SCHOOL OF PUBLIC HEALTH,  
COLLEGE OF HEALTH SCIENCES,  
UNIVERSITY OF GHANA

PREVALENCE OF HEPATITIS B AMONG PREGNANT WOMEN IN GHANA

BY

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DECLARATION
I, Rexford Bempong Adade, declare that under the guidance of my supervisor, Dr Kofi Mensah Nyarko, School of Public Health, University of Ghana-Legon, this dissertation is my original work, except for related works that have been duly referenced, and that no form of it has been presented elsewhere for another degree.

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ABSTRACT

Background: Hepatitis is a disease defined by the inflammation of the liver and it is characterized by the existence of inflammatory cells in the tissues of the liver leading to fibrosis or cirrhosis. Viral hepatitis has become a global public health threat affecting millions of people yearly, causing disability and mortality. There are an estimated five hundred million people who are chronically infected with hepatitis B virus (HBV) or Hepatitis C virus (HCV). Reliable data is required by policy making health officials on regular basis in planning and improving of relevant interventions. However, a common challenge health care-planners face often is lack of data and if data is available at all they are imperfect. A review of the national health facilities viral hepatitis data in Ghana indicated that 60% of viral hepatitis data are untyped. Currently the extent of the prevalence of Hepatitis B among sections of the Ghanaian population like pregnant women and the level of endemicity of Hepatitis B is unknown in Ghana.

Therefore this study aimed to determine the prevalence of Hepatitis B Surface Antigen (HBsAg), Hepatitis B Core Antibody (HBcAb) and Hepatitis B envelope-Antigen (HbeAg) among pregnant women in Ghana. And determine the prevalence of Hepatitis B in rural and urban HSS sites and to estimate the endemicity of hepatitis B in Ghana.

Method: A laboratory base cross sectional study involving the use of archived samples from the 2010 HSS was conducted in Ghana. Archived samples from the 2010 HSS were screened for HBV markers. Univeriate analysis was expressed as frequencies and percentages. Results were presented using appropriate chart and tables.

Results: In this study the overall prevalence HBsAg among pregnant women was estimated to be 14.33%. Prevalence of HBcAb was 75.3%; prevalence of HBeAg was 1.23%. HBsAg prevalence rate was highest in the northern sector {14.80% (95% CI 0.13, 0.17)} and
followed by the southern sector \(14.70\% (95\% \text{ CI 0.13, 0.17})\). HBsAg prevalence rate in the middle sector was \(13.20\% (95\% \text{ CI 0.11, 0.15})\), the lowest HBsAg prevalence rate among the sectors. However, there was no significant difference in the hepatitis B prevalence in the sectors \((p= 0.518)\). There was also no significant difference between the prevalence of hepatitis B virus infection among pregnant women in rural and urban sites \((p= 0.374)\). Hepatitis B prevalence rate was \(15.35\%\) among age group 25-29; this was the highest prevalence rate among all the age groups. There was no significant difference in hepatitis B prevalence among the age groups \((p=0.279)\). The overall HBeAg prevalence rate was \(1.23\%\).

**Conclusion:** The HBsAg and HBcAb prevalence rate among pregnant women in this study. Because hepatitis B prevalence does not differ between pregnant women and the general adult population this shows that a higher proportion of population of Ghana has ever been exposed to hepatitis B virus obtained in this study shows Ghana is a highly endemic country for hepatitis B.. Further, the northern sector recorded the highest prevalence of all Hepatitis B markers tested. Additionally, this study determined that there is no significant difference between hepatitis B viral infection between rural and urban sites. On the other hand, HBeAg prevalence is low, indicating a lower contribution of vertical transmission of HBV in Ghana.

**Recommendations:** Immunization of children against HBV infection should be scaled up to capture children who are not delivered in the hospital and those who do not get postnatal care. Current hepatitis B testing strategies should be changed to include other Hepatitis B virus markers in hospital blood bank units.

**Key words:** Ghana, Hepatitis B, HBsAg, HBcAb, HBeAg,
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LIST OF ABBREVIATIONS

ANC  Antenatal Clinic
HBsAg  Hepatitis B Surface Antigen
HBcAb  Hepatitis B Core Antibody
HBeAg  Hepatitis B Envelope Antigen
HSS  HIV Sentinel Survey
OD  Optical Density
WHO  World Health Organization
CHAPTER ONE

INTRODUCTION

1.1 Background
Hepatitis is a disease defined by the inflammation of the liver and it is characterized by the existence of inflammatory cells in the tissues of the liver leading to fibrosis or cirrhosis (Chang, 2007). Hepatitis can be caused by toxin, autoimmune disease, however, most cases of hepatitis worldwide is caused by a group of viruses known as hepatitis viruses. Viral hepatitis is mostly caused by five viruses called hepatitis A, B, C, D, and E. However, hepatitis B and C viruses are of most major concern because of their insidiousness at the early stage of infection and the eventual detection of the disease at a very late stage (World Health Organization, 2016).

Hepatitis B virus (HBV) is transmitted by exposure to body fluids through a myriad of ways. Potentially, HBV is transmitted longitudinally from mother to child during child birth or between family members within households by contact of non-intact skin or mucous membrane with secreting or saliva containing HBV. Additionally, HBV can be transmitted through unsafe sexual intercourse; transfusion of HBV infected blood and blood products; contaminated medical equipment and sharing of contaminated syringes and needles among injecting drug users (Petersen et al., 1976). Moreover, health workers are at high risk of HBV infection when they accidentally sustain needle stick injuries when treating HBV infected patients (World Health Organization, 2016).
Viral hepatitis has become a global public health threat affecting millions of people yearly, causing disability and mortality. There are an estimated five hundred million people who are chronically infected with viral hepatitis B virus (HBV) or Hepatitis C virus (HCV). Additionally, there are approximately one million deaths each year from cases related to viral hepatitis (World Health Organization. Secretariat, 2009). Furthermore, a projected 57% of cases of liver cirrhosis and 78% of cases of primary liver cancer result from HBV and HCV infection (Perz et al., 2006).

More than 2 billion people in the world are estimated to be infected with HBV; 240 million of these people are chronically infected and at risk of death and severe illness (Perz et al., 2006). Annually, mortality from illness (e.g. cirrhosis and hepatocellular carcinoma) due to HBV infection is estimated to be between 1 to 2 million worldwide (Zuckerman and Zuckerman, 2000).

In Ghana there has never been any nationwide population-based survey on Hepatitis B. Estimates from population-survey are more accurate reflection of population parameters. However, nationwide population base surveys are expensive, arduous and time consuming which makes them to be undertaken less often.

In Ghana, there is a sentinel surveillance system known as HIV Sentinel Survey. The HSS is used to monitor HIV and Syphilis prevalence rates all over the country. Data is collected from pregnant women during their first antenatal visit and this serves as a proxy indicator of HIV and Syphilis prevalence among adults in the general population. HIV and Syphilis prevalence are calculated as simple mean of HSS site specific prevalence within each region.
1.2 Problem Statement
Dongdem, et al. (2012) estimated the prevalence of hepatitis B among voluntary blood donors at the Tamale teaching hospital to be 10.79%. Additionally, the same study observed the prevalence of hepatitis B among replacement blood donors as 11.59%. Furthermore, Nkrumah and colleagues estimated hepatitis B prevalence among blood donors at 5.63% (Nkrumah et al, 2011).

Consistently and specifically epidemiological studies have shown an association between chronic HBV infection and hepatocellular carcinoma and other primary liver cancers (World Health Organization, 2012, Hou et al., 2005). Globally, 240 million people are chronically infected and are at risk of death from cirrhosis and HCC. There are an estimated 700,000 deaths annually as a result of HBV infection.

Liver cancer and cirrhosis of the liver have been among the leading cause of death at autopsy in Ghana over the past 4 decades (Edington, 1957, Wiredu and Armah, 2006).

Reliable data is required by policy making health officials on regular basis in planning and improving of relevant interventions as to how to design programmes to arrest the disease at an early stage of its detection. Population base interventions such as surveillance screening and individual/ community focused interventions such as counseling, consultation require good data. Additionally, policy areas such as legislation and guidelines will be better made based on evidence. However, a common challenge health care-planners face often is lack of data and if data is available they are incomplete.

