SEROPREVALENCE OF HEPATITIS B SURFACE ANTIGEN (HBsAg) AND ASSOCIATED FACTORS AMONG PREGNANT WOMEN IN JIGJIGA TOWN PUBLIC HEALTH FACILITIES, SOMALI REGIONAL STATE, ETHIOPIA

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Summary

Background: *Chronic Hepatitis B virus infection is the most common cause of hepatocellular carcinoma , accounting for 50% of hepatocellular carcinoma cases worldwide and up to 80% of cases in high hepatitis B virus endemic regions. Transmission of hepatitis B virus from carrier mothers to their babies can occur during the perinatal period, and appears to be the most important factor in determining the prevalence of the infection in high endemicity areas. Data on sero-prevalence and associated factors has paramount important in the prevention and control of the disease, therefore the present study was undertaken to generate data on sero-prevalence and associated factors for hepatitis B virus among pregnant women in the study area.*

Objective: *To determine the sero-prevalence of Hepatitis B Surface Antigen among Pregnant Women and to identify associated factors with the infection among pregnant women attending antenatal care clinic in Jijiga town public Health facilities, Somali Regional State.*

Methods: *Institution based cross-sectional study was conducted in the antenatal care clinics of Karmara Hospital and JIjiga Health Center from September26 to October 26/2013. Data including socio-demographic and economic characteristics were collected using a structured questionnaire, following the interview blood specimens were also collected for detection of hepatitis B virus surface antigen from 222 pregnant women attending antenatal clinics of both health facilities. Then bivarate and multivariate logistic regression analysis were employed to see the association between HBsAg sero-positivity and various factors, afterwards AOR 95% confidence level with P-value less than 0.05 was considered as statistically significant.*

Results: *In this study, 4.5% of the pregnant women were reactive for HBsAg. Among the various factors affecting HBsAg sero-positivity, history of hospitalization (AOR [95%CI] =6.4[1.6-26.3]) with P=0.009, and pregnant women that had dental procedure (AOR [95%CI] = 5.2[1.3-21.1) with P=0.02 were found as independent predictors.*

Conclusion: *This study has shown that HBV prevalence in pregnant women is of intermediate endemicity. Regarding risk factors history of hospitalization and dental procedures were the independent predictors for sero-positivity of HBsAg. Hence routine screening of pregnant women for HBV infection and improving aseptic techniques in health facilities is needed.*

Key words: *Hepatitis B surface antigen, pregnant women, Somali Regional State.*

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List of acronyms and abbreviations:

ALT	Alanine Amino Transferase
AOR	Adjusted Odds Ratio
APGAR	American Pediatric Gross Assessment Record
CHB	Chronic Hepatitis B
CI	Confidence Interval
CMV	Cytomegalovirus
COR	Crude Odds Ratio
CSA	Central Statistical Agency
DNA	Deoxyribonucleic Acid
HBcAg	Hepatitis B Core Antigen
HBeAg	Hepatitis B Envelop Antigen
HBIG	Hepatitis B Immunoglobulin
HBsAb	Hepatitis B Surface Antibody
HBsAg	Hepatitis B Surface Antigen
HBV	Hepatitis B Virus
HCC	Hepato Cellular Carcinoma
HIV	Human Immune Virus
HPV	Human Papiloma Virus
HSV	Herpes Simplex Virus
OR	Odds Ratio
PNC	Post-Natal Care
PMTCT	Prevention of Mother to Child Transmission
MCH	Maternal and Child Health
SOP	Standard Operating Procedures
STI	Sexually Transmitted Infections
WHO	World Health Organization

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CHAPTER 1: INTRODUCTION

1.1.   Background

Immunologic changes of pregnancy may induce a state of increased susceptibility to certain intracellular pathogens, including viruses, intracellular bacteria and parasites. In relation to this viral infections in pregnancy are a major cause of morbidity and mortality for both mother and fetus. Viral STIs occur as surface infection and then gradually infect immunologically protected sites, moreover, these are asymptomatic, hidden and hence under diagnosed, persistent and difficult to treat, of the different viral agents HSV, HPV, HBV, HIV and CMV are the common ones. Most of these are transmitted during intrapartum period hence proper screening, identification and treatment offered during prenatal period may help in preventing their complications [1].

Hepatitis B is a viral disease process caused by the hepatitis B virus (HBV). The virus is endemic throughout the world. It is shed in all body fluids by individuals with acute or chronic infection.[2]
It is   partially double-stranded DNA virus of the Hepadnaviridae family. Four major serotypes (adw, ayw, adr and ayr) and nine minor subtypes have been serologically identified at the hepatitis B surface antigen (HBsAg) level and the complete sequencing of DNA from HBV isolates worldwide has led to the identification of eight genotypes (from A to H). [3,]

Chronic hepatitis B (CHB) in pregnancy is an important and pervasive issue with unique challenges, however, for pregnant women with chronic hepatitis B virus (HBV) infection, unlike the general population, many special problems need to be considered, such as the influence of HBV infection on the mother and fetus, influence of pregnancy on HBV replication, effects of antiviral treatment on maternal and neonatal outcomes, immunization of newborns and the possible flare of hepatitis after delivery. [4, 5]

1.2. Statement of the problem.

In the last few years, more and more data have been produced in developing countries and areas with high or intermediate endemicity where the most common route of infection is still vertical transmission from mother to child and horizontal transmission between children, particularly siblings.[6]

Studies have shown that chronic HBV infection is associated with gestational diabetes mellitus, antepartum hemorrhage, threatened premature labor and lower APGAR score. Mothers with seriously abnormal liver function complications are prone to postpartum hemorrhage, puerperal infection, low body weight infants, fetal distress, premature birth, fetal death and neonatal asphyxia. A series of physiological changes occur during pregnancy, including vigorous metabolism and increased nutrient consumption, these changes occur to promote the metabolic needs of the mother as well as the needs of the growing fetus, abundant sex hormone produced by the mother needs to be metabolized and inactivated in the liver, metabolism and detoxification in the fetus also depend on the mother's liver, which correlates with aggravation of pre-existing liver diseases and exacerbation of liver damage. Although HBV infection during pregnancy can often be tolerated, severe hepatitis and hepatic failure induced by perinatal hepatic flare reactions still occur, and can have an unfavorable outcome. [4]

Two billion people worldwide have serologic evidence of past or present HBV infection, and 350 million are chronically infected and at risk of developing HBV-related liver disease. Some 15–40% of chronically infected patients will develop cirrhosis, progressing to liver failure and/or HCC. HBV infection accounts for 500,000–1,200,000 deaths each year.[2]

About 350 million people are infected with HBV worldwide and 50% of them have acquired their infection in the perinatal or neonatal period, especially in countries where HBV has a high prevalence, in these countries, women of childbearing age have a higher hepatitis B e antigen (HBeAg)-positive rate and a higher probability of mother-to-infant transmission, the younger they are when infected with HBV, the higher the risk of developing CHB. [4]

According to WHO   Africa has the second largest number of chronic carriers after Asia and is considered as a region of high endemicity, despite the fact that the exact burden of hepatitis B in Africa is difficult to assess due to inaccurate records and under-reporting, it is estimated that between 70 and 95% of the adult population show evidence of past exposure to HBV infection and the estimated HBsAg sero-prevalence ranges from 6-20% [7]

Sero-prevalence study of HBV conducted among pregnant study subjects in Nigeria, Gahana and Niger state Nigeria revealed that prevalence of HBsAg as 11%, 10.6% and 6.5% respectively. [8, 9,10]

In Ethiopia community based cross-sectional study conducted on seroepidemology of HBV revealed that prevalence of HBsAg was 7%, this finding placed Ethiopia under the region of intermediate indemicity according to WHO regions for HBV[11]. In addition   some institution based studies on sero- prevalence of HBV among pregnant women at Gondar and Jimma have shown 5.3 % and 3.7 % respectively. [12, 13]. Therefore these finding   in different African countries depict that perinatal transmission accounts for major share of transmission routes.

