
<td>8.58 + 4.63</td>
<td></td>
</tr>
<tr>
<td><strong>Hepatitis B Vaccination to Baby</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hepatitis B vaccine not given</td>
<td>11</td>
<td>12.4</td>
</tr>
<tr>
<td>Hepatitis B vaccine given</td>
<td>78</td>
<td>87.6</td>
</tr>
<tr>
<td><strong>Physical Abnormality Present in Baby</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No Physical Abnormality Present</td>
<td>89</td>
<td>100</td>
</tr>
<tr>
<td><strong>Baby Breastfed</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Breast fed</td>
<td>89</td>
<td>100</td>
</tr>
</tbody>
</table>
4.4 Association Between the Infection Status of Babies and Mother’s Demographic Characteristics

The Pearson’s chi-square test of association was used to assess the demographic characteristics of mothers that are associated with the infection status of the current child. From Table 4.3, none of the demographic characteristics showed significant association with the infection status of their current child (p-value > 0.05).

Of the seven mothers who were within the age range of 17-19 years, only one of their current children was infected, seven (17.07%) of the 41 mothers within the 20-29 years age range had their current children infected, and eight (19.51%) of those mothers within the 30-39 years age range had their children infected. Amongst the 12 mothers unmarried, 2 (16.67%) of them had their children infected whilst 14 (18.18%) of the 77 mothers who were married had the current children infected (Table 4.3).
Table 4.3: Association between demographic characteristics of Hepatitis B positive mothers and the infection status of their babies

<table>
<thead>
<tr>
<th>Variables</th>
<th>N</th>
<th>Negative n (%)</th>
<th>Positive n (%)</th>
<th>chi-square</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age group</strong></td>
<td></td>
<td></td>
<td></td>
<td>0.15</td>
<td>0.926</td>
</tr>
<tr>
<td>17-19 years</td>
<td>7</td>
<td>6 (85.71)</td>
<td>1 (14.29)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>20-29 years</td>
<td>41</td>
<td>34 (82.93)</td>
<td>7 (17.07)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>30-39 years</td>
<td>41</td>
<td>33 (80.49)</td>
<td>8 (19.51)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Marital status</strong></td>
<td></td>
<td></td>
<td></td>
<td>0.02</td>
<td>0.899</td>
</tr>
<tr>
<td>Unmarried</td>
<td>12</td>
<td>10 (83.33)</td>
<td>2 (16.67)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Married</td>
<td>77</td>
<td>63 (81.82)</td>
<td>14 (18.18)</td>
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</tr>
<tr>
<td><strong>Highest educational level</strong></td>
<td>5.00</td>
<td>0.082</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Basic</td>
<td>31</td>
<td>29 (93.55)</td>
<td>2 (6.45)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Secondary</td>
<td>38</td>
<td>30 (78.95)</td>
<td>8 (21.05)</td>
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</tr>
<tr>
<td>Tertiary</td>
<td>20</td>
<td>14 (70.00)</td>
<td>6 (30.00)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Occupational status</strong></td>
<td>1.43</td>
<td>0.49</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Government/Private</td>
<td>29</td>
<td>22 (75.86)</td>
<td>7 (24.14)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Self-employed</td>
<td>49</td>
<td>41 (83.67)</td>
<td>8 (16.33)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unemployed</td>
<td>11</td>
<td>10 (90.91)</td>
<td>1 (9.09)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

n: frequency. %: percentage.
4.5 Association Between Obstetric and Family Characteristics of Hepatitis B Positive Mothers and the Infection Status of their Babies

Using Pearson’s chi-square test the obstetric and family characteristics of the mother that were found to be associated with the infection status of their current child were determined. From Table 4.4, the weight of the baby was the only factor significantly associated with the infection status of the child (p-value <0.05).