WHO acknowledged that a kernel challenge to the fight against the viral hepatitis epidemic is the lack of adequate data needed to make evidence-based policy decisions (World Health Organization, 2012). Consequently, WHO has made the promotion of gathering of accurate data one of its areas in helping to address the problem.
Also, there are problems with data from earlier studies. This is because laboratory test for HBV markers were done with low sensitivity and specificity test kits which may have under estimate the HBV marker prevalence.

Although, Ghana has an integrated disease surveillance system, the system has been criticized for being untimely and poor in quality of output data. A review of the national health facilities viral hepatitis data in Ghana by Kye-Duodu(2011) indicated that 60% of viral hepatitis data are un-typed. This clearly shows a huge shortcoming in the national data on viral hepatitis and it will be virtually impossible to make a major policy decision using this data because the various types of viral hepatitis will require specific strategies in order to curb the hepatitis B problem in Ghana. Available estimates on hepatitis B prevalence are regional HBV estimates (Hou et al., 2005, World Health Organization, Goldstein et al., 2005b). Currently the extent of the prevalence of Hepatitis B among sections of the Ghanaian population like pregnant women and the level of endemicity of Hepatitis B is unknown in Ghana.
1.2.1 Conceptual Framework

Blood Transfusion

Social and Cultural practices
- Circumcision
- Female genital mutilation

Level of Endemicity/Prevalence of Hepatitis B

HB Surveillance
- Screening

Case Management
- Treatment
- Genotype of HBV
1.3 Justification of study
Today, the world is experiencing a silent epidemic due to viral hepatitis (World Health Organization, 2012). Furthermore, millions of people are living with viral hepatitis and millions more are at risk. Viral hepatitis, also, places a heavy burden on health care system because of the costs of treatment of liver failure and chronic liver disease.

Reliable data is required by health care-planners on regular basis in planning and improving relevant interventions. Data on viral hepatitis in Ghana is inadequate (Kye-Duodu, 2011). Also, obtaining country specific data on hepatitis B, which is estimated to have the highest global diseases burden (2 billion) and the highest mortality rate (five to seven hundred thousand deaths per year) among all the hepatotronic viruses will be more convincing to policy-making health officials.

Estimates obtained from population-based surveys are usually considered as more precise reflection of population parameters because they rely on probability sampling methods. However, they are expensive, tedious and they are sometimes confronted with major ethical challenges.

Therefore, information from this study which will provide HBV prevalence, the differences in HBV prevalence between rural and urban populations among pregnant women in Ghana, which will aid policy makers in making evidence base policies about hepatitis B in Ghana.
1.4 Study Hypothesis

Null Hypothesis:

There is no significant difference in Hepatitis B (HBsAg) prevalence rate among pregnant women in the various sectors (Northern, Middle, Southern) of Ghana.

Alternative Hypothesis:

Hepatitis B (HBsAg) prevalence rate among pregnant women is significantly different in the various sectors (Northern, Middle, Southern) of Ghana.

1.5 General Objective

To determine the prevalence of hepatitis B among pregnant women in Ghana.

1.5.1 Specific Objectives

The following are the specific objectives guiding the study:

1. To determine the prevalence of HBsAg and HBcAb among pregnant women in Ghana.

2. To determine the prevalence HBeAg among pregnant women in Ghana.

3. To determine HBsAg prevalence among pregnant women between the Northern, Middle and Southern sectors of Ghana.
CHAPTER TWO

LITERATURE REVIEW

Viral hepatitis is an inflammation of the liver caused by a group of viruses. These viruses—though unrelated are generically called viral hepatotropic virus. The most common include hepatitis A, hepatitis B, hepatitis C, hepatitis D and hepatitis E viruses. However, there are other viruses such as the yellow fever virus, Epstien-Barr, Herpes simplex, Cytomegalovirus virus that can also cause hepatitis. An estimated five hundred thousand people are chronically infected with HBV or HCV. Additionally, 70000 deaths and 3500 stillbirths are attributed to hepatitis E. Globally, there is an estimated 1,400,000 new hepatitis A infections every year. Moreover, 2.7% of total mortality recorded yearly in the world is viral hepatitis B related (Perz et al., 2006, World Health Organization. Secretariat, 2009).

2.1 Epidemiology

After several recorded outbreaks of serum hepatitis in epidemiological studies the hepatitis surface antigen (HBsAg) originally known as Australian antigen was first discovered in 1965 in the blood of indigenous Australians (Alter and blumberg, 1966). Further work by Dane et al, (1970) resulted in the discovery of the HBV viral particle (1970). Additionally, in the early 1980s came the sequencing of the HBV genome and the first testing of the hepatitis B vaccine.

There are about 350 million chronic carriers of HBV out of an estimated 2 billion people with serological evidence of past or present HBV infection. It has been reported that between 14-15% of HBV infected patients will progress to develop cirrhosis, liver failure or hepatocellular
carcinoma HCC (Lok, 2002). The annual HBV related mortality is between 500,000 to 1.2 million (Lee, 1997; Mahoney, 1999; World Health Organization, 2012). The percentage of people infected with HBV varies greatly worldwide. The age of infection is also associated with endemicity of HBV infection (Hou et al., 2005). There is also a correlation between the primary mode of transmission and the prevalence of HBV infection in an area (Redd et al., 2007). The prevalence of chronic HBV infection worldwide is therefore categorized into high, intermediate and low endemicity. Additionally, there are an estimated 160 million people living with chronic HBV and 360,000 HBV related deaths recorded annually in the Western Pacific region (Emiroglu, October 2010, Goldstein et al., 2005a, Nelson, June 2002). Most infections in the Western Pacific region occur at infancy and childhood. This makes rates of chronic liver disease and liver cancer in adults very high since infections in children are asymptomatic and there is little evidence of acute disease related to HBV (Alter, 2003). On the other hand, Eastern and Southern Europe, the Middle East, Japan, and part of South America are classified under regions with intermediate endemicity. Evidence of HBV infections ranges between 10-16% in these populations and 2-7% are chronic carriers (Hou et al., 2005). Because infections occur in adults and adolescents acute infections are common in intermediate endemic regions. Chronic HBV infection is also sustained in the population because of infections occurring in children and infants (Toukan, 1990). HBV infection in low endemic areas such as North America, Northern and Western Europe and Australia ranges between 5-7% of the population and chronic HBV carriers 0.5-2% of the population (McQuillan et al., 1989). Contrary to high endemic areas, low endemic regions have more acute HBV infections than chronic carriers. This is due to the fact that HBV infections occur in young adults and adolescents of well define risk group (intravenous drug users, homosexual male, health workers etc) (Hou et al., 2005). In highly endemic regions - usually developing regions- such as South East Asia, China, sub-Saharan Africa and the Amazon
Basin - at least 8% of the population are believed to be chronic HBV carriers and 70-90% of
the population in these areas also show past serological evidence of HBV infection (Hou et
al., 2005). The prevalence of HBV infection even though not accurately known in Africa is
believed to be one of the highest in the world: 8% in West Africa; 5-7% in Central, Eastern
and Southern Africa. Furthermore, there are an estimated 100 million people living with
chronic HBV infection in the South-East Asia region (World Health Organization, 2012).
Consequently, 65% of those with HBV infection are not aware of their status (Emiroglu,
October 2010) In Ghana estimated prevalence of hepatitis among voluntary blood donors was
shown to be 10.79% (Dongdem et al., 2012).

2.2 Mode of Transmission
Transmission of HBV occurs through exposure to infectious body fluids such as blood,
 saliva, semen and other body fluids. Reliable evidence shows three major modes of
transmission. They are perinatal, sexual and parenteral/percutaneous (Hou et al., 2005).
Accidental needle stick injuries are also a major consent for health workers when caring for
HBV infected people.

2.3 HBV Morphology
Table 1 Classification of Hepatitis B virus

<table>
<thead>
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<th>Classification</th>
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<tbody>
<tr>
<td>Family</td>
<td>Hepadnaviridae</td>
</tr>
<tr>
<td>Genus</td>
<td>Orthohepadnavirus</td>
</tr>
<tr>
<td>Species</td>
<td>Hepatitis B virus</td>
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</tbody>
</table>

Source: International Committee on Taxonomy of Viruses (2011)

The morphological type of HBV is icosahedral. The complete viral particle consisting of an
outer lipid envelope with an icosahedral nuclocapsid core composed of protein. The complete
hepatitis B viral particle (viron) is 42nM in diameter (Harrison, 2009); a viral DNA and DNA polymerase enclosed in a nucleocasid (Locarnini, 2004); an outer membrane embedded with protein that is involved in viral binding and entry into susceptible cells.