Previous studies on sero-prevalence of HBsAg among pregnant women in different parts of the world attempted to show the presence and strength of association between sero-status of pregnant mothers and independent predictors , in Ethiopia also previously conducted studies contribute a lot by providing baseline information for policy makers and researchers therefore this study also will generate data on HBV sero-epidemology following the fact that absence of data on HBV for researchers and policy makers

CHAPTER 2: LITERATURE REVIEW

2.1. Prevalence of HBV

Two billion people worldwide have serologic evidence of past or present HBV infection, and 350 million are chronically infected and at risk of developing HBV-related liver disease, some 15–40% of chronically infected patients will develop cirrhosis, progressing to liver failure and/or HCC. HBV infection accounts for 500,000–1,200,000 deaths each year. The prevalence of chronic infection ranges from over 10% of the population in South-East Asia, China, the Amazon area, and sub-Saharan Africa to less than 1% in Western Europe and North America. Overall, approximately 45% of the global population lives in areas of high endemicity. [2]

Transmission of HBV from carrier mothers to their babies can occur during the perinatal period, and appears to be the most important factor in determining the prevalence of the infection in high endemicity areas, particularly in China and Southeast Asia. Epidemiological studies on HBV intrauterine infection in China showed that intrauterine infection occurs in 3.7-9.9% pregnancy women with positive HBsAg and in 9.8-17.39% with positive HBsAg/HBeAg. [6]. In Africa also some institution based sero-prevalence study of HBV among pregnant study subject in Nigeria, Gahana and Niger state Nigeria revealed the prevalence as 11%, 10.6% and 6.5% respectively. [8,9,10] similarly in Ethiopia some institution based studies on sero-prevalence of HBsAg among pregnant women at Gondar and Jimma have shown 5.3 % and 3.7 % respectively. [12, 13]

2.2. Factors associated with sero-prevalence of HBV among pregnant women

Hepatitis B virus (HBV) infection is a viral infection spread by contact with infected blood or bodily fluids, including semen and saliva. Transmission of HBV from carrier mothers to their babies can occur during the perinatal period, and appears to be the most important factor in determining the prevalence of the infection [6]. Therefore in this study different literatures were reviewed to perceive factors that are associated with sero-prevalence of HBV among pregnant women.

2.2.1. Socio- demographic and economic factors

In most studies factors like age, educational level, marital status, gravidity, occupation and income, were considered to see its association with HBV infection as follows.

Age of the pregnant mothers considered to have significance relationship with HBsAg prevalence some literatures in different parts of the world revealed different findings. A study conducted in India shown that the highest prevalence rate was observed in the age group of 21–25 years with no significant relationship with other age group. [13] Another study conducted among pregnant mothers in Ghana also revealed that, there were no significant age-related differences between different age group and the sero- positivity. [9]. In Ethiopia also some t studies revealed that age has no significance association with incidence of HBV as shown by a study done in Jimma the highest age-specific prevalence was in the age group of >40, however there was no linear relationship between prevalence of HBsAg and age in all the study. [12] Similarly institution based cross-sectional conducted in South Gondar to determine the sero-prevalence of HBsAg among pregnant women attending antenatal clinic also revealed no statistically significant difference in HBsAg prevalence across various age groups (p>0.05). [13]

A study conducted in Nigeria among 180 pregnant mothers revealed that those subject under the age of 20-29 recorded higher percentage of HBsAg positivity 6.1% (χ^2=7.902, P-value=0.048) according to these finding age of acquiring infection was significantly associated with sero-status pregnant women [16].

Regarding educational attainment of the pregnant mother most studies in different parts of the world revealed that those with lower level of education has relatively higher risk of HBV infection compared to those with high level of education. A study done in Brazil among 541 pregnant women shown that the presence of serological markers for HBV infection was associated with lower educational levels [18]. Sero-prevalence survey done in Vietnam also shown that those with less than 6 years schooling were more likely to be HBsAg positive compared to those with higher education (14.0% vs. 6.6%, p=0.02). [19].

Regarding marital status of the pregnant women   some literature in different countries shown that marital status of has not been considered as predisposing factors for HBV for example , a study conducted in Manila revealed that , when marital status is considered as a risk for infection, the difference between the infected single and infected married cases is not significant. [15], similarly a study done in Nigeria shown that when marital status was considered as a predisposing factor to HBV infection, it was not statistically significant (χ^2 = 0.8109, p = 0.368) [24]

In relation to the association between gravidity with risk of acquiring HBV infection for pregnant women some literatures shown that no direct association between risks of HBV for example a cross-sectional study carried out among pregnant women attending antenatal clinics in Nigeria indicated that when premigravidea and multigravidea compared against the risk of infection with hepatitis B infection, there was no statistically significant relationship between the two groups (χ^2 = 2.244, p = 0.814) [24]. Similarly another study in Manila also revealed that, when the two groups (multigravidea and premigravidea) are compared against the risk of infection with hepatitis B infection, the difference is not statistically significant with odds ratio (OR) of 0.73. A survey1,500 pregnant women in Eastern region of Ghana and a studies in Mali and Egypt Also shown that there was no statistically significant difference in gravidity and risk of HBV infection (p-value 0.236) [9, 25, 26].

Regarding the occupation of pregnant women a study conducted in Manila shown when the different job categories were compared against hepatitis B infection, it is not statistically significant (OR = 1.24) [15]. Whereas another study conducted in Nigeria also shown out of the 36 healthcare workers, 8 (22.2%) tested positive to HBsAg, and this was statistically significant (χ^2 = 51.22, P = 0.001)[24]

2.2.2. Hospital associated factors

Regarding hospitalization history some literatures point out significant association between history of hospitalization and HBV incidence. A study done in Manila among pregnant mothers revealed that those who reported hospitalization was compared with those who did not have a history of hospitalization and there was no significant difference (OR = 1.07) in the risk of infection with Hepatitis B virus. [16]. Another study conducted in Saudi Arabia revealed pregnant women with history of hospitalization showed a significant association with anti- HBV sero-positivity (P-value 0.050 and 0.001 respectively) [28]

Related to hospital associated factors blood transfusion directly accountable for the transmission of most of microbial disease, some studies revealed significant association with incidence of HBV and blood transfusion. A study conducted among 544 volunteer family planning attending women at Family Planning Center, in Yemen found that there was a highly significant association of history of blood transfusion, with contracting HBV infection. Similarly risk factor assessment done in rural Vietnam, shown those with history of blood transfusion also had higher frequency of past HBV infections (68.4 vs. 45.7%, p=0.05). [23, 29]. Another study in India also shown that the infection was significantly more prevalent in HBsAg-positive women with history of blood transfusion (6/12; 50 %)   (p = 0.0015) [20]. Whereas a study done in Saudi Arabia revealed that transfusion history did not prove association with HBV.[23], likewise another study conducted in Manila also identified that transfusion history was not statistically significant with odds ratio of 1.0. [15]

Under hospital associated factors minor or major surgery and dental extraction are common practice that might be responsible for transmission of most of microbial infections.   Regarding surgical procedure a study conducted among pregnant women in Manila identified that those who had any surgical procedure, major or minor were compared with those who had none, of the 273 patients with surgery, 11 (3.99%) were infected and 25/652 (3.83%) patients who had no surgery were infected but the difference between the two groups was not significant (OR = 0.96%). [15].