A higher percentage of babies who weigh 3-3.9 kg were significantly infected compared to those babies who weighed 2.5-2.9 kg (27.45% vs. 6.06%) and those who weigh 4-4.5kg (27.45% vs. 0.00%). Of the 78 mothers who had not had previous caesarean section, 15 (19.23%) of them had their current child infected whilst only one child of the 11 mothers who have had previous caesarean section were infected. Equal number of the male children were infected as was the female children. Detailed analysis of the association between obstetric and family characteristics and the infection status of the current children of the hepatitis B positive mothers shown table 4.4.
<table>
<thead>
<tr>
<th>Variables</th>
<th>N (n=89)</th>
<th>Infection status of baby</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Negative n (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Positive n (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>chi-square</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>P-value</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Weight of baby</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2.5-2.9Kg</td>
<td>33</td>
<td>31 (93.94)</td>
<td>2</td>
<td>6.06</td>
<td>7.38</td>
<td>0.021*</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3-3.9Kg</td>
<td>51</td>
<td>37 (72.55)</td>
<td>14</td>
<td>27.45</td>
<td>0.67</td>
<td>0.681</td>
<td></td>
<td></td>
</tr>
<tr>
<td>4-4.5kg</td>
<td>5</td>
<td>5 (100.00)</td>
<td>0</td>
<td>0.00</td>
<td>0.67</td>
<td>0.681</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previous caesarean section</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No Previous Caesarean section</td>
<td>78</td>
<td>63 (80.77)</td>
<td>15</td>
<td>19.23</td>
<td>0.67</td>
<td>0.681</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previous Caesarean</td>
<td>11</td>
<td>10 (90.91)</td>
<td>1</td>
<td>9.09</td>
<td>0.67</td>
<td>0.681</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Preferred mode of Delivery</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Caesarean</td>
<td>5</td>
<td>3 (60.00)</td>
<td>2</td>
<td>40.00</td>
<td>1.74</td>
<td>0.219</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vaginal</td>
<td>84</td>
<td>70 (83.33)</td>
<td>14</td>
<td>16.67</td>
<td>1.74</td>
<td>0.219</td>
<td></td>
<td></td>
</tr>
<tr>
<td>History of Hepatitis B infection</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No knowledge</td>
<td>28</td>
<td>25 (89.29)</td>
<td>3</td>
<td>10.71</td>
<td>1.47</td>
<td>0.48</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No History</td>
<td>24</td>
<td>19 (79.17)</td>
<td>5</td>
<td>20.83</td>
<td>1.47</td>
<td>0.48</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Positive History</td>
<td>37</td>
<td>29 (78.38)</td>
<td>8</td>
<td>21.62</td>
<td>1.47</td>
<td>0.48</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Husband has hepatitis B</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No knowledge</td>
<td>38</td>
<td>32 (84.21)</td>
<td>6</td>
<td>15.79</td>
<td>1.78</td>
<td>0.381</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Husband not infected</td>
<td>46</td>
<td>38 (82.61)</td>
<td>8</td>
<td>17.39</td>
<td>1.78</td>
<td>0.381</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Husband Infected</td>
<td>5</td>
<td>3 (60.00)</td>
<td>2</td>
<td>40.00</td>
<td>1.78</td>
<td>0.381</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sex of current baby</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>40</td>
<td>32 (80.00)</td>
<td>8</td>
<td>20.00</td>
<td>0.13</td>
<td>0.721</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>49</td>
<td>39 (82.98)</td>
<td>8</td>
<td>17.02</td>
<td>0.13</td>
<td>0.721</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hepatitis B vaccine administered</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vaccine administered</td>
<td>5</td>
<td>5 (100.00)</td>
<td>0</td>
<td>0.00</td>
<td>1.03</td>
<td>0.586</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 4.4: Association Between Obstetric and Family Characteristics of Hepatitis B Positive Mothers and The Infection Status of Their Babies
Vaccine not administered 84 67 (82.72) 14 (17.28)

Mode of current delivery 0.016 0.969
Caesarean 17 14 (78.95) 3 (17.65)
Vaginal 72 59 (83.05) 13 (18.06)

n: frequency. %: percentage. *: P-value <0.05.

4.6 Differences in Means of Continuous Variables by the Infection Status of the Baby
The t-test was used to test the differences in means of continuous variables by the infection status of the baby. From Table 4.5, weight of baby at birth in kilograms was the only variable that showed significant difference with respect to the infection status of the baby. The mean weight at birth of babies with infection was significantly higher than the mean weight at birth of babies without infection (3.33kg vs. 3.12kg, p-value=0.043).
Table 4.5: Differences in Means of Continuous Variables by The Infection Status of the Baby