**Figure 1 HBV Morphology**

![HBV Morphology Diagram](2014)

### 2.4 HBV Markers

HBV markers are a distinct HBV-specific antigens and host antibodies that react to these antibodies. They are:

- **Hepatitis B Surface Antigen (HBsAg)**
- **Hepatitis B Core Antibody (HBeAb)**
- **Hepatitis B Envelope Antigen (HBeAg)**
- **Hepatitis B Surface Antibody**
- **Hepatitis B Envelope Antibody**
Hepatitis B Surface Antigen (HBsAg)

It is the first marker to appear after HBV infection. It is detectable in the host serum from 1 to 2 weeks after infection (Hoofnagle, 1981). Therefore it is the first marker test for during the diagnosis of HBV infection. However, HBsAg is undetectable in early and later stages of HBV infection as its being cleared from body. Chronic HBV infection is the presence of HBsAg in the blood for longer than six months or the absence of HBcAb in the blood. HBsAg and total anti-HBc indicates HBV infection

Hepatitis B Core Antibody (HBcAb)

It is an antibody produced by the immune system against the core of hepatitis B Virus. It is always positive in an infected individual and stays for life, regardless of whether the infection is cleared or not.(Mahoney, 1999)

Hepatitis B Envelope Antigen (HBeAg)

It is an indication of viral replicating activity. It is a marker of infectivity(Hoofnagle, 1981).

2.5 Pathogenesis

Even though a receptor required for HBV to enter into host cell is unknown it is believed to be identical to the duck carboxypeptidase D (DCPD) receptor required by duck hepatitis B virus (DHBV). DCPD has been identified for promoting virus binding and expression associated with internalization of virus particles (Glebe and Urban, 2007, Tong et al., 1999). Replication of HBV occurs in hepatocytes which leads to malfunction of the liver. First, HBV binds to the host cell by the preS domain of the viral surface antigen and afterwards it is internalized by endocytosis. Endocytosis is made possible by preS domain and IgA receptors. When host immune response (adaptive immune response) is activated virus-specific cytotoxic T lymphocytes (CTLs) eliminate HBV by killing infected cells and producing antiviral cytokines. Antiviral cytokines are used as signal to cleanse HBV from viable
hepatocytes (Iannacone et al., 2007). Injury to the liver is further worsen by antigen nonspecific inflammatory cells (Iannacone et al., 2005).

2.6 Diagnosis
Blood is the specimen of choice for laboratory diagnosis of HBV infection. The main diagnostic techniques employed in the laboratory diagnosis of HBV infection are serological/immunological and nucleic-acid base test. Serological techniques employ the use of an antigen to detect antibodies or an antibody to detect an antigen of a pathogen in a specimen. Examples of serological techniques used are Immunochromatography, enzyme immune assays, counter current immunoelectrophoresis and agar gel diffusion. Enzyme linked immunosorbent assay (ELISA) a subtype of heterogeneous, solid phase enzyme immune assay is a very sensitive technique capable of detecting HBsAg at concentrations of >0.1 ng/mL (Mahoney, 1999). Nucliec acid techniques on the other hand are based on the detection of HBV DNA in a specimen. HBV can cause both acute and chronic infection; this has important management with regards to treatment, vaccination and counseling. This therefore requires an arduous, yet, an important process of determining the correct HBV infection status of a person.

Chronic HBV infection is the presence of HBsAg in the blood for longer than six months or the absence of HBcAb in the blood. HBsAg is the first detectable viral antigen present during infection. Therefore it is the first marker test during the diagnosis of HBV infection. However, HBsAg is undetectable in early and later stages of HBV infection as its being cleared from body. Consequently, anti-HBc IgM is the only serological evidence of HBV infection in the window period. The presence of HBsAg, IgM anti-HBc indicates an early infection; IgM anti-HBc, total anti-HBc and/without anti-HBs indicates a recent infection; HBsAg and total anti-HBc indicates HBV infection; anti-HBs only indicates response to
hepatitis B vaccine; total anti-HBc and ant-HBs indicates past HBV infection (Mahoney, 1999).

2.7 Prevention
Immunity from both chronic and acute HBV infection can be passive or active. Passively acquired anti-HBs can protect individuals shortly after exposure leading to the development of specific Ig containing high titer of anti-HBs (HBIG). HBIG when used in combination with hepatitis B vaccine is efficacious after perinatal exposure for infants born to HBsAg-positive mother, percutaneous exposure to HBsAg-positive blood and sexual exposure. Active immunization can be attained by the use of commercially safe, immunogenic hepatitis B vaccine. Hepatitis B vaccines are composed of highly purified preparation of HBsAg. Efficacy of hepatitis B vaccines in neonates has been shown to be between 91.4-98.0% in many studies. For long-term protection from hepatitis B vaccines several studies have shown there is a rapid decline in protective antibody in the first 12 months after the first dose and a more decline over time (Mahoney, 1999).
2.8 The HIV Sentinel Survey (HSS)
The HIV Sentinel Survey is a cross sectional survey targeting pregnant women attending clinics at selected ANC sites in Ghana. The HSS is done annually and was initiated based on the premise that prevalence of HIV among pregnant women is a good proxy indicator of the spread of the infection among the populace. HSS data is used as a primary data source for the National HIV and AIDS estimate in Ghana. The HSS report thus represents prevalence among pregnant women. There are 40 sentinel sites in the HSS system in all the ten regions with each region having at least three sites. There are 23 urban sites and 17 rural sites. Base on the size of the population a site is classified as a rural or urban community. Urban sites are settlements with a population of 5000 or more, while sites with population less than 5000 are designated as rural sites.

Target population includes all clients of ANC aged 15 to 49 years attending antenatal clinic for the first time during their current pregnancy within the survey period. However, antenatal clients reporting for repeated visits during the period are excluded(National AIDS/STI Control Programme, 2010).
Figure 2 Map showing Location of HIV Sentinel Sites

Source: 2010 HSS Report

HSS Sample Size
The expected sample size for an ANC site is 500 with an acceptable minimum of 251. The expected sample sizes are based on the estimated HIV prevalence in the population, a confidence interval of 95%, a 5% acceptable margin of error and the need for a sample size large enough to enable analyses by age groups (National AIDS/STI Control Programme, 2010).
HSS Sampling Scheme

Eligible clients are sampled consecutively until the required sample size is obtained or sample period is over (National AIDS/STI Control Programme, 2010).
CHAPTER THREE

METHODS AND MATERIALS

3.1 Study Type

The study was a laboratory base cross sectional study involving the use of archived samples of pregnant women in HSS.

3.2 Study Area

The study area was Ghana. Ghana is located along the Gulf of Guinea and the Atlantic Ocean in the western part of Africa. It is bordered by the Ivory Coast in the west, Burkina Faso in the North, Togo in the east and the Gulf of Guinea in the south. Ghana has a land mass of 238,535 km². Ghana is located in latitude 8° 00′ above the Equator and 2° 00′ west of the Greenwich Meridian. Ghana is divided into 10 administrative divisions: They are the Brong Ahafo, Ashanti, Volta, Northern, Eastern, Greater Accra, Western, Central, Upper East and Upper West region. Ghana had a multi ethnic population of 24 million in 2010 census; 48.7% (9,877,036) were males and 51.3% (10,112,121) were females. 53% of Ghanaians live in urban areas. Accra the political and commercial capital of Ghana is also the administrative capital of GAR. Accra (5°33′00″N 0°12′00″W), occupies a land area of approximately one hundred and seventy-three square kilometers (173 km²). GAR is divided into 10 administrative districts: 2 metropolis and 8 municipalities, Accra and Tema Metropolis, Adentan, Ashaiman, Ga West, Ga East, Ga South Dangbe East, Dangbe West and Ledzokuku. Ghana saw a decline in fertility rate from 3.99 in the year 2000 to 3.28 in the year 2010.
Malaria continues to be the leading cause of death among under five year olds. Adult mortality rate of 263 per 1000 population has been observed in the male population and a 227 mortality rate per 1000 population for women.