Pertaining to dental procedure and HBV incidence HBsAg sero-prevalence study conducted in Iran among pregnant women revealed that history of dental procedure has no significant relationship between the two groups with positive and negative serum surface [21].Another study done in Manila also identified the risk of infection with a history of a dental procedure and comparison was between with those who had none (40.8%), and of those with dental procedure, 3.82% was positive for HBsAg while it was 3.96% for those who had no dental procedure, the difference between the two groups was not significant with OR = 1.04. [15] Another   study in Pakistan also revealed tooth extraction as a risk for maternal infection of HBV. [30]

2.2.3. Risky socio cultural and behavioral factors.

Socio-cultural practices like tattooing, ear/ nose piercing and multiple sexual partners   are some of the common factors that exist today responsible for the exchange infectious materials between different individuals, being HBV found in various body fluids. Parenteral or percutaneous routes of HBV transmission, such as needle stick injury and mucus membrane splash in healthcare setting, as well as tattooing, piercing, sharing razors or toothbrushes, are also important in spreading the virus.[6]

Regarding practice of tattooing, ear/nose piercing and risk of HBV among pregnant women a study conducted in Nigeria to assess the risk of HBV infection during pregnancy   revealed that two groups (HBsAg reactive or non-reactive) did not differ with respect to presence of tattoo or scarification marks,(p>0.05 [22]. Another systematic review of prevalence, risk factors, awareness status and genotypes in Pakistan also revealed that risk factors in pregnant women (antenatal) include ear and nose prick were not significantly associated [30]. Whereas Unlike another study conducted in Gondar, Ethiopia revealed that pregnant women with tattoo on their body had statistically significant association with the HBsAg sero-positivity (p<0.05). [13]

Abortion is also one of risky behavioral factors that increase the incidence of HBV infection among pregnant women, for example a study done in Jimma revealed that pregnant women who experienced abortion had higher prevalence of HBsAg (7.3%) and the odds of having HBsAg was more than twice with those pregnant women that had history of abortion than with other risk factors. Similarly a study conducted in Nigeria revealed that, there was no statistical relationship between previous termination of pregnancy and acquisition of HBV o infection [13, 31]

In relation to risky behavioral practice sexual behavior of the individual might determine the risk of HBV infection. A prospective case control study done in Nigeria revealed that of the 1,052 women attending the antenatal clinic, 6.08% (n = 64) were positive for HBsAg, among the different significant risk factors history of multiple sexual partners was found to be significantly associated with sero-positivity of HBsAg (AOR = 2.02; 95% CI = 1.02-3.98; P = 0.0427).[23] .Similarly a study conducted in India to asses various factors in relation to HBV infection shown that the infection was significantly more prevalent in HBsAg-positive women with high risk sexual behavior of the husband (8/12; 66.6 %). (P = 0.0015) [20]

2.3. Conceptual frame work

This conceptual frame work was designed to show the association between HBV sero-status of pregnant women (dependent variable) and associated factors (independent variables). The bidirectional arrow depicts the bilateral relationship of variables and the single arrow describes the effect of one variable to the other towards the direction of the arrow.
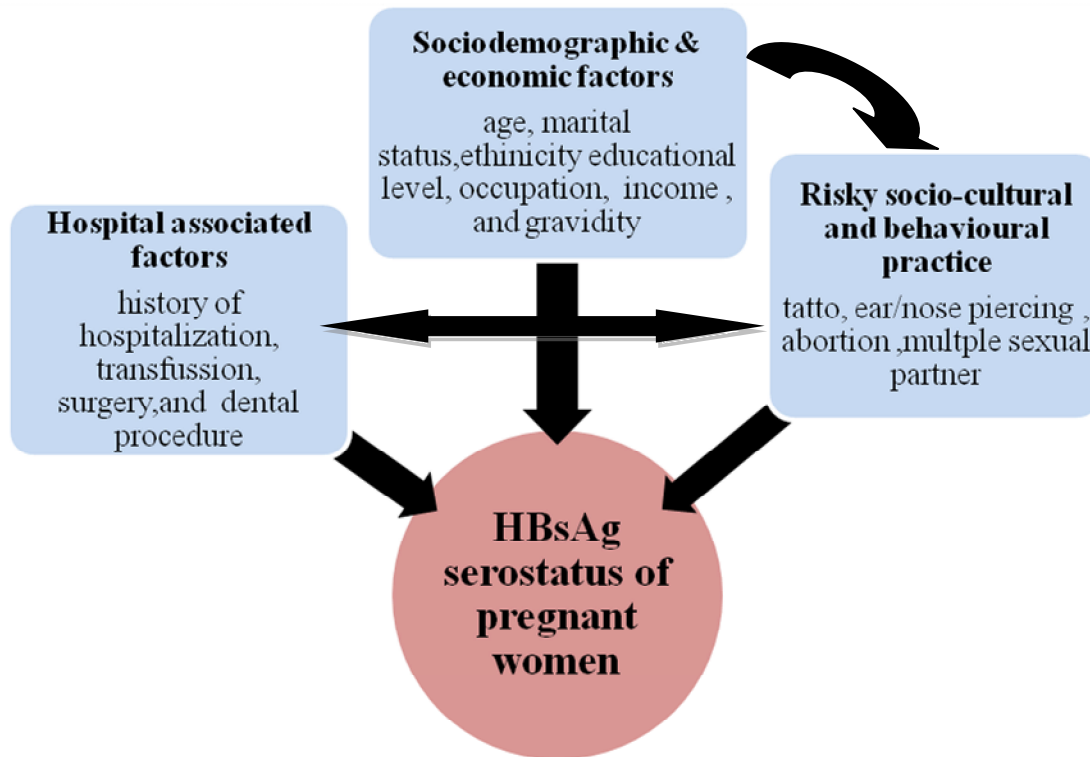
Figure 1: Conceptual frame work showing the relationship between the dependent variable and independent variables *(designed following review of literatures)*

2.5. Significance of the study

The prevalence of HBV infection among antenatal population may give some indication on prevalence in the general population. Hence, screening antenatal women for HBsAg can give clue to estimate prevalence of the disease in a population and provide an avenue for preventing mother to child transmission of the virus. Moreover prevention of vertical transmission is extremely important because HBV infection in early life can result in a chronic carrier state. [25]

It is obvious that the introduction of routine hepatitis B screening among pregnant women and its prevention control requires updated data on the magnitude of HBV, besides HBV infected pregnant women are at risk of infecting their babies with a consequence of developing fulminant HBV infection, therefore following the fact that lack of data on HBV among pregnant women in the study area, the present study will provide valuable information on the magnitude and associated factors on HBV and helps to draw possible recommendation for prevention and control purpose .

CHAPTER THREE: OBJECTIVES

3.1. General objective:

To determine the sero-prevalence of HBsAg among pregnant women and to identify associated factors among pregnant women attending ANC follow up in Karamara Hospital and Jigjiga Health center Somali Regional State by 2013/14.