<table>
<thead>
<tr>
<th>Continuous variables</th>
<th>Infection Status of Baby</th>
<th>t-statistic</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Negative n=16</td>
<td>Positive n=16</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Mean ± SD</td>
<td>Mean ± SD</td>
<td></td>
</tr>
<tr>
<td>Number of Live Children</td>
<td>2.23 ± 1.29</td>
<td>2.19 ± 1.11</td>
<td>0.14</td>
</tr>
<tr>
<td>Weight of Baby at Birth (Kg)</td>
<td>3.12 ± 0.39</td>
<td>3.33 ± 0.35</td>
<td>-2.13</td>
</tr>
<tr>
<td>Age of mother (years)</td>
<td>29.07 ± 5.97</td>
<td>29.38 ± 5.46</td>
<td>-0.20</td>
</tr>
<tr>
<td>Length of Baby at Birth (cm)</td>
<td>56.78 ± 0.89</td>
<td>56.55 ± 0.69</td>
<td>0.94</td>
</tr>
<tr>
<td>Head Circumference of Baby (cm)</td>
<td>36.68 ± 0.63</td>
<td>38.68 ± 6.17</td>
<td>-1.08</td>
</tr>
<tr>
<td>Duration of Delivery (hours)</td>
<td>9.32 ± 4.47</td>
<td>10.5 ± 4.77</td>
<td>-0.84</td>
</tr>
<tr>
<td>Gestational Age at 1ST ANC (weeks)</td>
<td>10.68 ± 4.27</td>
<td>9.81 ± 3.06</td>
<td>0.95</td>
</tr>
<tr>
<td>Number of Deliveries Done so far</td>
<td>2.29 ± 1.24</td>
<td>2.19 ± 1.11</td>
<td>0.33</td>
</tr>
</tbody>
</table>

SD: standard deviation.

4.7 Equality of Means of Viral Load of Mothers and the Infection Status of their Babies

Viral load of 32 of the 89 mothers were obtained by testing all 16 mothers whose babies were HBsAg positive and randomly selecting and testing an equal number of mothers (16) whose babies tested HBsAg negative. The Welch’s t-test was used to test the equality of means of the viral load of mothers whose current baby was infected and those whose current baby was not infected. The mean viral load of the mothers whose current baby was not infected was 935.18 IU/mL which was significantly lower compared to the mean viral load of 4178.63 IU/mL of mothers whose current baby was infected (p-value <0.001) in Table 4.6.
Table 4.6: Equality of Means of Viral Load Of Mothers by the HBsAg Status of Their Baby

<table>
<thead>
<tr>
<th>HBsAg Status of Baby</th>
<th>n</th>
<th>Mean</th>
<th>SD</th>
<th>95% CI</th>
<th>t-statistic</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>HBsAg Negative</td>
<td>16</td>
<td>935.18</td>
<td>728.00</td>
<td>[560.87, 1309.48]</td>
<td>-7.05</td>
<td>&lt;0.001***</td>
</tr>
<tr>
<td>HBsAg Positive</td>
<td>16</td>
<td>4178.63</td>
<td>1698.71</td>
<td>[3273.45, 5083.80]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>32</td>
<td>2507.76</td>
<td>2080.22</td>
<td>[1770.15, 3245.37]</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

SD: standard deviation. n: frequency. CI: confidence interval. ***: P-value <0.001.

The One-way ANOVA test was also used to test the equality of means of the viral load of mothers and their demographic characteristics. None of the demographic characteristics of the mother (Age group, Marital status, highest level of education, and occupational status) showed significant association with the viral load of hepatitis B positive mothers.
Table 4.7: Equality of Means of Viral Load of Mothers by Their Demographic Characteristics

<table>
<thead>
<tr>
<th>Variables</th>
<th>Frequency</th>
<th>Mean</th>
<th>Standard deviation</th>
<th>Test Statistic</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age group</strong></td>
<td></td>
<td></td>
<td></td>
<td>2.02°F</td>
<td>0.150</td>
</tr>
<tr>
<td>17-19 years</td>
<td>2</td>
<td>2500.00</td>
<td>2893.48</td>
<td></td>
<td></td>
</tr>
<tr>
<td>20-29 years</td>
<td>10</td>
<td>3564.40</td>
<td>2509.92</td>
<td></td>
<td></td>
</tr>
<tr>
<td>30-39 years</td>
<td>21</td>
<td>2005.33</td>
<td>1687.28</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Marital status</strong></td>
<td></td>
<td></td>
<td></td>
<td>1.42ᵗ</td>
<td>0.242</td>
</tr>
<tr>
<td>Unmarried</td>
<td>3</td>
<td>3865.33</td>
<td>3127.06</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Married</td>
<td>30</td>
<td>2372.00</td>
<td>1972.52</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Highest educational level</strong></td>
<td></td>
<td></td>
<td></td>
<td>2.18°F</td>
<td>0.130</td>
</tr>
<tr>
<td>Basic</td>
<td>11</td>
<td>1499.64</td>
<td>1365.07</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Secondary</td>
<td>12</td>
<td>2835.25</td>
<td>2108.15</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tertiary</td>
<td>10</td>
<td>3223.70</td>
<td>2434.94</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Occupational status</strong></td>
<td></td>
<td></td>
<td></td>
<td>0.13°F</td>
<td>0.877</td>
</tr>
<tr>
<td>Government/Private worker</td>
<td>12</td>
<td>2728.50</td>
<td>2189.29</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Self-employed</td>
<td>19</td>
<td>2343.32</td>
<td>2082.41</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unemployed</td>
<td>2</td>
<td>2745.50</td>
<td>2546.29</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

SD: standard deviation. t = t test. F = One-way ANOVA test.