Figure 3 Map of Ghana Showing 10 Administrative Regions

3.3 Study Site
The study sites were HIV sentinel sites in the 2010 HIV Sentinel Survey. They are: Northern Sector Bawku (Urban), Builsa(Rural); Middle- Amansie West(Rural), Obuasi(Urban); Southern- Korle Bu(Urban), Fanteakwa(Rural). Upper West and East constituted the northern sector, while the middle sector included Brong Ahafo, Eastern and Ashanti and the Western, Eastern, central and Greater Accra and Volta regions concludes the southern sector as indicated in the figure above.
Builsa district is located in the upper west region of Ghana. Capital of the district is Sandema. Major occupation in the district is agriculture. It has a population of 92,991 (45,892 are male, 47,099 are female) according to the 2010 Ghana population and housing census.

Bawku municipal is located in the upper west region of Ghana. Capital of the district is Bawku. Major occupation in the district is agriculture. It has a population of 217,791 (104,382 are male, 113,409 are female) according to the 2010 Ghana population and housing census.

Amansei West district is located in the southwestern part Ashanti region of Ghana. Capital of the district is Manso Nkwanta. Major occupation in the district is agriculture. It has a population of 134,331 (67,485 are male, 66,846 are female) according to the 2010 Ghana population and housing census.
The Korle Bu sentinel site is located in Accra Metropolitan Area. Accra Metropolitan Area is located in the Greater Accra region of Ghana. Accra is the Administrative capital of the metropolitan area. It has a population of 217,791 (104,382 are male, 113,409 are female) according to the 2010 Ghana population and housing census.

Obuasi municipal is located in the Ashanti region of Ghana. Capital of the district is Obuasi. Major occupation in the district is agriculture and mining. It has a population of 168,641 (81,015 are male, 87,626 are female) according to the 2010 Ghana population and housing census.

Fanteakwa district is located in the Eastern region of Ghana. Capital of the district is Fanteakwa. Major occupation in the district is agriculture. It has a population of 134,331 (67,485 are male, 66,846 are female) according to the 2010 Ghana population and housing census.

The study was carried out at the National Public Health and Reference Laboratory (NPHRL). The NPHRL is the national and reference laboratory under the Ghana Health Service which serve to among other things screen and confirm HBV infection.
### 3.4 Variables

<table>
<thead>
<tr>
<th>VARIABLE</th>
<th>OPERATIONAL DEFINITION</th>
<th>INDICATOR</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>Age of respondent in completed years</td>
<td>Age as recorded in the record book</td>
</tr>
<tr>
<td>Geographical Location</td>
<td>The location (region) where sample was taken</td>
<td>The location as recorded in the record book</td>
</tr>
<tr>
<td>HBsAg status of the specimen</td>
<td>Presence or absence of HBsAg in serum of specimen</td>
<td>Optical density of test sample as indicated by photospectrometre reading</td>
</tr>
<tr>
<td>HBeAb status of the specimen</td>
<td>Presence or absence of HBeAb in serum of specimen</td>
<td>Optical density of test sample as indicated by photospectrometre reading</td>
</tr>
<tr>
<td>HBeAg status of the specimen</td>
<td>Presence or absence of HBeAg in serum of specimen</td>
<td>Optical density of test sample as indicated by photospectrometre reading</td>
</tr>
</tbody>
</table>

### 3.5 Study Population

The study population was pregnant women who were involved in the 2010 HIV Sentinel Survey (HSS) in Ghana.

### 3.6 Sample Size and Sampling Method

The HSS consist of 40 sentinel sites distributed all over Ghana. Therefore, the country was divided into three sectors: namely northern, middle and southern sectors. The northern sector consists of the Upper East Region, Upper West Region and the Northern Region. Further, the middle sector includes Ashanti Region and the Brong Ahafo Region. The southern sector includes Western, Central, Greater Accra and the Volta regions.

Stratified sampling was done to ensure one rural and one urban site for each sector is selected. This was done by grouping sentinel sites under rural and urban for each sector by using the HSS protocol. However, actual selection of sentinel sites for each site was done using lottery sampling method. Names of sentinel sites were written on pieces of paper and
folded. They were placed in a bowl and two sentinel sites were selected one by one for each sector.

However, sample size large enough to accurately estimate HBsAg and HBeAg prevalence and to enable analyses by age groups was of 500 for each selected site as prescribed in the WHO protocol (EPI and World Health Organization). Consequently, 3,000 samples were screened.

3.7 Serologic Assay

Archived serum samples collected for HSS were tested to detect HBsAg and anti-HBc with HBsAg and HBsAb ELISA test kit. Subsequently, all HBsAg positive samples were tested for HBeAg with HBeAg ELISA test kit.

3.8 Laboratory Techniques

3.8.1 Hepatitis B Surface Antigen (HBsAg), Hepatitis B Core Antibody (HBcAb) and Hepatitis B Envelope Antigen (HBeAg) ELISA test

Archived serum samples from 2010 HSS stored at -70°C were allowed to thaw at room temperature. 100 μL of positive control and 100 μL of negative control were dispensed in duplicates into respective wells. One well was set blank as a background control. 100μL of the thawed serum was added to each well. The microtiter plates were placed into a humidified box, and incubated at 37°C for 60 minutes. 50μL of enzyme conjugate was added to each well except the blank well. The microtiter plates were swirled gently for one minute to facilitate mixing. The microtiter plates were placed into a humidified box, and incubated at 37°C for 30 minutes. Each well was washed 5 times after the incubation period. 50μL of substrate A(HRP-substrate) and 50μL of substrate B(TMB) were added to each well respectively.
Mixing was done gently by swirling and the microtiter plates were incubated at 37°C for 30 minutes. 50μL of stop solution was added to each well to stop the color reaction. The optical density OD value for each plate was read at 450nm.

Figure 5HBsAg ELISA Microtiter Plate

![Figure 5HBsAg ELISA Microtiter Plate](image)

Figure 6HBcAb ELISA Microtiter Plate

![Figure 6HBcAb ELISA Microtiter Plate](image)

1 Yellow color change is an indication of a reactive specimen

2 Yellow color change is an indication of a reactive specimen
Figure 7HBeAg ELISA Microtiter Plate

3 Yellow color change is an indication of a reactive specimen
3.9 Quality Control

In house or standard control samples across the analytical range of the assay that is low, medium and high controls as well as negative controls in previous assays was obtained from the National Public Health and Reference Laboratory and included in all specimen assays. Results were considered valid if the control values lie within their expected ranges.

Additionally, ten percent (10%) of the specimen were randomly selected and their laboratory analysis replicated in order to test the validity of the results. All assays were conducted by the researcher with supervision from experienced microbiologist at the NPHRL to address quality control measures such as calibration of pipettes and incubation at correct temperature. (Appendix B)

3.10 Ethical Consideration

The proposal was reviewed and approved by the Ethical Review Board of the Ghana Health Service (Ethical clearance ID No. GHS-ERC 89/03/13).

All data was handled anonymously and confidentially. Only investigators of this study were privy to the data. Research information will not be shared with third parties not directly involved in the research and will be used purely for academic purposes. References to identities that would have compromised anonymity were removed before preparation of research reports. Soft copies were encrypted whilst hard copies were stored under lock and key.
3.11 Data Analysis
Data was double entered and cleaned in EpiData and analysis done with STATA software package version 11.

Univariate analysis was expressed as frequencies and percentages. Results were presented using appropriate chart and tables. Statistical significance were determined at a level of 5%.

Any statistically significance in prevalence between the different study sites, between geographical locations was determined with Chi square test.

3.13 Limitations
Because the research study was based on the use of HSS specimen it was liable to any limitation in the HSS.

This includes:
- Collection, storage and transport of specimen.

Pre-analytical phase is an important phase in any laboratory assay. How samples are collected transported and stored can affect the outcome of a test result. In this study collection, transportation and storage was not done by the investigator. However, before testing for HBV commenced, samples were randomly selected and tested to determine the integrity of the samples by comparing the outcome of the test to that obtained in the HSS.
CHAPTER FOUR

RESULTS

Table 2 shows the total number of ANC clients screened in each age group, prevalence of hepatitis B (HBsAg) among age groups, site type and sentinel sites. Overall, the highest number of samples was obtained from the 25-29 year age group 29.1% (873). Sixty-five percent of the samples screened were below the age of thirty. Furthermore, 15-24 year age group contributed 36.33% of the samples; 10.2% (306) percent was from age group 15-19 years. The least number of samples came from the 45 to 49 age group which accounted from less than one percent 0.53% (16) of the total samples collected. Moreover, this trend was observed in all the individual sites.

