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ADDIS ABABA UNIVERSITY COLLEGE OF HEALTH SCIENCE
DEPARTMENT OF MICROBIOLOGY, IMMUNOLOGY AND PARASITOLOGY

SERO-PREVALENCE AND RISK FACTORS OF HEPATITIS B AND C VIRUSES AMONG HEALTHY ADULT BLOOD DONORS IN WOLITA ZONE, ETHIOPIA

BY

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Finally my diction is too poor to translate my appreciation to my wife for her support and without whom I was not able to move footstep.

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Acronyms

AAU: Addis Ababa University
ALT: Alanine Aminotransferase
ANC: Antenatal Care
Anti-HBcAg: Antibody for Hepatitis B core Antigen
Anti-HBsAg: Antibody for Hepatitis B surface antigen
Anti-HCV: Antibody for Hepatitis C virus
BBVs: Blood Borne Viruses
CDC: Center for Disease Prevention and Control
DNA: Deoxyribonucleic Acid
ELISA: Enzyme Linked Immunosorbent Assay
EPPs: Exposure-prone Procedures
ERCS Ethiopian Red Cross Society
HBcAg: Hepatitis B core Aantigen
HBeAg: Hepatitis B e Antigen
HBsAg: Hepatitis B surface Antigen
HBV: Hepatitis B Virus
HCC: Hepatocellular Carcinoma
HCV: Hepatitis C Virus
HIV: Human Immunodeficiency Virus
IgG: Immunoglobulin G
IgM: Immunoglobulin M
mRNA: messenger RNA
NANB: Non- A Non B hepatitis
NGO Non Governmental Organization
NSI: Needle Stick Injury
OR: Odd Ratio
PEP: Post-Exposure Prophylaxis
PI: Percutaneous Injury
PTH: Post-transfusion hepatitis
RIBA: Recombinant Immunoblotting Assay
RNA: Ribonucleic Acid
SNNPR: Southern Nations Nationalities Peoples Region
SOPs: Standard Operating Procedures
TTIs: Transfusion-Transmissible Infections
USA: United State of America
WHO: World Health Organization

Abstract

Background: Hepatitis B virus (HBV) and hepatitis C virus (HCV) remains a major global health problem. More than three-quarters of HBV infections occur in Asia, the Middle East and Africa. HCV causes an acute and necroinflamatory disease of liver. Blood serves as a vehicle for transmission of blood-borne pathogens including hepatitis viruses. Determination of the prevalence of HBV and HCV in a population in general, and blood-donors in particular will certainly help in reviewing the screening procedures and making health policy decisions.

Objective: The study aims at investigating the seroprevalence of HBV, HCV and associated risk factors among healthy adult blood donor in Wolaita Sodo Hospital, and Christian hospital, Wolaita Zone, Ethiopia.

Method: A cross-sectional study was conducted to determine the seroprevalence of HBV and HCV infections among blood donors at Wolaita Sodo Hospital, and Christian hospital, Wolaita Zone, Ethiopia. The blood samples were screened for HBV and HCV. All serum samples were tested for HBsAg and anti-HCV using rapid screening kit method according to the manufacturer's instruction. The socio-demographic characteristics and associated risk factors of blood donors were assessed using structured questionnaire.

Results: A total of 148 blood donors were tested .The mean age of study participants was 28.8 years ± 5.8.The overall prevalence of HBV and HCV were 10.1% (15/148) and 8.8% (13/148) respectively. The risk factors associated with significant association was observed in sharp injury and teeth extraction for HBsAg (p=0.036 (OR=4.06) and p=0.035 (OR=3.36)).

Conclusion: Screening blood donors for both HBC and HCV is indispensable for safe blood transfusion. It is important to screen donated blood with highly sensitive and specific tests and to counsel donors who are positive to any of the above infections. It is absolutely necessary to avoid the further transmission of infection.

Key words: Hepatitis B virus, Hepatitis C virus, Prevalence, Blood donors.