4.8 Association Between Infection Status of Baby and the Hepatitis B Envelope-Antigen Status of Mother and Mode of Delivery of Baby

From Table 4.8, the prevalence of infection among babies whose mothers were hepatitis B e-Antigen positive was significantly higher than babies whose mothers were hepatitis B e-Antigen negative (81.82% vs. 31.82%, p-value = 0.007). The mode of delivery of the current child did not show significant association with the infection status of the baby as 4 (21.05%) of the 17 babies who were delivered through caesarean section were infected compared to 10 (16.95%)
of the 72 babies who were delivered by spontaneous vaginal delivery were also infected (p-value = 0.685) Table 4.7.

Table 4.8: Association Between Infection Status of Baby and the Hepatitis B e-Antigen Status of Mother and Mode of Delivery of Baby

<table>
<thead>
<tr>
<th>Variables</th>
<th>Infection Status of Baby</th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>n</td>
<td>Negative n (%)</td>
<td>Positive n (%)</td>
<td>chi-square</td>
</tr>
<tr>
<td>Hepatitis B e-Antigen Status</td>
<td></td>
<td>7.34</td>
<td>0.007**</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hepatitis B e-Antigen Negative</td>
<td>22</td>
<td>15 (68.18)</td>
<td>7 (31.82)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hepatitis B e-Antigen Positive</td>
<td>11</td>
<td>2 (18.18)</td>
<td>9 (81.82)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mode of Delivery</td>
<td></td>
<td>0.016</td>
<td>0.969</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Caesarean</td>
<td>17</td>
<td>14 (82.35)</td>
<td>3 (17.65)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vaginal</td>
<td>72</td>
<td>59 (81.94)</td>
<td>13 (18.06)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

n: frequency. %: percentage. **: p-value <0.01.

4.9 Binary Logistic Regression Analysis of Mode of Delivery and Hepatitis B e-Antigen on Positive Infection Status of Baby

From Table 4.8, the mode of delivery of the baby did not show any significant influence on the infection status of the baby. However, the Hepatitis B e-Antigen status of the mother showed significant influence on the baby both in the unadjusted (simple binary logistic) model and the adjusted (multiple binary logistic) model (p-value<0.05).

From the unadjusted model, the odds of a baby being infected was 7.85 (95% CI:1.52-40.63) times higher among those whose mothers were Hepatitis B e-Antigen positive compared to
those who were negative. After adjusting for the weight at birth and mode of delivery of the baby, the adjusted odds of a baby being infected was 8.32 (95% CI:1.52-45.55) times higher among those whose mothers who were hepatitis B e-Antigen positive compared to those who were negative (Table 4.9).

Table 4.9: Binary Logistic Regression Analysis of Mode of Delivery and Hepatitis B e-Antigen on Positive Infection Status of Baby

<table>
<thead>
<tr>
<th>Variables</th>
<th>Simple Binary Logistic</th>
<th>Multiple Binary Logistic</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>COR (95% CI)</td>
<td>P-value</td>
</tr>
<tr>
<td>Weight of Baby at Birth</td>
<td>3.46 (0.96 - 12.48)</td>
<td>0.058</td>
</tr>
<tr>
<td>Mode of Delivery</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Caesarean Section</td>
<td>1.00 (reference)</td>
<td></td>
</tr>
<tr>
<td>Vaginal Delivery</td>
<td>0.94 (0.25 - 3.48)</td>
<td>0.926</td>
</tr>
<tr>
<td>Hepatitis B e-Antigen Status</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Negatives</td>
<td>1.00 (reference)</td>
<td></td>
</tr>
<tr>
<td>Positive</td>
<td>7.85 (1.52 - 40.63)</td>
<td>0.014*</td>
</tr>
</tbody>
</table>

COR: Crude odds ratio. AOR: Adjusted odds ratio. CI: Confidence Interval. *: p-value<0.05.
5.1 Prevalence of infections among Babies born to Hepatitis B positive mothers

This study sought to determine maternal factors associated with transmission of hepatitis B virus infection from mothers to their babies in selected health facilities in the Western Region of Ghana. Positivity status of neonates was determined by their testing positive to Hepatitis B surface antigen.