Age group 25-29 had the height hepatitis B prevalence rate – 15.35% (95% CI 0.13, 0.18). Second was age group 20-24 with 15.05% (95% CI 0.13, 0.17). Additionally, prevalence rate for age groups 30-34, 35-39, 40-44 was 14.40% (95% CI 0.12, 0.17), 14.14% (95% CI 0.10, 0.19) and 10.34% respectively. The lowest hepatitis B prevalence rate was recorded among age group 45-49{6.25% (95% CI 0.002, 0.30), followed by age group 15-19{10.13% (95% CI 0.07, 0.16). Further, there was no significant difference in hepatitis B prevalence among the age groups (p=0.279) (Table 1).

Prevalence rate for all rural and urban sites put together was 13.67% (95% CI 0.12, 0.16) and 14.80% (95% CI 0.13, 0.17) respectively. Additionally, there was no significant difference in hepatitis B prevalence rate among the site types (p= 0.374).

Among the individual sites Builsa had the highest hepatitis B prevalence rate of 16.80% (95% CI 0.14, 0.20). Korle Bu sentinel site had the second highest hepatitis B prevalence rate of 16.20% (95% CI 0.13, 0.20). Also, Obuasi, Fanteakwa, Bawku and Agroyesum sites had hepatitis B prevalence rate of 15% 15% (CI 95% 0.12, 0.19), 13% (CI 0.10, 0.17), 12.8%
(CI 0.10, 0.17) and 11% (CI 0.08, 0.14) respectively. There was no significant difference in hepatitis B prevalence rate among the study sites (p = 0.065).

**Table 3 Hepatitis B Prevalence among ANC Clients in HIV Sentinel Sites, Ghana, 2010**

<table>
<thead>
<tr>
<th>Age Group</th>
<th>Hepatitis B Positive</th>
<th>Hepatitis B Negative</th>
<th>Total</th>
<th>Prevalence %</th>
<th>95% CI</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>15-19</td>
<td>275</td>
<td>31</td>
<td>306</td>
<td>10.13</td>
<td>0.07, 0.14</td>
<td>0.279</td>
</tr>
<tr>
<td>20-24</td>
<td>666</td>
<td>118</td>
<td>784</td>
<td>15.05</td>
<td>0.13, 0.17</td>
<td></td>
</tr>
<tr>
<td>25-29</td>
<td>739</td>
<td>134</td>
<td>873</td>
<td>15.35</td>
<td>0.13, 0.18</td>
<td></td>
</tr>
<tr>
<td>30-34</td>
<td>539</td>
<td>91</td>
<td>630</td>
<td>14.4</td>
<td>0.12, 0.17</td>
<td></td>
</tr>
<tr>
<td>35-39</td>
<td>261</td>
<td>43</td>
<td>304</td>
<td>14.14</td>
<td>0.10, 0.19</td>
<td></td>
</tr>
<tr>
<td>40-44</td>
<td>78</td>
<td>9</td>
<td>87</td>
<td>10.34</td>
<td>0.05, 0.19</td>
<td></td>
</tr>
<tr>
<td>45-49</td>
<td>15</td>
<td>1</td>
<td>16</td>
<td>6.25</td>
<td>0.002, 0.30</td>
<td></td>
</tr>
</tbody>
</table>
| Site Type | Rural                | 1295                 | 205   | 1500         | 13.67        | 0.12, 0.16 | 0.374  
|           | Urban                | 1278                 | 222   | 1500         | 14.80        | 0.13, 0.17 |         |  
| Site Name | Agroyesum            | 445                  | 55    | 500          | 11.00        | 0.08, 0.14 | 0.065  
|           | Bawku                | 436                  | 64    | 500          | 12.80        | 0.10, 0.17 |         |  
|           | Builsa               | 416                  | 84    | 500          | 16.80        | 0.14, 0.20 |         |  
|           | Fanteakwa            | 434                  | 66    | 500          | 13.20        | 0.10, 0.17 |         |  
|           | Korle Bu             | 419                  | 81    | 500          | 16.20        | 0.13, 0.20 |         |  
|           | Obuasi               | 423                  | 77    | 500          | 15.40        | 0.12, 0.19 |         |  

\(^a\) \(^b\) Hepatitis B positive\(^a\)/Hepatitis B Positive\(^a\) + Hepatitis B Negative\(^b\) * 100.

\(^5\) \(^d\) Chi square test for difference between the groups of Age, Site type and Site respectively

Abbreviations: ANC, Antenatal Clinic; CI, Confidence Interval
Table 3 shows HBsAg prevalence rate in the sectors and age groups. In the northern sector HBsAg prevalence rate was 16.80% (95%, CI 0.14, 0.20) in the rural sites which is higher than that of the urban sites (12.80% (95%, CI 0.10, 0.16). In the northern sector there was no significant difference between the HBsAg prevalence rate in the rural and urban sites (p=0.075). On the other hand, in the middle sector, urban sites had the highest HBsAg prevalence rate of 15.40 % (95%, CI 0.12, 0.19), while the rural site had 11% (95%, CI 0.12, 0.19). There was however, a significant difference between the HBsAg prevalence rate in the rural and urban sites (p=0.040).

For the southern sector urban sites had 16.20% (95%, 0.13, and 0.20) prevalence rate. HBsAg prevalence rate was 13.20% (95% CI 0.10, 0.16) in rural sites. However, there was no significant difference in HBsAg prevalence rate among the site type (rural/urban) (p= 0.180).
Table 4 HBsAg Prevalence among Age Groups, Rural/Urban Prevalence Rate in Sectors, Ghana, 2010

<table>
<thead>
<tr>
<th>Age Group</th>
<th>HBsAg Positive(^a)</th>
<th>HBsAg Negative(^b)</th>
<th>Prevalence(^c) (%)</th>
<th>C.I</th>
<th>P Value(^d)</th>
</tr>
</thead>
<tbody>
<tr>
<td>15-19</td>
<td>31</td>
<td>275</td>
<td>10.13</td>
<td>0.07, 0.14</td>
<td>0.279</td>
</tr>
<tr>
<td>20-24</td>
<td>118</td>
<td>666</td>
<td>15.05</td>
<td>0.13, 0.17</td>
<td></td>
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<tr>
<td>25-29</td>
<td>134</td>
<td>739</td>
<td>15.35</td>
<td>0.13, 0.18</td>
<td></td>
</tr>
<tr>
<td>30-34</td>
<td>91</td>
<td>539</td>
<td>14.44</td>
<td>0.12, 0.17</td>
<td></td>
</tr>
<tr>
<td>35-39</td>
<td>43</td>
<td>261</td>
<td>14.14</td>
<td>0.10, 0.19</td>
<td></td>
</tr>
<tr>
<td>40-44</td>
<td>9</td>
<td>78</td>
<td>10.34</td>
<td>0.05, 0.19</td>
<td></td>
</tr>
<tr>
<td>45-49</td>
<td>1</td>
<td>15</td>
<td>6.25</td>
<td>0.002, 0.30</td>
<td></td>
</tr>
<tr>
<td>Sector</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Northern</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rural</td>
<td>84</td>
<td>416</td>
<td>16.80</td>
<td>0.14, 0.20</td>
<td></td>
</tr>
<tr>
<td>Urban</td>
<td>64</td>
<td>436</td>
<td>12.80</td>
<td>0.10, 0.16</td>
<td>0.075(^e)</td>
</tr>
<tr>
<td>Middle</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rural</td>
<td>55</td>
<td>445</td>
<td>11.00</td>
<td>0.08, 0.14</td>
<td></td>
</tr>
<tr>
<td>Urban</td>
<td>77</td>
<td>423</td>
<td>15.40</td>
<td>0.12, 0.19</td>
<td>0.040(^e)</td>
</tr>
<tr>
<td>Southern</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rural</td>
<td>66</td>
<td>434</td>
<td>13.20</td>
<td>0.10, 0.16</td>
<td></td>
</tr>
<tr>
<td>Urban</td>
<td>81</td>
<td>419</td>
<td>16.20</td>
<td>0.13, 0.20</td>
<td>0.180(^e)</td>
</tr>
</tbody>
</table>