3.2. Specific objectives:

- To determine prevalence of HBsAg among pregnant women
- To assess socio-demographic and economic factor and their association with HBV
- To assess hospital associated factors and its association with HBV
- To identify socio cultural and behavioral factors and their association with HBV

CHAPTER FOUR: METHODS AND MATERIALS

4.1 Study setting and period

Somali Regional state is one of nine regions of the Federal democratic Republic of Ethiopia. Located in the eastern Ethiopian lowlands bordering Djibouti, Somalia and Kenya, locally bordering with Oromia, Harari, and Afar region. According to Ethiopian Central Statistics Agency the estimated region's population at just over 4.5million in (2007 ECSA).

The region covers   271,970.91 square km-second in Ethiopia, next to Oromia regional state with topography of 500-1600 meters above sea level and the climate is warm, hot and arid. The region has 9 zones and 67 districts (Woreda) administrative. The Capital city is Jigjiga located 635 km from Addis Ababba, divided into 10 Kebele & 67 sub-kebeles. The income of inhabitants of the city mainly depends on small and large scale trading. Regarding health facilities the town has one referral hospital (under construction), one general hospital, two health centers (one new) and ten health posts (government)   there is one private general hospital, 27 higher clinics, 10 medium clinics and the study was conducted at Karamara hospital and Jijiga health center from September 26-October 26/2013.

4.2. Study design

Institution based cross sectional study design was used.

4.3. Population

4.3.1 Source population

The source populations were all pregnant mothers attending both health facilities.

4.3.2 Study population

The study population were those mothers attending ANC clinic of the study area who fulfilled the eligibility criteria.

4.4 Sample size determination and sampling procedures

4.4.1 Sample size determination

The single population proportion formula was used with the following assumptions; the proportion of pregnant mother with positive HBsAg is 5.3. % according to similar study conducted in Gondar[12] with margin of error 3% and 95% confidence interval.

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

$$n = \frac{(1.96)^2 * (0.053)*(1-0.053)}{(0.03)^2}$$

$$n = 214 + Nr(5\%) = 225$$

Where:

n -Minimum sample size

z- Confidence interval level 95%

p- Proportion of pregnant women with HBV = 5.3%

d – Margin of error 3%

Nr – Non- response rate

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4.4.2. Sampling technique.

In Jigjiga town there are two public health centers (one new) and one public general hospital, of which Karamara Hospital and Jigjiga Health center provides full scale ANC service therefore study participants were selected from these two health facilities, then the number of study units to be sampled from selected public health facilities was determined using proportional allocation to size based on the number of client flow in the previous quarter total average one month case load of the selected health facilities (258) and those pregnant women who fulfill the illegibility criteria were enrolled conveniently until the required sample size were achieved.
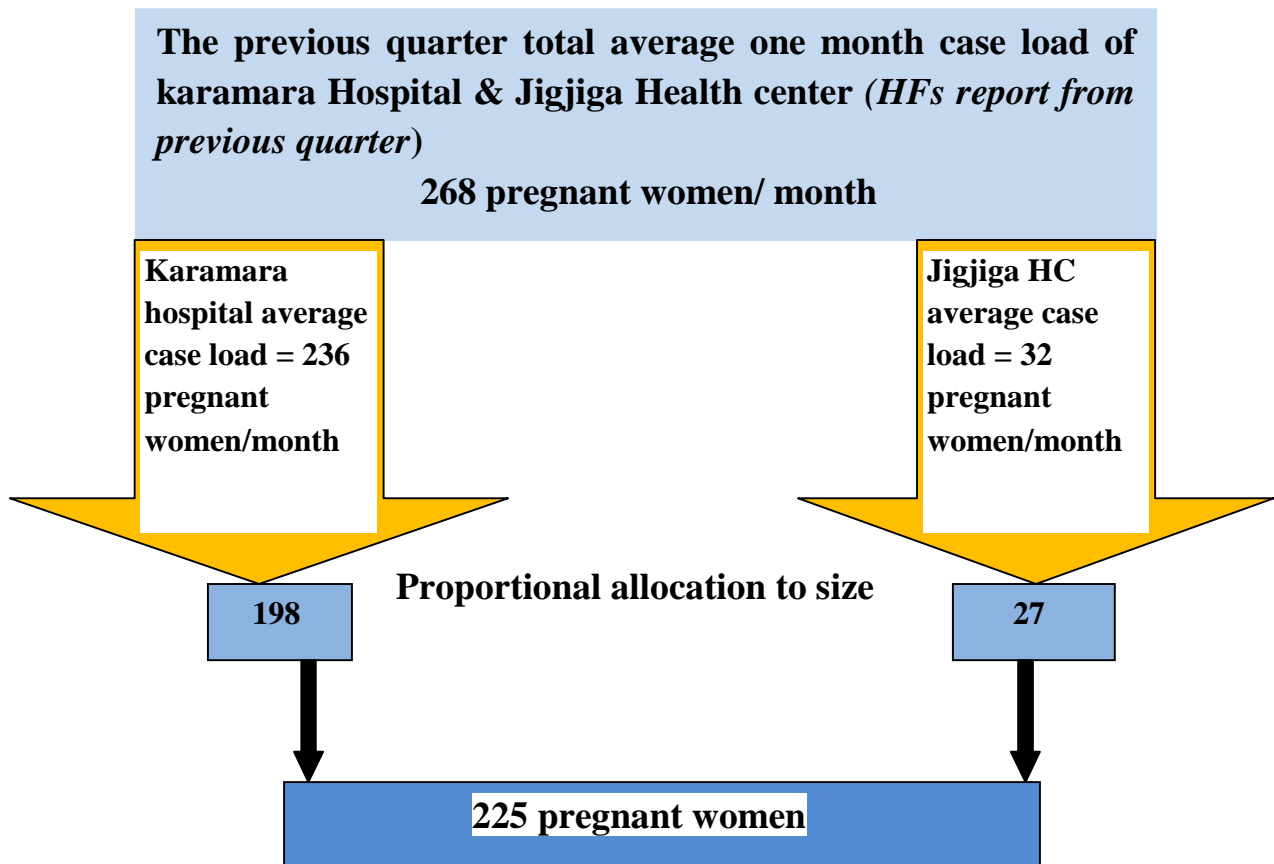
Fig 2. Schematic presentation of sampling procedure for the selection of study units in Jijgjiga town public health facilities, Somali Regional State 2013

4.4.3. Sampling unit.

Pregnant women visiting the ANC clinic of both health facilities.

4.4.4 Inclusion and exclusion criteria

- *Inclusion criteria*

Pregnant women, those who came for ANC.

- *Exclusion criteria*

Pregnant women, attending ANC but unwilling to participate in the study and severely ill

4.5 Study variables

4.5.1. Dependent variable

HBsAg sero-status

4.5.2. Independent variables

- Age
- Educational level
- Income level
- Occupation
- Marital status
- Gravidity
- History of dental procedure
- History of surgical procedure
- Tattooing
- Abortion
- Blood transfusion
- Ear piercing
- Risky sexual behavior

4.6 Data collection process

A structured questionnaire was originally developed in English and translated into Somali and then back translated to English. The questionnaire was pre-tested in the study area. The content of the questionnaire includes socio-demographic and economic variables, hospital associated factors, and risky socio-cultural and behavioral practice. After brief explanation about the aim of the study the participants were requested to take part in the study voluntarily and also informed about 4 ml blood will be drown to determine their hepatitis B sero-status and other ANC related laboratory profiles, after the verbal consent was accepted by the subjects the data collector in charge (senior Midwifery nurse) was conducted the interview followed by coding and transferring of the subjects to the laboratory, then senior laboratory technicians in charge of the data collection (one sample collector and one analyzer) collect the blood sample

Regarding specimen collection 4ml of blood was collected aseptically in anticoagulant free test tube, using SOP in each health institutions then sample kept for about 30 minutes at room temperature for natural clotting to take place then serum was separated at 3000rpm for 5 minute using electric centrifuge afterward the serum sample was tested immediately using one step NOVA cassette style HBsAg Test kit.