Overall, 17.9% of the babies were found to be positive for Hepatitis B virus infection. This was found to be close to an earlier report by Yelemkoure (2018) from Burkina Faso, where mother-to-child transmission was found to be 23.8%. Conversely, Liu and associates in a study among mothers who were given immunoprophylaxis detected a 3.9% prevalence which was lower than what has been found in the current study though there is variation in the study design and used in both instances, with a shorter duration in their study (Liu, Zeng, Zhou, Chen, & Hu, 2014).

Torii and colleagues in a study in Japan using cord blood of babies showed that five out of 17 (29.4%) babies had prenatal HBV infection at birth. There is currently routine testing for HBV at the ante-natal clinics in Ghana, but no definitive government policy on at-birth dose vaccine with its established significant reduction in perinatal transmission of Hepatitis B viral (HBV) infections (Awuku & Yeboah-Afihene, 2018 ); (Henderson, Webber, & Bean, 2015).

According to the WHO and UNICEF estimates of national immunization coverage Report, 2019, no estimates were made for infant immunization on hepatitis B in Ghana since estimates of hepatitis B birth dose coverage are made for countries with a universal birth dose policy
(WHO and UNICEF, 2019). Consequently, the prevalence determined in this study is high, and is similar to the universal claims made in other studies that the prevalence of Hepatitis B infection among neonates of infected pregnant women is high in developing countries.

5.2 Association between Obstetric and family characteristics of Hepatitis B positive mothers and the infection status of their babies

The weight of the babies was the only factor among these characteristics determined to be significantly associated with the infection status (p-value <0.05) and majority of babies who weigh 3-3.9 kg were infected compared to those babies who weigh 2.5-2.9 kg (27.45% vs. 6.06%) and those who weigh 4-4.5kg (27.45% vs. 0.00%).

Kashyap & Bineeta (2017) illustrated that preterm and Low Birth Weight (LBW) infants are at a greater risk of increased morbidity from vaccine-preventable diseases such as Hepatitis B Viral (HBV) infection owing to the fragility and ability to develop protective immunity due to immature or impaired cellular and humoral immune systems of such infants. This study detected that a proportionally higher number of hepatitis B positive babies with high birth weight, possibly due to the use of a small sample size. Such babies tend to have a stronger, well-developed immune response to the HBV, and hence a better prognosis compared to the HBV-positive babies with LBW.

This finding conforms similarly to that of Bajema et al (2014) in a Population-Based Cohort Study in the United States which reported that women with HBV had a reduced risk of delivering Small for Gestational Age (SGA) infants (13% among HBV-positive vs 10% among HBV-negative (Bajema, Karita, Tenforde, Hawes, & Heffron, 2014).

A population-based cohort study by Liu et al, 2014 in China, however, found that the preterm birth rate, with its consequential low birth weight was 5-2% for women who were not infected with
hepatitis B virus, 6.5% for women who were positive for HBsAg and negative for HBeAg, and 6.2% for women who were positive for both HBsAg and HBeAg.

Multivariable adjusted analyses also showed a significant association between maternal pre-pregnancy hepatitis B virus infection, preterm birth and low birth weight (Liu et al., 2014).

5.3 Association between Maternal Viral Load and Babies Born to Hepatitis B-Positive Mothers
The mean viral load of the mothers whose current child was not infected was significantly lower as compared to that of mothers whose current child was infected (p-value <0.001), and mother-to-child transmission was 18.0%.

Maternal HBV DNA level is the single most important prognosticator of mother-to-child transmission (MTCT) and levels of 106 to 108 copies/mL were associated with very high transmission risk.

MTCT occurs, especially during the third trimester of pregnancy and intrapartum period. Yelemkoure and colleagues, in a study in Burkina Faso among 21 pregnant women, reported a rate of 23.81% (5/21) (Yelemkoure et al., 2018).

Burgis and colleagues (2017) also found that perinatal transmission of chronic Hepatitis B infection occurred in infants with maternal HBV DNA levels < 2 × 107 IU/mL, received appropriate post-exposure prophylaxis (PEP) at birth and completed the HBV vaccine series in the appropriate time period (Burgis et al., 2017). These infants were born at term via normal spontaneous vaginal deliveries.