1. Introduction

1.1. Background

During World War II and the immediate post war period the demand for blood and blood components in the United States of America (USA) increased substantially. This resulted in the establishment and growth of blood banks, transfusion services and other blood and laboratory services. Post-transfusion hepatitis (PTH) was first reported in the USA by Beeson in 1943.Beeson commented that the expanding number of blood and plasma transfusion could lead to the occurrence of a considerable number of PTH cases (Tobler and Busch, 1997). By 1971, greater than 5400 organizations were involved in the field of transfusion medicine. Each year in the USA, 4 million patients' gets transfusion of greater than 20 million units of whole blood and blood components. Concerns about the high rate of PTH lead to the development of the National Blood Policy in 1973 and the move from paid donors to an all-volunteers system of whole blood donation (Chamberlan, 2002).

The national blood bank had established in 1969, the Red Cross National Blood Bank (NBB) is a unique public service that has been operating for the last 4 decades in the country.
ERCS has one Central Blood Bank in Addis Ababa and 11 Blood Bank branches in the country. Annually 42,000 units of blood on average are collected, screened and distributed to health institutions. The NBB contribute close to 50% of safe blood demand of the country. Club 25 is also providing to be of value during periods of seasonal shortage. 59% of blood collection comes from replacement while 41% comes from voluntary donations (ERCS, 2010).

Hepatitis B and C are major disease affecting mankind and a serious global public Health problem. According to WHO studies, out of the 2 billion people who have been infected with the hepatitis B virus (HBV), more than 350 million have chronic (lifelong) infection. These chronically infected persons are at high risk of death from cirrhosis of the liver and liver cancer. In case of Hepatitis C virus (HCV) infection WHO estimates that about 170 million people, 3% of the world's population are infected with HCV and 50% of all cases become chronic carriers at risk of liver cirrhosis and liver cancer. The prevalence of HCV infection in some countries of

Africa, the eastern Mediterranean, south-east Asia and the western pacific is high compared to countries in North America and Europe [WHO, 2000].

Hepatitis B is contracted through the blood, semen, vaginal fluids, and other body fluids of an infected individual having hepatitis B infection. Individuals at risk are intravenous drug users, children of mothers with HBV, men who have sex with men, patients on hemodialysis and those exposed to blood or blood products [Seegar and Mason, 1988]. HCV however, can only be contracted through blood to blood contact. The transmission risk of these diseases is more among patients receiving blood transfusions or injection drug users [Maheshwari and Thuluvath, 2010].

Unfortunately, once inflicted, these infections show poor response to the available treatment modalities. Therefore precautionary methods are considered the best way to avoid spreading of this disease. Unlike HCV, several vaccines have been developed for HBV that provide long lasting immunity to individuals [Vandamme and Van Herck, 2007]. It is the most important precautionary measure of HBV as a vaccinated individual may never contract the infection. Both infections, especially the risk of HCV, can be further avoided by use of disposable syringes, screened blood transfusion, avoidance of sexual abuse, antiseptic shaving and use of proper antiseptic measures in hospitals, clinics and operation theaters [Kevorkyan et al., 2011]. Safe blood and blood products should be offered to all patients in need for blood transfusion. Therefore we were interested to determine the prevalence of HBV and HCV among Health adult blood donors.

1.2. Statement of the problem

Both HBV and HCV are major cause of dreadful liver diseases including acute hepatitis, chronic liver diseases and hepatocellular carcinoma (HCC). It is estimated that 30% of the global population (about 2 billion persons) have serologic evidence of HBV infection while over 350 million people are carriers of chronic HBV worldwide [Singhal et al.]. Approximately 3% of the world's population (approximately 170 million people) have chronic HCV infection and 3-4 million people are newly infected annually [Mukhopadhya, 2008].