NOVA cassette style HBsAg rapid test strip was used for the detection of hepatitis B surface antigen (HBsAg), the test has sensitivity and specificity of approximately 99.7% and 99.3%, respectively when performed according to the instructions of the manufacturer. The test strip is a one step rapid direct binding test for the visual detection of hepatitis B surface antigen (HBsAg) in serum as an aid in the diagnosis of hepatitis B infection. One step HBsAg Test is based on the principle of sandwich immunoassay for determination of HBsAg in serum, monoclonal and polyclonal antibodies are employed to identify HBsAg specifically. This one step test is very sensitive and only takes about 10-20 minutes. Test results are read visually without any instrument.

Test procedure

1. When the serum sample is ready after processing, open the sealed pouch by tearing along the notch, remove the test from the pouch
2. Draw 0.2ml (about 4drops) sample in the pipette, and dispense it in to the sample well on the cassette
3. Wait 10-20minutes and read result. Do not read result after 30 minutes.

Interpretation

Non-Reactive: only one colored band appears on the control (C) region. No apparent band on the test (T) region.

Reactive: In addition to a pink colored control (C) band, a distinct pink colored band will also appear in the test (T) region.

Invalid: A total absence of color in both regions is an indication of procedure error and /or the test reagent has deteriorated.

4.7 Data processing and analysis.

Data were entered into a computer using Epi data and double entry verification was made. Then data were exported in to SPSS version -16 statistical software followed by data cleaning and analysis. Descriptive analysis was made to check for outliers, inconsistencies and missed values for the variables, subsequently bivariate logistic regression was made to see the association between each covariate and outcome variable, then to control the effect of confounding factors and to get independently associated variables bivariate analysis with 20 % level of significance was selected and entered in to a multiple logistic regression model using backward stepwise method. Wald test with 5% level of significance was used to identify independent covariates for HBsAg sero-status of pregnant women. Furthermore collinearity diagnosis was made to see interaction among independent predictors (history hospitalization and of dental procedures). Finally the goodness of fit of the final model was checked using Hosmer and Lemeshow statistics.

4. 8. Data quality assurance.

In order to achieve   good data quality data collectors were recruited based on their profession and qualification for that reason senior Midwifery nurse   and laboratory technician   were involved in the   data collection moreover   ability to speak the local language and previous experience of data collection was considered. Before the data collection begins orientation was provided to the data collectors on objectives of the study, steps and approach for interviewing, proper sample collection, processing and testing and recording of the result. Vague points and other problems encountered about the questionnaire was explained and clarified prior to the data collection. During the time of data collection firm supervision was undertaken by the investigator and supervisors to ensure good quality of data and appropriateness of the blood sample collection and processing in accordance with SOP in each health facilities. At the end of each data collection date the questionnaire was crosschecked daily by the supervisors and the principal investigator and problems faced was discussed over night with data collectors and the supervisors.

Regarding quality and specification of the test kits currently different brands of rapid HBsAg test kits are commercially available, with high sensitivity and specificity for example according to Comparative Evaluation of Immunochromatographic Rapid Diagnostic Tests (Strip and Device) and PCR Methods for Detection of Human Hepatitis B Surface Antigens in Iran, device or strip from six companies compared with PCR and found >97.5% sensitivity and specificity [27]. Therefore in this study NOVA cassette style HBsAg rapid test strip was used for the detection of hepatitis B surface antigen (HBsAg).The test has sensitivity and specificity of approximately 99.7% and 99.3%, respectively.

4.9 Ethical considerations.

The study was obtained ethical clearance from ethical committee of Jimma University, College of Public Health and Medical Science, permission paper was also obtained from Somali Regional State Health Bureau Research and Technology Sub-process coordinator and respective heads of the study institutes. Afterward clear discussion about the actual study has been made for each subject, then following the acceptance of verbal informed consent, pre-test counseling was made by the MCH nurse in charge, issue of confidentiality also discussed therefore identifying participants by name was avoided, lastly after the result of the participants obtained from the lab post test counseling and referral linkage to the physician was made for those pregnant women with reactive HBsAg test.

4.10 Plan for dissemination of findings.

The final result of this study will be presented to Jimma University, College of Public Health and Medicine and disseminated to Somali Regional State Health Bureau, Karamara Hospital and Jigjiga Health Center other concerned governmental and nongovernmental organization in addition effort will be made to publish on local or international journal.

4.11. Operational definition.

Reactive: In addition to a pink colored control (C) band, a distinct pink colored band will also appear in the test (T) region, that indicate the presence of HBsAg in the serum.

Non-Reactive: only one colored band appears on the control (C) region. No apparent band on the test (T) region that indicates the absence of HBsAg in the serum.

Invalid:-No visible band at all or there is a visible band only in the test region but not in the control region

History of hospitalization: - refers to the previous history of hospital admission of the study subject for any disease in the past.

Blood transfusion history: - refers to the transfusion history of the study subject for any problem in the.

History of dental procedures: - refers any surgical procedures in the study subject in the past

History of surgical procedures: - refers to any surgical procedures minor or major practiced on the study subject in the past.

Risky Socio cultural factors: - refers to some of the social and cultural activities that were practiced by the study subject and able to serve as a means of exchange of body fluids and germs between individuals for example ear or nose piercing, contact with family member during visiting and caring of the sick in the past.

Risky behavioral factors: - refers to some of the behavioral malpractice of the subjects that exposed them to the risk of acquiring HBV infection from different source like abortion in the past.

CHAPTER FIVE: RESULTS

5.1. Socio-demographic and economic characteristics

Two hundred twenty two (98.67%) pregnant women were included in the analysis, the rest (1.32%) were excluded due to incomplete result on major outcome variables. Of 222 pregnant women enrolled in the study, 195 (87.8%) were from Karamara Regional Hospital and the remaining 27(12.2%) were from Jigjiga Health Center. The mean age of the mothers' was 26.65 (± 4.92) years old and majority of the pregnant women 154(69.4%) were under 28 years old. Amongst the 222 pregnant mothers 181 (81.5%) were from Somali ethnic group, the remaining18.5% were from, Oromo, Amhara, Tigre and Gurage. Regarding marital status, almost all (96.8%) of the study participants were married.

In terms of the educational level, 124 (55.8%) women couldn't read and write, 19 (8.6%) attended informal education and the remaining 79 (35.6%) attended formal education. Regarding occupation, majority (83.8%) of them were housewife and the remaining were formal employ (16.2%). Concerning the average monthly income of the participants, 83(37.4%) were had an average monthly income of <1634 Birr and the remaining139 (62.6%) had >1634 Birr. As to the number of pregnancies, 57(25.6%) of them were primigravidae and the remaining, 165(74.4), were multigravidea. (Table 1)

Table 1. Socio-demographic and economic characteristics of pregnant women (n=222) attending antenatal clinic of karamara Hospital and Jigjiga Health Centre, Jigjiga , Eastern Ethiopia November, 2013.