Transmission with high HBV DNA levels were, however, independent of the HBeAg status of the mothers (Burgis et al., 2017). This agrees with findings by (Schillie and associates, 2015)
that a greater proportion of infected infants were born to women with viral loads \( \geq 2000 \) IU/mL compared with \(<2000\) IU/mL (Schillie, Walker, Veselsky, Crowley, & Dusek, 2015). On the contrary, Belopolskaya (2015) found minimal, non-significant correlation between HBsAg and HBV DNA levels which can be explained by a large group of patients with an unmeasurable value of HBV DNA (between 50 and 150 IU/mL) in that study Hepatitis B Surface Antigen (HBsAg) level and hepatitis b viral load correlation with focus on pregnancy (“HBsAG level and hepatitis b viral load correlation with focus on pregnancy,” 2015).

5.4 Association between Maternal Hepatitis B e-Antigen Positive and Babies Born to Hepatitis B-Positive Mothers

The prevalence of HBV infection among children whose mothers were hepatitis B e-Antigen positive was significantly higher than children whose mothers were hepatitis B e-Antigen negative (81.82% vs. 31.82%, p-value 0.007). After adjusting for the weight at birth and mode of delivery of the child, the odds of a child being infected was 8.32 (95% CI:1.52-45.55) times higher among those whose mothers were hepatitis B e-Antigen positive compared to those who were negative. Geeta and Riyaz (2013) reported that without immune prophylaxis in mothers who are both HBsAg and HBeAg positive, the risk for transmission to the baby is between 70% and 90%, whereas it is less than 10% in HBeAg negative mothers (Riyaz, 2013).

Additionally, increased risk of transmission with HBeAg positive status resulted in 38% of babies born to HBsAg positive mothers (Keane et al., 2016)(Dyson et al., 2014) (Rufai, , Mohamed Mutocheluh, & Elliot Dogbe, 2014). Liu et al (2015) also recounted that all infected infants were born to HBeAg-positive mothers with a high HBV DNA level. Conversely, Yelemkoure (2018) found the rate of MTCT in HBeAg-positive mothers to be low at 4.5%, whilst Fessehaye and
colleagues (2018) reported 3.9% MTCT among HBsAg positive pregnant women who are HBeAg negative, compared to 3.2% in HBeAg-positive, indicating a reduced infectivity (Fessehaye et al., 2018).

The low HBeAg prevalence amongst mothers of infected babies suggest HBeAg-negative hepatitis B infection is highly prevalent among the study population as has been determined worldwide. It can also be explained by mothers with virological mutant variants which offer it a replication advantage, facilitate immune escape or cause resistance to antiviral drugs that can be preferentially selected (Alexopoulou & Karayiannis, 2014).

5.5 Association between Mode of Delivery and Babies Born to Hepatitis B-Positive Mothers

The mode of delivery of the current child did not show significant association with the infection status of the child as 21.05% (4/19) of children who were delivered through caesarean section were infected compared to 16.95% (10/59) who were delivered by spontaneous vaginal delivery (p-value = 0.685).

Yang and colleagues (2017) found out that mother-to-child transmission (MTCT) of hepatitis B virus was 6.76% (670/9906), where MTCT was 4.37% (223/5105) for mothers who underwent cesarean section and 9.31% (447/4801) for vaginal delivery, indicating a statistically significant reduction in HBV vertical transmission with cesarean section (Yang, Qin, Fang, Jiang, & Nie, 2017). A significant effect of cesarean section on the MTCT of HBV was found in a larger percentage of infants who had been administered Hepatitis B Immunoglobulin (HBIG) at birth (Yang et al., 2017).
Nguyen (2013) agrees with these findings, and reported a statistically significant absolute risk reduction of 17.5% and a relative risk of 0.41 among 789 pregnant mothers who underwent elective cesarean section with appropriate HBIg immunoprophylaxis given to their babies compared with immunization of HBV vaccine at birth alone (Nguyen & Nguyen, 2013). They also found that only among HBeAg-positive mothers at birth, 47.8% (670/1401), mother-to-child transmission occurred in 5.2% of infants with the lower rate in the elective cesarean section group 2.6% (7/273) compared with both vaginal delivery 6.3% (23/365). The observed differences in study results can be attributed to the large study population and long duration of study in the meta-analysis.

5.6 Limitations
The ideal study type for this research should have been cohort study that would have reduced loss to follow up of participants recruited at the beginning of the study, instead of the cross-sectional approach employed.