In Ethiopia as in other Sub-Saharan Africa, the prevalence of liver disease is high. They account for 12% of the hospital admissions and 31% of the mortality in medical wards of Ethiopian Hospitals [Tsega, 2000]. In an earlier study done to define the mode of transmission of Hepatitis B infection in Ethiopia, 5% of pregnant women were reported to be positive for HBsAg [Tsega , et al.1988]. A study among Ethiopian women attending antenatal care (ANC) in Addis Ababa hospitals, the prevalence of HBsAg was 5% [Duncan et al., 1995].
Blood and blood products are the main routes through which the virus is transmitted. Only a very small amount of blood is needed for transmission (down to 0.00004 ml intradermally). The risk for transfusion-associated HBV infection has been greatly reduced since the screening of blood for HBV markers and the exclusion of donors who engage in high-risk activities, the transmission is still possible when the blood donors are asymptomatic carrier with HBsAg negative (Lau , 1993).

In Ethiopia only few community-based studies on sero-epidemiology of HBV and HCV prevalence have been previously done and indicated that HBV and HCV are endemic in Ethiopia with regional variation .However, in Wolaita zone institution based studies are not conducted about the prevalence of HBV and HCV among health adult blood donors. The current study measured the level of HBV, HCV and risk factors among the blood donors at Wolaita zone.

1.3. Significance of the study

Blood donors were volunteers, unpaid and in many cases were relatives or friends of patients who were having medical or surgical treatment. All the donors were screened thoroughly based on the history, physical and hematological examinations before donating blood. Blood donation saves millions of lives; however, although blood transfusion plays an important role in the supportive care of medical and surgical patients, unsafe transfusion practices also put millions of people at risk of transfusion-transmissible infections. This study has the following Significance

- It will provide information on prevalence of HBV and HCV infection on the community.
- It will give clues for means of preventions.
- It can be used as base line information to those who are interested for further study in the same area.
- It can be used by those who are responsible for planning health development activity.

2. Literature review

2.1. Microbiology and Pathogenesis

Hepatitis B virus (HBV) is a relatively small, partially double-stranded,

Enveloped DNA virus and a member of the Hepadnaviridae (**hepa** = liver, **dna** = deoxyribonucleotide, **viridae** = virus) family of hepatotropic viruses, that causes **hepatitis.** Generally, the term *hepatitis* describes any inflammation of the liver, caused by a virus or a toxin such as alcohol, and characterized by jaundice, liver enlargement, and fever. Today, hepatitis B remains a major global concern. The portion of the world's population currently infected with the virus is estimated at 3 to 6 %, far greater than 0.6 % of the world infected with HIV[Murray et al.2005].

Hepatitis B virus genome (genetic make-up) is unique in that the minus-strand is full length, while the plus-strand is less than full length, leaving 15% to 50% of the molecule single stranded or uneven. Its DNA is 3020 to 3320 nucleotides long (for the full length strand) and 1700-2800 long (for the shorter strand). Its genomic circularity is maintained by 5' (five prime) cohesive ends linked to viral DNA polymerase [Murray et al.2005].

Hepadnaviridae viruses are characterized by their unique mode of replication, which involves using an RNA template for DNA synthesis even though it is a double-stranded DNA virus. The DNA is housed within an isometric nucleocapsid that is surrounded by an envelope 42 to 47 nm in diameter, made of host lipid membrane. This spherical envelope sometimes extends into a tubular tail of filamentous particles. HBV is also one of the smallest viruses to infect humans [Murray et al.2005].

HCV is a positive-stranded RNA virus, classified as family Flaviviridae, genus *Hepacivirus.* Various viruses can be differentiated by RNA sequence analysis into at least six major genotypes (clades) and more than 100 subtypes. Clades differ from each other by 25–35% at the nucleotide level; subtypes differ from each other by 15–25%. The genome is 9.4 kb in size and encodes a

core protein, two envelope glycoproteins, and several nonstructural proteins. Most cases of post transfusion NANB hepatitis were caused by HCV [Jawetz et al. 2007].

There are six major groups of variants (clades), which differ in their worldwide distribution [Brian C, et al., 2004]. Although, it is much easier to talk of the hepatitis as a single organism, it is group of viruses, similar enough to be called HCV, yet different enough to be classified into sub groups. Several identifiable 'families' of HCV have been observed around the world, differing slightly from each other in their DNA sequencing (genetic makeup). The most commonly used classification system lists these families' as HCV genotype 1, 2, 3, etc. There are multiple sub types described as HCV sub type 1a, 1b, 2a, 2b, etc [Jefferey et al., 2003].