Variables	No (%)
Age in years	
<28 years old	154(69.4)
>28 years old	68(30.6)
Ethnicity	
Somali	181(81.5)
Others*	41 (18.5)
Marital status	
Married	215(96.8)
Others**	7(3.2)
Educational level	
Cannot read write	124 (55.8)
Read and write (informal)	19 (8.6)
Formal education	79 (35.6)
Occupation	
House wife	186 (83.8)
Formal employee	36 (16.2)
Monthly income	
<1634 Birr	83 (37.4)
>1634 Birr	139 (62.6)
Gravidity	
Premigravidea	57(25.7)
1-2	86(38.7)
3-4	36(16.2)
>4	43(19.4)

NB. *Others *= Oromo, Amhara, Tigre and Gurage*

*Others **= Single divorced, Cohabited*

5.2. HBsAg sero-status distribution by socio-demographic and economic characteristics

Among 222 pregnant women included in the study 10 of them were reactive for HBsAg which makes the overall prevalence 4.5 % in the study area. In relation to the educational status of the study subjects, 124(55.8) couldn't read and write, of which 4(3.2%) of them were reactive for HBsAg, and 19(8.6%) were attended informal education of which 1(5.3%) was reactive for HBsAg (Table2). Marital status of pregnant women also considered in this study and 215(96.8%) were married of which 10(4.7%) were reactive for HBsAg. Regarding occupation of the study subject 186(83.8%) were housewife of which 9(4.8%) were reactive for HBsAg test, and 36(16.2%) were formal employee of which 1(2.8%) was reactive for HBsAg (Table2)

Table 2. HBsAg sero-status with general characteristics of pregnant women (n=222) attending antenatal clinic of karamara Hospital and Jigjiga Health Centre, November, 2013.

Variables	No (%)	Reactive No (%)	Non-Reactive No (%)
Ethnicity			
Somali	181(81.5)	9(5)	172(95)
Others	41(18.5)	1(2.4)	40(97.6)
Marital status			
Married	215(96.8)	10(4.7)	205(95.3)
Others	7(3.2)	0(0)	1(100)
Educational level			
Cannot read write	124 (55.8)	4(3.2)	120(96.8)
Read and write (informal)	19 (8.6)	1(5.3)	18(94.7)
Formal education	79 (35.6)	5(6.3)	74(93.7)
Occupation			
House wife	186(83.7)	9(4.8)	177(95.2)
Formal employee	36(16.3)	1(2.8)	35(97.2)
Monthly income			
<1634 Birr	83 (37.4)	3(3.6)	80(96.4)
>1634 Birr	139(62.6)	7(5)	132(95)
Gravidity			
Premigravidea	57(25.7)	5(8.8)	52(91.2)
1-2	86(38.7)	2(2.3)	84(97.7)
3-4	36(16.2)	2(5.6)	34(94.4)
>4	43(19.4)	1(2.3)	42(97.7)

NB. *Others *= Oromo, Amhara, Tigre and Gurage*

Others **= Single divorced, Cohabited

5.3. HBsAg sero-status of pregnant women by exposure to hospital associated factors and risky socio-cultural behaviours.

Regarding HBsAg sero-status the pregnant women in relation to exposure with hospital associated factors and risky socio-cultural behaviours 23(3.6%) pregnant women had   previous history of hospitalization of which 4(17.4%) were reactive for HBsAg test, among 12(4%)pregnant women had previous history of blood transfusion of which 3(25%) were reactive for HBsAg. Concerning socio-cultural risky behaviour practice by the study subjects 15(6.7%) pregnant women had tattoo on their body parts and none of them were reactive for HBsAg (Table3).

Table 3. HBsAg sero-status with exposure to hospital associated factors and risky socio-cultural behaviours (n=222) of pregnant women attending antenatal clinic of karamara Hospital and Jigjiga Health Centre, November, 2013.

Expected risk factors		Total N (%)	Reactive N (%)	Non-Reactive N (%)
Hospitalization history				
	Yes	23(10.4)	4(17.4)	19(82.6)
	No	199(89.6)	6(3)	193(97)
Transfusion history				
	Yes	12(4)	3(25)	9(75)
	No	210(96)	7(3.3)	203(96.7)
Dental procedure				
	Yes	24(10.8)	4(16.7)	20(83.3)
	No	198(89.2)	6(3)	192(97)
Surgical history				
	Yes	13(5.9)	3(23.1)	10(76.9)
	No	209(94.1)	7(3.3)	202(96.7)
Spouse with polygamous marriage				
	Yes	20(9)	3(15)	17(85)
	No	202(91)	7(3.5)	195(96.5)
Marriage before current husband				
	Yes	16(7.2)	3(18.8)	13(81.2)
	No	206(92.8)	7(3.4)	199(96.9)
Tattoo				
	Yes	8(3.6)	0(0)	8(100)
	No	214(96.4)	10(4.7)	204(95.3)
Ear piercing				
	Yes	15(6.8)	0(0)	15(100)
	No	207(92.2)	10(4.8)	197(95.2)
Nose piercing				
	Yes	191(86)	10(5.2)	181(94.8)
	No	31(14)	0(0)	31(100)
Abortion				
	Yes	13(6.3)	1(7.1)	13(92.9)
	No	199(93.7)	9(4.3)	199(95.7)

5.4. Bivariate logistic regression results of hospital associated factors and risky soci-cultural behaviors.

The bivariate logistic regression analysis of hospital associated factors and risky socio-cultural behaviors revealed that previous history of hospitalization, dental procedure, surgical procedure and spouse with polygamous marriage were significantly associated with HBsAg sero-positivity and found to be candidates for the multivariate logistic regression (Table 4)

Table 4. Bivariate logistic regression results of hospital associated factors and risky soci-cultural behaviors. (n=222) of pregnant women attending antenatal clinic of Karamara Hospital and Jigjiga Health Centre, November, 2013.

Factors	Reactive N (%)	Non-reactive N (%)	COR[95%C]	P-value
Surgical history				
	4(17.4)	19(82.6)	5.6[1.3-24]	0.02
	6(3)	193(97)	1	
Hospitalization history				
	3(25)	9(75)	6.7[1.8-26.1]	0.005
	7(3.3)	203(96.7)	1	
Dental procedure				
	3(15)	17(85)	5.5[1.4-20.9]	0.013
	7(3.5)	195(96.5)	1	
Transfusion history				
	3(25)	9(75)	3.9[0.9-16.2]	0.06
	7(3.3)	203(96.7)	1	
Spouse with polygamous marriage				
	3(15)	17(85)	4.9[1.2-20.8]	0.03
	7(3.5)	195(96.5)	1	

5.5. The independent predictors of HBsAg sero-posistivity

Following the bivarate logistic regression analysis, the multivariate analysis was made accordingly previous hospitalization and dental procedure histories were found to be independent factors associated with HBsAg sero-positivity (Table 5). The final model was fit to explain the independent predictors of HBV infection in the study area as it was explained by Hosmer and Lemeshow test result of $\chi^2 = 0.354$ with P-value of 0.552, furthermore multi collinearity and two way interactions were checked and none was found significant.

Table 5. Multivariable logistic regression results of factors affecting HBsAg sero-positivity (n=222) of pregnant women attending antenatal clinic of karamara Hospital and Jigjiga Health Centre , November,2013.