A larger sample size using the upper limit of the prevalence would have been more appropriate to detect associations for mother-to-child transmission of hepatitis B in this study. Additionally, only hepatitis B positive pregnant women recruited whose babies were HBsAg positive had their tested hepatitis B viral DNA and e-antigen tested, which may have masked some true associations in the maternal factors for perinatal transmission.

Not all pregnant women delivered during the study period due to improper ultrasound scan dating of pregnancies, or post-dates, and it was not feasible that all babies born to hepatitis B positive pregnant women might have been screened during the study period. Selection bias might have occurred during recruitment of study participants by including all hepatitis B positive mothers, with no randomization in the selection of study participants.
CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusion

In this study, maternal factors associated with transmission of hepatitis B viral infection from mothers to their babies in selected health facilities were determined in Sekondi Takoradi in the Western Region of Ghana. The study demonstrated a prevalence of 17.98% neonatal hepatitis B viral infection amongst babies delivered by the hepatitis B positive mothers. This level of prevalence is higher than the global prevalence of up to 10%.

Prevalence of infection among children whose mothers were hepatitis B e-Antigen positive was significantly higher than children whose mothers were hepatitis B e-Antigen negative. The odds of a child being infected was 8.32 times higher among those whose mothers were Hepatitis B e-Antigen positive compared to those who were negative.

The weight of the baby at birth was the only demographic characteristic significantly associated with the infection status of the baby. The mean birth weight of infected babies was significantly higher than the mean birth weight of those without infection and highest amongst babies who weighed 3-3.9 kg.

The mean viral load of the mothers whose current baby was not infected was 935.18 IU/mL which was significantly lower compared to the mean viral load of 4178.63 IU/mL of mothers whose current baby was infected.

The mode of delivery of the current baby, whether vaginal or Caesarean, however, did not show significant association with the infection status of the baby. Hence, mother-to-child
transmission of Hepatitis B Virus (HBV) in this study occurred in all babies with higher birth weight, and whose mothers had high Hepatitis B viral load and Hepatitis B e-Antigen positive.

6.2 Recommendations

1. The Ministry of Health, together, with the Ghana Health Service, Regional and District Directors of Health Services and all stakeholders to institute protocols that incorporate HBV DNA and Hepatitis B profile screening as part of routine Antenatal care to detect mothers with high viral load and e-Antigen positive status.

2. The Ministry of Health and other stakeholders needs to increase efforts to use antivirals to reduce viral load and at-birth vaccination within 24 h of the newborn combined with the current EPI protocol in Ghana need to be encouraged to reduce transmission significantly.
REFERENCES


APPENDICES

Appendix I: Questionnaire

Maternal Factors Associated with Hepatitis B Transmission to Neonates at Selected Antenatal Clinics, Sekondi Takoradi Metropolis and Effia Kwesimintsim Municipality, 2019

Dear Respondent,

Kindly take a few minutes to answer these questions. We assure you of strict confidentiality of the answers you provide.

Unique identifier for respondents .................................

Date of interview ....................................................
A. Demographic Characteristics:

1. Name of Facility:

<table>
<thead>
<tr>
<th>2. Age (years)</th>
<th>3. Sex:</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Male</td>
</tr>
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</table>

<table>
<thead>
<tr>
<th>Parity</th>
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</thead>
</table>

5. Occupation:

B. Medical History

Do you have any of these medical conditions? Tick under appropriate heading

<table>
<thead>
<tr>
<th>Medical Condition</th>
<th>Yes</th>
<th>No</th>
<th>Don’t Know</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIV</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Hypertension</td>
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<td></td>
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<tr>
<td>Diabetes</td>
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<tr>
<td>Asthma</td>
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<td></td>
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<tr>
<td>Sickle Cell</td>
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<tr>
<td>Epilepsy</td>
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</table>

<table>
<thead>
<tr>
<th>Medical Condition</th>
<th>Yes</th>
<th>No</th>
<th>Don’t Know</th>
</tr>
</thead>
<tbody>
<tr>
<td>G6PD</td>
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<tr>
<td>Thalassemia</td>
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<tr>
<td>Chronic Kidney Disease</td>
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<tr>
<td>Chronic Liver Disease</td>
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</tbody>
</table>
C. Obstetric History

1. Last Menstrual Period (dd/mm/yyyy): 

2. Previous Caesarean Section? 
   Yes ☐ No ☐ Don’t Know ☐

3. Complications during Current Pregnancy? 
   Yes ☐ No ☐ Don’t Know ☐
   (If Yes, please specify …………………………………………………………………)