\(^a\) (HBsAg Positive\(^a\)/HBsAg Positive\(^a\) + HBsAg Negative\(^b\)) * 100.
\(^d\) Chi square test for difference between the age groups and Sector
\(^e\) Chi square test for difference between the age groups and Sector

Abbreviations: C.I, Confidence Interval; HBsAg, Hepatitis B Surface Antigen
Table 4 shows HBcAb prevalence rate in the sectors and age groups. HBcAb prevalence rate in age group 45-49 was 81.25% (95% CI 0.54, 0.96) which was the highest among the age group. This is followed by age group 40-44 which had HBcAb prevalence rate of 79.31% (95% CI 0.69, 0.87). HBcAb prevalence rate for 35-39, 30-34, 25-29, 20-24 was 79.28% (95% CI 0.74, 0.84), 78.73 (95% CI 0.75, 0.82), 76.29% (95% CI 0.73, 0.79) and 71.30% (95% CI 0.68, 0.74) respectively. Additionally, there was a significant difference in HBcAb prevalence rate among the age groups (p=0.004).

Furthermore, in the northern sector, HBcAb prevalence rate was 94.8% (95% CI 0.92, 0.96) in the rural sites which is higher than that of the urban sites (87% (95% CI 0.84, 0.0900). In the northern sector there was a significant difference between the HBcAb prevalence rate in the rural and urban sites (p<0.0001).

HBcAb prevalence rate in the middle sector was 79.2% (95% CI 0.75, 0.83) for the rural sites. HBcAb prevalence rate of HBcAb was 68.6% (95% CI 0.64, 0.73) in urban sites. There was a significant difference between the HBcAb prevalence rate in the rural and urban sites (p<0.0001).

Southern sector HBcAb prevalence rate in rural and urban sites was 63.6% (95% CI 0.59, 0.68) and 58.6% respectively. Consequently, there was no significant difference in HBcAb prevalence rate among the rural and urban sites (p=0.105).
Table 5 Prevalence of HBcAb among Age Groups, Rural/Urban Prevalence rate in Sectors, Ghana, 2010

<table>
<thead>
<tr>
<th>HBcAb</th>
<th>Positive&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Negative&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Prevalence&lt;sup&gt;b&lt;/sup&gt; %</th>
<th>C.I</th>
<th>P Value&lt;sup&gt;d, e&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age Group</td>
<td>15-19</td>
<td>215</td>
<td>91</td>
<td>70.26</td>
<td>0.65, 0.75</td>
</tr>
<tr>
<td></td>
<td>20-24</td>
<td>559</td>
<td>225</td>
<td>71.30</td>
<td>0.68, 0.74</td>
</tr>
<tr>
<td></td>
<td>25-29</td>
<td>666</td>
<td>207</td>
<td>76.29</td>
<td>0.73, 0.79</td>
</tr>
<tr>
<td></td>
<td>30-34</td>
<td>496</td>
<td>134</td>
<td>78.73</td>
<td>0.75, 0.82</td>
</tr>
<tr>
<td></td>
<td>35-39</td>
<td>241</td>
<td>63</td>
<td>79.28</td>
<td>0.74, 0.84</td>
</tr>
<tr>
<td></td>
<td>40-44</td>
<td>69</td>
<td>18</td>
<td>79.31</td>
<td>0.69, 0.87</td>
</tr>
<tr>
<td></td>
<td>45-49</td>
<td>13</td>
<td>3</td>
<td>81.25</td>
<td>0.54, 0.96</td>
</tr>
<tr>
<td>Sector</td>
<td>Northern Rural</td>
<td>474</td>
<td>26</td>
<td>94.8</td>
<td>0.92, 0.96</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Urban</td>
<td>435</td>
<td>65</td>
<td>87</td>
</tr>
<tr>
<td></td>
<td>Middle Rural</td>
<td>396</td>
<td>104</td>
<td>79.2</td>
<td>0.75, 0.83</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Urban</td>
<td>343</td>
<td>157</td>
<td>68.6</td>
</tr>
<tr>
<td></td>
<td>Southern Rural</td>
<td>318</td>
<td>182</td>
<td>63.6</td>
<td>0.59, 0.68</td>
</tr>
<tr>
<td></td>
<td>Urban</td>
<td>293</td>
<td>207</td>
<td>58.6</td>
<td>0.54, 0.63</td>
</tr>
</tbody>
</table>

<sup>a</sup> HBcAb Positive<sup>a</sup>/HBcAb Positive<sup>a</sup> + HBcAb Negative<sup>b</sup> * 100.
<sup>d, e</sup> Chi square test for difference between the age groups and Sector
Abbreviations: C.I, Confidence Interval; HBcAb, Hepatitis B Core antibody
Table 5 shows HBeAg prevalence rate in the sectors and age groups. HBeAg prevalence rate in age group 40-44 was 11.11% (95% CI 0.003, 0.50) which was the highest among the age group. This is followed by age group 25-29 which had HBeAg prevalence rate of 12.5% (95% CI 0.08, 0.19). HBeAg prevalence rate for 20-24, 30-34, 15-19, 35-39, 45-49 was 8.6% (95% CI 0.04, 0.15), 7.69% (95% CI 0.03, 0.15), 3.32% (95% CI 0.001, 0.17), 2.33% (95% CI 0.001, 0.12) respectively. Additionally, there was a significant difference in HBeAg prevalence rate among the age groups (p=0.408).

Furthermore, in the northern sector, HBeAg prevalence rate was 10.71% (95% CI 0.05, 0.19) in the rural sites which is higher than that of the urban sites (9.38% (95% CI 0.04, 0.20)).

In the northern sector there was no significant difference between the HBeAg prevalence rate in the rural and urban sites (p=0.789).

HBeAg prevalence rate in the middle sector was 5.45% (95% CI 0.01, 0.15) for the rural sites. HBeAg prevalence rate of HBeAg was 9.09% (95% CI 0.04, 0.18) in urban sites.

There was however, no significant difference between the HBeAg prevalence rate in the rural and urban sites (p=0.436).

The southern sector HBeAg prevalence rate in rural and urban sites was 10.61% (95% CI 0.04, 0.21) and 6.17% (95% CI 0.02, 0.14) respectively. Consequently, there was no significant difference in HBcAb prevalence rate among the rural and urban sites (p=0.329).
Table 6 Prevalence of HBcAb among Age Group, Rural/Urban Prevalence Rate IN Sectors, Ghana, 2010

<table>
<thead>
<tr>
<th>Age Group</th>
<th>HBeAg Positive</th>
<th>Negative</th>
<th>Prevalence (%)</th>
<th>C.I</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>15-19</td>
<td>1</td>
<td>30</td>
<td>3.23</td>
<td>0.001, 0.17</td>
<td>0.408</td>
</tr>
<tr>
<td>20-24</td>
<td>10</td>
<td>106</td>
<td>8.62</td>
<td>0.04, 0.15</td>
<td></td>
</tr>
<tr>
<td>25-29</td>
<td>17</td>
<td>119</td>
<td>12.5</td>
<td>0.08, 0.19</td>
<td></td>
</tr>
<tr>
<td>30-34</td>
<td>7</td>
<td>84</td>
<td>7.69</td>
<td>0.03, 0.15</td>
<td></td>
</tr>
<tr>
<td>35-39</td>
<td>1</td>
<td>42</td>
<td>2.33</td>
<td>0.001, 0.12</td>
<td></td>
</tr>
<tr>
<td>40-44</td>
<td>1</td>
<td>8</td>
<td>11.11</td>
<td>0.003, 0.50</td>
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<tr>
<td>45-49</td>
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<table>
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<tr>
<th>Sector</th>
<th>Rural</th>
<th>Urban</th>
<th>Prevalence (%)</th>
<th>C.I</th>
<th>P Value</th>
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<tbody>
<tr>
<td>Northern</td>
<td>9</td>
<td>6</td>
<td>10.71</td>
<td>0.05, 0.19</td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>0.789e</td>
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<td>Middle</td>
<td>3</td>
<td>7</td>
<td>5.45</td>
<td>0.01, 0.15</td>
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<td></td>
<td>0.436e</td>
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<tr>
<td>Southern</td>
<td>7</td>
<td>5</td>
<td>10.61</td>
<td>0.04, 0.21</td>
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</tr>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>0.329e</td>
<td></td>
</tr>
</tbody>
</table>

---

10  a HBeAg Positive / HBeAg Positive + HBeAg Negative * 100.
11  d, e Chi square test for difference between the age groups and Sector

Abbreviations: C.I, Confidence Interval; HBeAg, Hepatitis B Envelope antigen
Figure 8 shows prevalence rate of hepatitis B markers in the sectors and the overall HBsAg marker prevalence rate. HBsAg prevalence rate was highest in the northern sector \(14.80\%\) (95% CI 0.13, 0.17) and followed by the southern sector \(14.70\%\) (95% CI 0.13, 0.17). HBsAg prevalence rate in the middle sector was 13.20% (95% CI 0.11, 0.15); the lowest HBsAg prevalence rate among the sectors. Further, the overall HBsAg prevalence rate was 14.23% (95% CI 0.13, 0.16). There was no significant difference in HBsAg prevalence rate in the sectors \(p = 0.518\).