Pathogenesis

Shortly after the HBV enters a new host, it initially infects liver cells, called hepatocytes. The virus' main target is the liver because the virus possesses surface antigens specific for receptors found on liver cells only. The binding of these viral antigens to hepatocyte receptors induces viral entry by receptor-mediated endocytosis and uncoats in the cytoplasm. Generally, the liver is responsible for purifying blood and processing nutrients. A healthy liver is essential to the functioning of blood, lymph, and bile production. If the liver fails, all other organs in the body will soon start to fail [Murray et al.2005].

Within the cytoplasm, the core particle of the virion translocates its content of viral DNA and DNA polymerase into the hepatocyte nucleus. The DNA is then organized to form a viral mini-chromosome. Once within the cell nucleus, the hepatitis B genome is transcribed into messenger RNA (mRNA), where it is subsequently translated into hepatitis B viral surface proteins, viral core proteins, DNA polymerase, and hepatitis B e-antigen protein. The cell then assembles live copies of the virus. The copies of the virus are released from the liver cell membrane into the bloodstream and from there can infect other liver cells and thus replicate effectively [Murray et al. 2005].

HCV infects only humans and chimpanzees. Patients infected with HCV have an 80 to 85 percent chance that the HCV infection will persist and that they will go on to have a chronic HCV infection. Chronic HCV infection has multiple manifestations, the most common of which

is chronic progressive liver disease associated with inflammation and fibrosis that is some patients may progress to cirrhosis. Cirrhosis or end–stage liver disease is associated with multiple complications including visceral bleeding, ascites, encephalopathy, and HCC. These conditions are common causes of morbidity and mortality in HCV infected patients [Weinstock, 1999].

2.2. Epidemiology of the Hepatitis

The global epidemiology of HBV infection has been described according to three categories of endemicity as high, intermediate, and low depending on the proportion of the population that is seropositive for HBsAg. HBsAg seroprevalence has marked geographic variations, and the degree of HBV endemicity often correlates with the predominant mode of transmission. In highly endemic settings, perinatal and horizontal (exposure to chronically infected household members) routes are responsible for most disease transmission, and 70–90 % of the adult population has serologic evidence of prior infection [Alter et al., 1999].

Africa, infections with HBV play a major role in the etiology of most liver diseases. By country, estimated HBsAg seroprevalence ranges between 5% and 19%, and the total number of carriers may approach 58 million with as many as 12.5 million likely to die prematurely due to hepatitis B-induced liver disease[Brian C, et al., 2004]. Southern Europe, the Middle East, and South Asia have an intermediate level of HBV endemicity. HBsAg seroprevalence in India is approximately 5 %, and the major modes of HBV transmission are perinatal, child-related/ horizontal, and health-care-related, particularly unsafe injections but highly endemic in Brazil and Peru with observed HBsAg seroprevalence rates greater than 10 % [Kurien et al., 2005].

In Ethiopia, a nationwide sero-epidemiological study of hepatitis B markers prevalence was conducted in Ethiopia on 5,270 young males from all regions of the country. Overall prevalence rates were 10.8% for HBsAg and 73.3% for "at least one marker positive"; a remarkable geographical and ethnic variability of marker prevalence was observed, reflecting the wide differences existing in Ethiopia in socio-cultural environment and activities such as tribal practices and traditional surgery [Kefenie et al., 1988]. A community based seroprevalence study

in the capital city of Ethiopia; Addis Ababa has shown a 7% seroprevalence of HBsAg, higher in males than females. Overall HBV seroprevalence rose steadily to over 70% in 49 year olds [Abebe, 2003].

HCV has emerged as the cause of the second major epidemic of viral infection after HIV infection within the past two decades. In the USA, an estimated 4,000,000 people are infected with HCV. There are currently about 30,000 new cases of hepatitis C infections in each year. HCV infection occurs among persons of all ages with the highest incidence of acute hepatitis C (new cases) occurring highest among groups with specific risk factors especially injection drug users, patients with hemophilic or on long term hemodialysis, prisoners and people who receive blood /blood products prior to June 1992 [Soriano et al.,1999].