4. Preferred Mode of Delivery: 
   A. SVD ☐
   B. Cesarean Section ☐

5. Antiviral Therapy during Pregnancy 
   Yes ☐ No ☐ Don’t Know ☐

D. Family History

1. History of Child with Hepatitis B Infection 
   Yes ☐ No ☐ Don’t Know ☐

2. Husband Has Hepatitis B Infection 
   Yes ☐ No ☐ Don’t Know ☐
E. Babies Birth History

1. Hepatitis B Vaccine Administered                   Yes ☐ No ☐ Don’t Know ☐

2. Birth weight (Kg) ................................................

3. Gestation                                      Term ☐ Preterm ☐ Don’t Know ☐

4. Mode of Delivery                                Vaginal ☐ Cesarean ☐ Assisted Delivery ☐

5. Duration of Delivery                            ..................................................

6. Physical Abnormalities                          Yes ☐ No ☐ Don’t Know ☐

   If yes to 6 above, please state of abnormality             ..................................................

7. Breastfeeding                                   Yes ☐ No ☐ Don’t Know ☐

Thank you.

Appendix II: Information on The Study

School of Public Health
**Introduction**

My name is Dr. Christabel Ayepah, a student of the School of Public Health, University of Ghana, Legon. As part of the academic requirements, I am conducting a research to determine Maternal Factors Associated with Hepatitis B Transmission to Neonates At Selected Antenatal Clinics in the Sekondi Takoradi Metropolis and Effia Kwesimintsim Municipality, 2019. The study will involve all pregnant women who have Hepatitis B infection at these Clinics. As part of the study, you have been selected to help in obtaining information for this study. We will obtain this information that from you, by the use of a questionnaire and taking blood samples from you and your newborn baby. We will keep the information confidential and use for research purpose only. If you agree to be part of this study, it will involve answering some questions that we will ask you. This is an academic research, which forms part of my work for the award of Master of Philosophy Degree. I will be very grateful to have you as part of this study. The process is expected to last between 20 to 30 minutes.
Potential Risks and Discomforts

In participating in this study, we will ask you some questions and we will take blood samples from you and your newborn babies in the labour ward. You may however experience some minor discomfort when answering certain questions, and you and your babes will experience some temporary pain at the site of the needle puncture. We will however, adhere to all the Infection Prevention and Control Practices to ensure a sterile procedure. You may refuse to answer any question if you feel uncomfortable about it. You may also withdraw from the study at any time that you wish. We will give you will an opportunity at the end of the interview to review your responses, and you can ask to change any responses that you want. You do not have to give me any reason for withdrawing.

Additional Costs and Compensation

No money will be demanded from you for taking part in this study and you do not have to participate in this study if you do not wish to. Your refusal to participate will not attract any penalty. If you agree to participate, you can withdraw consent and discontinue participation at any time. This will not affect you in any way. Your participation in this study is purely voluntary and no financial compensation will be given for participating in the study.

Confidentiality
We will not disclose information shared to any of your community members or to anyone who is not part of the study team and your name is not needed in this study. We will carry out data analysis at the aggregate level to ensure anonymity. We will use the information that we will collect from this study for academic purposes only.

**Dissemination of results**

We will mail the result of this study to you if you provide your address below. Before taking the consent, do you have any question you wish to ask about the study?

Yes □  (if yes, questions to be noted)
No □

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**Contacts for Additional Information**
If you have any questions, you can ask them now or later. If you wish to ask questions later, you may contact the principal investigator Dr. Christabel Ayepah, School of Public Health, Legon on this telephone number 0206 300 443.

You can also contact the GHS-ERC Administrator, Madam Hannah Frimpong on 0507041223 for any clarifications on this research.
Appendix III: Certificate of Consent

I have had the information read and adequately explained to me for the study on “Maternal Factors Associated with Hepatitis B Transmission to Neonates in the Sekondi Takoradi Metropolis and Effia Kwisimintsim Municipality”. The document describing the nature and purpose as well as risks and benefits of the study has been read and explained to me. The research team has me given an opportunity to have any questions about the study answered to my satisfaction. I agree voluntarily to participate in this study.

......................................................... ..............................................
Signature or Thumbprint of Participant   Date

If a participant cannot read the document, then a Witness is needed:
I was present during the reading and explanation of the consent document to the participant. All questions from the participant were duly answered and the participant agreed to participate in the study.

......................................................... ..............................................
Signature of Witness                      Date

I certify that the purpose and nature of the research, the potential benefits and possible discomforts associated with participating in this research have been explained to the participant who has agreed to voluntarily participate.

......................................................... ..............................................
Signature of Person Who Obtained Consent  Date