Figure 8
Figure 9 shows prevalence rate of HBcAb marker in the sectors and the overall pHBcAb marker prevalence rate.

HBcAb prevalence rate was highest in the northern sector (90.90% (95% CI 0.90, 0.93)) and followed by the middle sector (73.90% (95% CI 0.71, 0.77)). HBcAb prevalence rate in the southern sector was 61.11% (95% CI 0.11, 0.15); the lowest HBcAb prevalence rate among the sectors. Further, the overall HBcAb prevalence rate was 75.30% (95% CI 0.13, 0.16). There was a significant difference in HBcAb prevalence rate in the sectors.

Figure 9
Figure 10 shows prevalence rate of HBeAg marker in the sectors and the overall HBeAg marker prevalence rate. Additionally, figure 10 shows prevalence rate of HBeAg among HBsAg positive in the study.

HBeAg prevalence rate was highest in the northern sector {1.50% (95% CI 0.008, 0.020)} and followed by the southern sector {1.2% (95% CI 0.006, 0.021)}. HBeAg prevalence rate in the middle sector was 1.0% (95% CI 0.005, 0.018); the lowest HBeAb prevalence rate among the sectors. Further, the overall HBeAg prevalence rate was 1.23% (95% CI 0.008, 0.020) which was higher than the middle and southern sectors. There was no significant difference in HBeAg prevalence rate among the sectors (p = 0.598). Further, HBeAg prevalence rate among HBsAg positives was highest in the northern sector {10.14% (95% CI 0.06, 0.16)} and followed by the southern sector {8.16% (95% CI 0.04, 0.14)}. HBeAg prevalence rate among HBsAg positives in the middle sector was 7.58% (95% CI 0.04, 0.13); the lowest HBeAb prevalence rate among the sectors. The overall HBeAg prevalence rate among HBsAg positives was 8.66% (95% CI 0.062, 0.12). There was no significant difference in HBeAg prevalence rate among the sectors (p = 0.723).
Figure 10

Prevalence of HBeAg Against Sectors In HSS, Ghana, 2010

<table>
<thead>
<tr>
<th>Sector</th>
<th>Prevalence rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Northern</td>
<td>10.14</td>
</tr>
<tr>
<td>Middle</td>
<td>8.16</td>
</tr>
<tr>
<td>Southern</td>
<td>8.66</td>
</tr>
<tr>
<td>Overall</td>
<td>8.66</td>
</tr>
</tbody>
</table>

- Prevalence of HBeAg (p=0.598)
- Prevalence of HBeAg among HBsAg positive (p=0.723)
CHAPTER FIVE
DISCUSSIONS

More than 2 000 million people alive today have been infected with HBV at some time in their lives. Of these, about 350 million remain infected chronically and become carriers of the virus. Every year there are over 4 million acute clinical cases of HBV, and about 25% of carriers, 1 million people a year, die from chronic active hepatitis, cirrhosis or primary liver cancer (World Health Organization 2016). This study assessed the prevalence of hepatitis B among pregnant women in Ghana.

The overall prevalence of hepatitis B viral infection (HBsAg) among pregnant women screened in the study was 14%. Because hepatitis B prevalence does not differ between pregnant women and the general adult population it can be deduced that the endemicity of hepatitis B in Ghana is in the high endemic category according to WHO categorization. This parallels with WHO categorization which categorizes sub-Sahara as high endemic region of the world (World Health Organization, 2012). However, in comparison with Germany, Jilg et al. (2001) estimated HBsAg prevalence in the population at 0.63% thereby putting the endemicity of Hepatitis B of Germany at lower category. This is further corroborated by their estimation of HBcAb in the German population at 8.7%. In addition the status quo is the same in most parts of Europe where countries like the United Kingdom, France, Italy, and Finland have an estimated HBsAg prevalence of less than 1%. This puts the entire continent of western Europe in the lower endemic category (van de Marita, 2006). Further, in the United States an analysis of the national health and nutrition examination survey data estimated hepatitis B prevalence of 0.33%. This also puts the US in a lower endemic category (C.D.C, 2008).

Comparing hepatitis B prevalence in this study to India, India’s level of hepatitis B endemicity is also estimated to be intermediate as determined by a point prevalence of
3.7% (National Centre for Disease Control (NCDC 2014)). Further, a study by Silveira et al. (1999) shows a varying hepatitis B endemicity levels in countries in South America. Silveira et al. (1999) estimated point hepatitis HBcAb prevalence for Dominican Republic to be 21.4%. This makes Dominican Republic a moderately endemic country. However, further estimates puts Argentina, Brazil, and Venezuela at lower endemic countries.

Further, HBV prevalence in the sectors did not vary greatly. HBV prevalence in middle and southern sectors were 14.8, 13.20% and 14.70% respectively. This is in contrast with previous studies by Tandon et al. (1996) in India. Tandon et al. (1996) established that there was difference in HBsAg prevalence in states in India with different socioeconomic status (Chandigarh and Madras). Chandigarh has a higher gross domestic product than Madras; Chandigarh’s HBsAg prevalence was 1% and Madras prevalence was 5.5%. In Ghana the Northern sector is less endowed economically than the middle and the southern sector. However, there was no significant difference in HBsAg prevalence in the various sectors in this study.

Additionally, the overall prevalence rate of HBV infection in rural and urban sites were 13.67% and 14.80% respectively. This difference, however, is insignificant as determined by the test statistic. This finding however disagrees with the work of Mehmet et al. (2005) where they found there was a statistically significant difference between urban and rural regions in terms of HBsAg positivity in southeastern Turkey. In the United State of America urban areas have higher hepatitis B prevalence rate than rural areas (Te and Jensen, 2010). Additionally, contrary to the finding of this study Kew (1996) also found a higher hepatitis B prevalence rate in rural areas than urban areas in South Africa.

Overall prevalence varied from 10.13% in age group 15-19 to 15.35% in age group 25-29. Age group 20-24 (15.05 %) had the second highest prevalence, followed by age group 30-
Moreover, prevalence of HBsAg ebbs among age group 45-49(6.25%). The number of samples tested were fewer in the higher age groups. Also, Ghana integrated the Hepatitis B vaccine into her EPI schedule in 2002 as one of the component of the pentavalent vaccine, many of pregnant women tested in this survey may have not been exposed to hepatitis B vaccine. This may have contributed the differences in prevalence rate among the age groups.

This corresponds favorably with findings of Ott et al., (2012); it was established in their study that prevalence of HBsAg in parts of Turkey – region A(Marmara & Aegean), region B (Black sea, Central Anatolia, Mediterranean), region C (Eastern and South Eastern) – was lower in 0-14 year olds, highest among 15-24 age groups and falls among 35-45 age groups. Also, Kim et al. (2004) and the C.D.C (2006b) found a similar trend. In the study by Kim et al. (2004) they determined that hepatitis B prevalence rate was highest among age group 20-29, HBV prevalence was 25.2%, followed by 21.5% among those of age group 30-39%.