The risk of occupational exposure for health care workers has been estimated to be 1.8% per incident of hollow-bore needle stick exposure to HCV infected blood. Prenatal transmission is estimated as being about 5% although if the mother is co-infected with HIV the risk may be increased to approximately 15-25 % [Chin J, 2000]. Epidemiological studies have suggested that household contact with an infected person may be associated with non sexual transmission of HCV. Seroprevalence studies have found an average anti HCV rate of 4% (0 to 11%) among house hold contacts with no other apparent risk factors for infection [Bjoro et al.1994].

Furthermore, HCV is especially prevalent in southern Italy, Spain, Central Europe, Japan and parts of Middle East (for instance almost 20% of Egyptian blood donors are HCV positive), and high prevalence rates are found in the Middle East. Infection rates of 2.5%- 10% occur in parts of South America and Asia. The prevalence of HCV seropositivity in various populations of Africa ranges from 0.2-40%. In Egypt and Cameroon, seropositvity rates of 30-40 % have been reported [Murray et al.,2005].

A survey of antibodies to HCV was measured in Ethiopian subjects representing urban and rural populations. The overall confirmed seroprevalence was 2%. Less than 1% was confirmed to be seropositive in rural communities with 1.4% positive among blood donors, 1.6% positive among

patients with dermatological disorders, 3.6% among leprosy patients, and 6.0% among patients attending a university hospital clinic for neurological disorders [Tsega et al., 1992].

Moreover, the prevalence and possible etiological association of HCV with chronic liver disease and hepatocellular carcinoma and antibodies to HCV were determined by ELISA and recombinant immunoblotting assay (RIBA) in 500 (1.4%) healthy volunteer blood donors, 14 (21%) patients with chronic hepatitis, 156 (36%) cirrhosis and 68 (46%) cases of hepatocellular carcinoma in Ethiopia. There were no apparent risk factors to suggest the mode of transmission of HCV. HCV infection was significantly more common in patients with chronic liver disease and hepatocellular carcinoma than HBV infection in this population [Frommel et al., 1993].

2.3. Modes of Transmission

a) HBV

HBV is spread through contact with infected blood and body fluids. Blood is the most important vehicle for transmission, but other body fluids have also been implicated, including semen and saliva. In most of sub-Saharan Africa, early life horizontal transmission is thought to be the predominant mode of transmission. Groups at increased risk for HBV infection includes persons with a history of sexually transmitted disease, household contacts of HBV infected persons, health care workers, hemodialysis patients, intravenous drug users, infants born to HBV-infected mothers, immigrants and children of immigrants from hyper endemic areas, homosexuals, persons who have more than one sexual partner in a six-month period, sexual partners of HBV-infected persons (Kirchner and Lin, 2004). Currently, three important modes of HBV transmission have been recognized: perinatal, sexual and parenteral/percutaneous transmission of HBV (Ellett and Marsha, 1999). In Southeast Asia, China, and sub-Saharan Africa, HBV infection usually is acquired prenatally or in early childhood. In contrast, 80 percent of infections in the United States, Canada, and Western Europe occur in adults via sexual contact or intravenous drug use (Atkins and Nolan, 2005).

Perinatal transmission is important mode of transmission in areas of intermediate and high prevalence (Schweitzer, 1975). Before HBV vaccine was integrated into the routine immunization program, the proportion of babies that become HBV carriers is about 10-30% for mothers who are HBsAg-positive but HBeAg-negative. However, the incidence of perinatal

infection is even greater, around 70-90%, when the mother is both HBsAg-positive and HBeAg-positive. The rate of infection depends up on the maternal status of HBeAg / anti-HBe (Chakarvarti et al., 2005). In one transmission study of HBV infection in Ethiopia, 19 of the 25 HBsAg positive mothers had anti-HBe none had HBeAg. There was only one case of vertical transmission, during the follow up period, new horizontal infection occurs in 2 infants and 2 older siblings, demonstrating the lesser importance of prenatal transmission (Tsega et al., 1989).