Predictors		Reactive N (%)	Non-reactive N (%)	AOR[95%CI]
Surgical history				
	Yes	4(17.4)	19(82.6)	1.7[0.6 -21.8]
	No	6(3)	193(97)	1
Hospitalization history				
	Yes	3(25)	9(75)	**6.4[1.6-26.3]***
	No	7(3.3)	203(96.7)	1
Dental procedure				
	Yes	3(15)	17(85)	**5.2[1.3-21.1]***
	No	7(3.5)	195(96.5)	1
Transfusion history				
	Yes	3(25)	9(75)	1.2[0.9-6.2]
	No	7(3.3)	203(96.7)	1
Spouse with polygamous marriage				
	Yes	3(15)	17(85)	2.8[1.8-14.9]
	No	7(3.5)	195(96.5)	1

AOR=p-value<0.05*

Therefore pregnant women with past history of hospitalization were six times more likely to be reactive for HBsAg than without history of hospitalization AOR [95%CI] = 6.4[1.6-26.3] with P=0.009 and those pregnant women with past history of dental procedure were five times more likely to be reactive for HBsAg than without history of dental procedure AOR [95%CI] = 5.2[1.3-21.1] with P=0.02 (Table 5).

CHAPTER SIX: DISCUSSION

In this study an attempt has been made to determine the prevalence of HBsAg and associated factors among pregnant women in karmara Hospital and JigJiga Health center, and the study revealed that the prevalence of HBV in the study area was 4.5% hence according to WHO regions of Hepatitis B epidemiology the study area placed under intermediate endemicity [2] This is almost in agreement with two earlier institution based studies conducted among pregnant women in Jimma and Gondar that revealed prevalence of 3.7% and 5.3% respectively [12, 13], but this is lesser than studies conducted in Nigeria and Ghana with prevalence of 11%, 10.6% respectively. [6, 7,], the difference might be due to that the Nigerian study employed simultaneous detection of HBsAg and HBeAg and the Ghanaian study employed large sample about 7 times than the present study.

  The multivariable analysis revealed that among the various factors only history of hospitalization and dental procedure were found as independent predictors of HBsAg sero-positivity, as a result 12(5.4%) pregnant women had at least one episode of hospital admission of which 3(25%) were reactive for HBsAg with AOR [95%CI] =6.4[1.6-26.3] with P=0.009, this implies that those pregnant women with history of hospitalization are six times more likely to be reactive for HBsAg than those without history of hospitalization, which is consistent with Saudi Arabia study that reported pregnant women with history of hospitalization showed a significant association with anti- HBV sero-positivity (P-value 0.001) [28], however this finding is inconsistence with study conducted in Manila that reported hospitalization was compared with those who did not have history of hospitalization and there was no significant difference (OR = 1.07) [15]   the inconsistencies from the present study might be earlier study use radioimmunoassay technique which is highly specific gold standard method than the rapid HBsAg test(chromatographic technique) which employed in the current studies and also due to the difference in sample size in that the present study use 1/4 of the earlier sample size.

In the present study history of dental procedure was also one of independent risk factor for HBsAg sero-positivity, accordingly 20(9%) participants had dental procedure in the past of which 3(15%) were positive for HBsAg test with AOR [95%CI] =5.2[1.3-21.1] with P=0.02 which means pregnant women with history of dental procedure were five times more likely to be

reactive for HBsAg than without dental procedure, this is in agreement with study conducted in Jimma town [9] however inconsistence with study conducted in Iran that revealed history of dental procedure has no significant relationship [21]. The inconsistency from the present study might be the differences in sample size were the Iranian study employed five times larger sample size than this study.

Regarding history of blood transfusion as risk factors for HBsAg sero-positivity in the current study it was not statistically significant with AOR[95% CI=1.2[0.9-6.2], p-value =0.72 and this was different from study conducted in Yemen the discrepancy might be due to the difference in sample size where the Yemeni's study used more than twice of the current[23].

In relation to history of surgical procedure the present study revealed that there is no difference in HBsAg sero-status between pregnant women with history of surgical procedure and without history of surgical procedure with AOR 95% CI] =1.7[0.6-21.8] ,p=0.68 and this was in line with HBV sero-prevalence study conducted among pregnant women in Manila this might be due to that in both studies pregnant women with history of surgical procedure having reactive HBsAg result were few [15.]

Concerning the sexual behavior of the pregnant women for the sake social and cultural value of the participant this study employed indirect approach by asking whether their husband had polygamous marriage or not and the finding was no difference between the two groups AOR 95% CI] =2.8[1.8-14.9], p=0.87 and this was inconsistent with study conducted in India that identified HBsAg positive women with high risks sexual behavior of the husband were significant compared to HBsAg positive women with less risk sexual behavior of the husband the difference might be in the present study the proportion of pregnant women their husband in polygamous marriage were less (3/15; 20%) where as the Indian study were (8/12; 67%) [20.]

Strength and limitation of the study

Strength of the study

- Pre-tested data collection instrument was used, based on the finding the questionnaire were modified to explore the risky sexual behavior of pregnant women indirectly in order to respect the social and religion value of the participants, therefore the final questionnaire was corrected as whether the pregnant women had previous official marriage before the current and whether their husband has polygamous marriage or not.

Limitation of the study

- Unable to explore risky sexual behavior due to cultural and social values might result under reporting
- Due to absence of positive result from the Health Center it might be difficult to generalize the risk factors for both institutions
- Due to finical and time constraint the study employed small sample size.

CHAPTER SEVEN: CONCLUSION AND RECOMMENDATION

7.1. Conclusion

This study has found that the sero-prevalence of HBV infection among pregnant women place the study area under intermediate endemicity (with prevalence of 2-8%) according to WHO classification. Regarding the risk   factors pregnant women with previous history of hospitalization and dental procedure were found to be at risk of HBsAg sero-positivity.

7.2. Recommendation

To Regional Health Bureau and Health facilities

- Based on the current finding the most common risk for HBV infection were nosocomial associated therefore effort should be made to improve the quality of service in health facilities especially on improving the aseptic technique in different procedures..
- Routine screening and immunization of all pregnant women and their infants should be incorporated in the antenatal and postnatal programmes in health facilities to prevent postnatal infection of the infants by their infected mothers.

For researchers

- Large epidemiological studies need to be conducted preferably to have a good contrast both pregnant and non-pregnant women should be included in a future study.
- Further the prevalence of HBeAg should be determined among the HBsAg carriers to determine the extent of perinatal transmission.
- Qualitative studies should be incorporated in further studies to explore risk factors related to sexual behavior of the pregnant women and some risky behaviors like abortion.

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Annex 1. Questionnaire

Department of Epidemiology, College of Public Health and Medical Science, Jimma University, Structured Questionnaire to Assess Sero-prevalence of Hepatitis B Surface Antigen (HBsAg) and Associated Factors among Pregnant Women in Jigjiga Town Government Health Institutions, Somali Regional State,

Information on verbal consent Form

Dear Participant, My Name is _____ I am working in a research team of Jimma University. This questionnaire is prepared to conduct a study titled: *Sero-prevalence of Hepatitis B Surface Antigen (HBsAg) and Associated Factors Among Pregnant Women In Jigjiga Town Public Health Facilties, Somali, Regional State, Ethiopia* You are being asked to enroll in a cross-sectional study; this informed consent form gives you information about the study, which will be discussed with you. Once you understand the study, and if you agree to enroll, you will be asked to sign this consent. Please note that your participation in this research is entirely voluntary. The main purpose of this research study is to determine the sero-prevalence of HBV infection and associated factors among pregnant women at Antenatal Clinic (Karamara Hospial and Jigjiga Health Center) . A total of 225 pregnant women are expected to participate in the study. Once you have agreed to enroll in the study, you will be asked some questions concerning the study and to have blood drawn about 4ml for HBV testing and you may receive no monetary benefit from this study. However, knowledge gained from this study may help in the management of infectious diseases in the future. There is no cost to you for enrolling in the study. Your research records will be confidential. You will be identified by a participant identification number, and personal information from your records will not be released without your written permission. You will not be personally identified in any publication about this study.