The overall prevalence of HBcAb in the study was 75.3% and because hepatitis B prevalence does not differ between pregnant women and the general adult population this shows that a higher proportion of population of Ghana has ever been exposed to hepatitis B virus. Also, HBcAb prevalence was highest compared to all the other markers tested. This finding is consistent with those of Moghaddam et al (2010) Hamidi et al,(Unknown) indicating a high percentage of HBcAb prevalence in the population they studied. On the other hand, a lower HBcAb prevalence rate of 8.7% was estimated by Jilg et al. (2001) in Germany. Additionally, some of these cases may be due to occult hepatitis B infection. Again, comparing sector by sector it was established in this study that northern sector had a high HBcAb prevalence followed by the middle and southern sector respectively. Rural sites had higher HBcAb prevalence compared to urban sites. On the other hand, HBsAg prevalence was higher in urban sites than rural sites.
The results of the study shows that HBeAg prevalence decreases by age with 25-29 age group recording highest prevalence. In all HBeAg prevalence was significantly lower than the other markers tested in this study. Additionally, there was no significant difference in HBeAg prevalence rate among the sectors and site type.

HBeAg prevalence among HBsAg positive women was low in this study. This corresponds favorably with a study by Kiire (1996). Kiire indicated that HBeAg prevalence among pregnant women in is lower in pregnant women in sub-Saharan Africa than Asia.

HBeAg prevalence (1.23%) in this study was low. Data from the 2008 Ghana demographic and health survey established that 26.2% (Ghana Statistical Service, 2008) of lactating mothers do not attend postnatal clinic. Moreover, there is little less than 50% (W.H.O, 2014) out of institutional delivery and only 36% of newborns receiving hepatitis B vaccine birth dose in high endemic countries(C.D.C. 2006a), Some children are not exposed to the hepatitis B vaccine and this may increase the contribution of vertical transmission of hepatitis B. However, transmission from other routes like childhood horizontal transmission are also important.

HBsAg can be detected in high levels in serum during acute or chronic hepatitis virus infection but disappears between one to two months in acute infection. HBeAg is an indicator of active viral replication, which means an infected person is infectious; HBeAg is detectable between twelve to sixteen weeks of infection. HBcAb appears at the onset of the symptoms hepatitis B infection and persists for life (CDC, 2006). Therefore prevalence of HBcAb in any population will be higher than other hepatitis B markers (HBsAg and HBeAg). This corroborates the findings in this study where HBcAb prevalence was higher in all the sectors than other markers tested. Further, because HBsAg disappears when hepatitis B infection is resolved and HBcAb persists for life after infection; HBsAg prevalence provides a true
prevalence for the various sectors. HBcAb prevalence factors in those with current and past infections in this study. Occult hepatitis B virus infection (OBI) is infection with detectable hepatitis B viral DNA and undetectable HBsAg in patients blood. However, commonly used serological assays are based on detection of HBsAg. Prevalence of OBI is higher in seropositive patients, especially those who are positive for HBcAb. HBcAb positivity is therefore a surrogate to marker for OBI (Raimondo et.al, 2008). Considering the high HBcAb prevalence in this study it will therefore be prudent to add HBcAb testing to hepatitis B screening in blood bank units.
CHAPTER SIX
CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions
The HBsAg and HBcAb prevalence rate among pregnant women in this study was high. Because hepatitis B prevalence does not differ between pregnant women and the general adult population this shows that a higher proportion of population of Ghana has ever been exposed to hepatitis B virus. This study shows Ghana is a highly endemic country for hepatitis B. Further, the northern sector (i.e. Northern, Upper East, and Upper West regions) recorded the highest prevalence of all Hepatitis B markers tested. Additionally, this study determined that there is no significant difference between hepatitis B viral infection between rural and urban sites. Also, there was no significant difference between the sectors. On the other hand, HBeAg prevalence is low, indicating a lower contribution of vertical transmission of HBV in Ghana.

6.2 Recommendations
The following are cited recommendations for some health institutions and related blood bank units in Ghana:

Ghana Health Service
- The program of immunizing of children at birth should continue. However, in a setting where there is a significant level of out of institutional delivery the immunization of children should be scaled up to capture these children.
- Also, special attention and studies should be conducted to determine the contribution of other route of transmission of the virus in Ghana.
Hospital Blood Bank Units

Because of the high prevalence of HBcAb prevalence shown in this study which may be an indicator of window HBV infection or occult HBV infection it will be prudent to change current testing strategies to include other markers of HBV in order to make apt decision on which blood to reject.
REFERENCES


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### APPENDICES

#### APPENDIX A

Table 7 Quality Control Results

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<thead>
<tr>
<th>Test</th>
<th>Total Positive</th>
<th>True Positive</th>
<th>False Positive</th>
<th>Total Negative</th>
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<td>100</td>
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<td>200</td>
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<tr>
<td>HBcAb</td>
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<td>200</td>
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<tr>
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<td>0</td>
<td>32</td>
<td>32</td>
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APPENDIX B

List of Materials
Sodium Hypochloride

Laboratory Coat

Single Channel pipette

Multichannel pipette

Gauze

Incubator

Distilled water

Dispossession pipette tip

Humidified Box

Laboratory coat

Discard jars

Examination gloves

Equipment
Spetrophotometer

Microtiter plate washer

Timer

Fridge (2°C and 8°C)

Freezer (-20°C and -80°C)
Reagents

Bioneovan HBsAg ELISA Kit

Bioneovan HBeAg ELISA Kit

Bioneovan HBcAb ELISA Kit
**Interpretation of HBsAg Results**

Negative control values must have an absorbance of $\leq 0.10$ after subtracting the blank.

Positive control value must have absorbance $\geq 1.00$ after subtracting the blank.

Cut-off Value (COV) Calculation:

Mean of Negative Controls (NC) $\times 2.1$

When the OD value of negative control is less than 0.05, it is reported as 0.05. If it is more than 0.05, it is reported as the actual OD value measured.

Positive reading: $\geq$ Cut-off value

Negative reading: $< \text{Cut-off value}$

**Interpretation of HBcAb Results**

Cut-off Value (COV) Calculation:

$\text{COV} = 0.5 \times \text{the average Optical Density (OD)}$ of negative controls (If the absorbence of negative controls is below 1.50, Calculate it was calculated as original value. If the absorbance of the negative controls is above 1.50, it was calculated as 1.50)

Positive OD$_{450}$ of sample $\leq \text{COV}$

Negative OD$_{450}$ of sample $> \text{COV}$

Borderline (S/CO = 0.9-1.1): Samples with absorbance to Cut-off ratio between 0.9 -1.1 are considered borderline samples were retested. Repeated reactive samples were considered positive for anti-HBc.
Interpretation of HBeAg Results

Cut-off Value (COV) Calculation:

The average of OD values of Negative control multiply by 2.1:

If the OD value of the negative control is less than 0.07, it should be reported as 0.07. If it was more than 0.07, it was reported as the actual OD value measured.

Positive OD reading: ≥ Cut-off value

Negative OD reading: < Cut-off value
APPENDIX C
Ethical Clearance
GHANA HEALTH SERVICE ETHICAL REVIEW COMMITTEE

In case of reply the number and date of this Letter should be quoted.

My Ref. : ERC-
Your Ref. No.

Rexford Bempong Adade,
University of Ghana,
College of Health Sciences,
Accra.

Protocol ID NO: GHS-ERC 89/03/13
Country: Ghana
Protocol Title: “Endemicity of Hepatitis B in Ghana”

Dear Rexford Bempong Adade,

Please find the review summary of the Protocol titled: “Endemicity of Hepatitis B in Ghana” that was submitted to the ERC Secretariat for review. This proposal underwent full general meeting review. In a cover letter please address your responses to each of the in sections A POINT BY POINT, and submit a revised, amended protocol accordingly. All changes should be marked in bold.

Issues of Concern to be addressed
A. Main Protocol
i. There is the need for an abstract. Please provide one.
ii. Indicate which HIV sentinel survey you will use. (2010 or 2011; pages 15 and 16)

Decision
Based on the above comment, the Committee has made the following decision for this protocol.
The proposal is approved conditionally, subject to the Amendments requested above being incorporated into the proposal to the satisfaction of the Responsible officer and ERC.

Ag. Administrative Secretary, Ghana Health Service Ethical Review Committee
For: Chairman
Name: Abena Kwaa Addai-Donkoh

May 10th, 2013

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