Instruction for data collectors:

Interview pregnant women, who come for ANC, encircle the response of the respondents for multiple choice questions and for short answers write on the space provided.

Code No		HBV sero-status _____	
Part I. Socio-demographic and economic characteristics of the respondent.			
No	Question	Response category code	Skip to
100	Date of interview		
101	What is your age in years?	_____yr	
102	Ethnicity	Somali…………………………..1 Oromo…………………………...2 Amhara………………….……3 Gurage …………………....…4 Tigre....……………………....5 Others…………………………6	
103	What is your marital status	Single …………..………..…1 Cohabited………………..…..2 Married ………………......3 Divorced………………..……4	
104	What is your educational level?	Cannot read write…………….1 Read and write (informal)…….2 Elementary (1-6)………………3 Junior (7-8)……………………4 High school (9-12)……………5 Above grade 12………………..6	
105	What is your main occupation?	Health sector……………..……1 Petty trader…………………...2 House wife……………..……...3 Non –employee………..……….4 Formal employee……….…….. 5 Others……………….……….... 6	
106	What is your monthly income?	Cash (in Birr)/month _____ '	
107	Have you ever had any pregnancy before?	YES……………………………………..1 NO……………………………………….2	**201**
108	If yes on question **107**, how many pregnancies have you had? **Enter in number**	_____	
109	How many live births have you had? Enter no	Number _____	

Part II. Questionnaire related to hospital associated factors.			
201	Have you ever been admitted in hospital or clinics in the last 6 months?	YES……………………………..……1 NO…………………………….......…..2	→ **204**
202	Have you ever had dental procedure in hospital or clinics	YES…………………………………...1 NO……………………………....…2	
203	Have you ever been operated for surgical problem in hospital?	YES…………………………………1 NO……………………………………...2	
Part III. Questionnaire related to risky socio cultural and behavioral factors			
301	Have you ever-experienced abortion?	YES……………………………………1 NO……………………………….....2 No response …………………………....3	→ **303** → **303**
302	If yes in question **301**How many times in number	_____	
303	Have you ever had tattoo on your body	YES……………………………………1 NO…………………………………..2	
304	Have you ever had ear piercing	YES…………………………………1 NO…………………………………...2	
305	Have you ever had nose piercing	YES…………………………………1 NO…………………………………...2	
306	Does your husband have another wife?	YES…………………………………1 NO……………………………….....2	→ **308**
307	If yes on question **306** how many?	One ………………………………………1 Two…………………………………..2 More than two……………………………3	
308	Have you had official marriage before current husband?	YES……………………………………1 NO…………………………………..…2	

Annex2. Translated questionnaire (Somali Version)

Awaamirta uu raacayo xog ururiyuhu:

Waraysiga hooyada uurka leh ee timi xanaanada, gobaabin gali jawaabta uu bixiyo la waraystuhu ee su,aalaha doorashooyinka leh , kuwa jawaabaha gaabana ku buuxi barta loogu talo galay.

Code N<u>o</u> _____	HBV serostatus_____	
Qaynta-1aad. su'aalo ku saabsan xaalada dhaqan dhaqaale ee la waraystaha		Ugudub su'aasha
Lam	Su'aasha	Nooca jawaabta koodhka la siiyay	
100	Taariikhda la waraystay _____		
101	*Da,adaadu waa imisa?*	_____Sano	
102	Sinjiyad	Somali ……………………………………1 Oromo ……………………….…….…..2 Axmaaro…………………………………3 Uraago…………………………………..4 Tigre ……………………………………5 Qabiilo kale………………………………6	
103	Waa maxay xaaladaada lamaane ama guur	Kali noolasho……………..……………..1 Wada noolaasho……………..…………...2 Guursatay ……………………...…………..3 Kala tagay/isfuray …………………..……4	
104	*Waa maxay heerka aqaoontaadu* ?	Mana qoro mana akhriyo……....................1 Qorikara akhriyi karana (informal)………..…2 Dugsi Hoose (1-6)………………………….3 Dugsi Dhexe (7-8)………………………….4 Dugai Sare (9-12)………………………….5 Heer Jaamacadeed……………………….....6	
105	Waa maxay shaqada hada/wakhti xaadirkan aad ka shaqaysid?	Xirfadle caafimaad…………………………1 Baayac mustar ……………………………2 Guri jog ……………………...…………....3 Camal la'aan………………………………4 Shaqaali caadi ah…………………………5 *Wax intaa ka baxsa_____*	

106	Waa imisa dhaqaalaha bishii ku soo gala?ETB	Lacag ahaan (Birr)/Bishii _____ '	
107	Hada ka hor wax uur ma yeelatay?	Haa …………………………………...1 Maya …………………………………2_____	**201**
108	Hadii jawaabta su'asha **107** ay tahay Haa,imisajeer ayaad uur qaaday? **Qor tirada**	_____	
109	Imisa nolol ku soo dhalatay?	Tiro _____	
Qaynta-2aad . Su,aalaha la xidhiidha jiibsi cusbitaal.			
201	Lixdii bilood ee ugu danbaysay ma lagu dhigay/jiifiyay cusbitaal?	YES…………………………………..1 NO…………………………………..2_____	**204**
202	Waligaa malagugu sameeyay qaliin ilkaha ah cusbitaalka/kiliniga dhexdiisa?	Haa…………………………………...1 Maya………………………………….2	
203	Waligaa malagugu sameeyay qaliin jidhka ah oo cusbitaalka dhexdiisa ah?	Haa…………………………………...1 Maya………………………….………2	
Qaynta-3aad. Su'aalaha la xidhhiidha ku dhaqanka dabeecadaha iyo dhaqan bulsheed halista ah.			
301	Waliga ilmo ma kaa daatay/dhicisay?	Haa……………………….………..1 Maya……………………………....2_____ **303** Jawaab male ……………………….\3 **303**	
302	Hadii jawaabta su'asha **301** ay tahay Haa,Imisa jeer ayuu kaa daatay?	_____	
303	Waligaa jidhkaaga ma tuntay?	Haa……………………………..1 Maya…………………………….2	

304	Waligaa dhagaha ma duleeshatay	Haa………………….…………………1 Maya……………………………….…..2	
305	Waligaa Sanka ma duleeshatay	Haa…………………..……………..…1 Maya……………………………….…..2	
306	Ninkaagu maleeyahay xaas kale?	Haa…………………………………..1 Maya…………………………………..2	**308**
307	Hadii jawaabta su'asha **306** ay tahay Haa waa imisa dumara	Hal……………………………….……1 Laba……………………………….…....2 Laba iyo wax kabadan……………………3	
308	Hada ka horn in kale ma guursatay	Haa………………….……………..……1 Maya……………………………….…..2	