Sexual transmission of hepatitis B is a major source of infection in all areas of the world, especially in the low endemic areas, such as North America (Lavanchy, 2004). HBV is very efficiently transmitted sexually during heterosexual and homosexual contact. Heterosexual transmission can still be important as shown by the 40% transmission rates to non-immune partners of patients with acute or chronic hepatitis B (Van Damme et al., 1995).

Parenteral/percutaneous transmission can occur during surgery, after needle-stick injuries, intravenous drug use, and following procedures such as ear piercing, tattooing, acupuncture, circumcision and scarification. The nosocomial spread of HBV infection in the hospital, particularly in dialysis units, as well as in dental units, has been well described, even when infection control practices are followed. As with other modes of transmission, high vial titers have been related to an increased risk of transmission. People at high-risk of infection include those requiring frequent transfusions or hemodialysis, physicians, dentists, nurses and other healthcare workers, laboratory technicians, intravenous drug users, police, firemen, laundry workers and others who are likely to come into contact with potentially infected blood and blood products (Margolis et al., 1991). In one study of hepatitis B transmission no clear risk factor is found in 20-30% of patients (Lee, 1997).

b) HCV

HCV is chiefly contracted through parenteral exposure to infected material such as blood-transfusion, or injection with dirty needles. Risk factors for HCV infection include: drug injection, history of hepatitis, health care workers who are at risk for needle sticks and other exposures history of previous surgical procedure and blood transfusion (Zaller, et al. 2004). Relatively weak risks associated are sex with an infected partner, high-risk sexual behavior, and low socioeconomic status. The risk of vertical transmission is 5%. A mother's co-infection with HIV significantly increases this risk (Shehab, 2004). The accumulated evidence indicates that hepatitis C virus (HCV) can be transmitted by sexual contact but much less efficiently than other

sexually transmitted viruses, including hepatitis B virus and human immunodeficiency virus (HIV). However, because sex is such a common behavior and the reservoir of HCV-infected individuals is sizable, sexual transmission of HCV likely contributes to the total burden of infection. HIV confections appear to increase the rate of HCV transmission by sexual contact (Terrault, 2002).

2.4. Clinical Manifestations

Hepatitis B is a serious infection that affects the liver. It can cause acute (short-term) and Chronic (long-term) infection illness. Acute infections can lead to loss of appetite, diarrhea and vomiting, tiredness, jaundice (yellow skin or eyes), pain in muscles, joints, and stomach acute illness, with symptoms, is more common among adults. Children who become infected usually do not have symptoms. Chronic (long-term) infection: Some people go on to develop chronic hepatitis B infection. Most of them do not have symptoms, but the infection is still very serious, and can lead to liver damage (cirrhosis), liver cancer, and death. Chronic infection is more common among infants and children than among adults. People who are chronically infected can spread hepatitis B virus to others, even if they don't look or feel sick [Murray et al.,2005].

HCV infection causes an acute or chronic necroinflammatory disease of the liver. Only relatively small fractions of HCV infections are symptomatic. Most infected individuals remain asymptomatic and presumably, undiagnosed [Alter et al., 1999]. Major clinical manifestation is progressive hepatic fibrosis, which leads to cirrhosis and an increased risk of hepatocellular carcinoma [Frommel et al., 1993]. Primary infection with HCV leads to persistent viremia in approximately 85% of patients with development of chronic liver disease in >60% of cases. Approximately 20% of individuals with chronic hepatitis C eventually develop medically significant sequel including cirrhosis end stage liver disease or HCC [Feinstone et al., 1975].

2.5. Laboratory Diagnosis

When a person is infected with HBV, the first virological marker detectable in the serum is HBsAg. It precedes the elevation of serum aminotransferase and clinical symptoms. In a majority of cases, HBsAg becomes undetectable one to two months after the onset of jaundice and rarely persists beyond six months. During the recovery phase, HBsAg becomes undetectable,

while antibodies to HBsAg (Anti-HBs) become detectable in the serum and remain so indefinitely thereafter. In addition, anti-HBs antibody is the only detectable serological marker in those who successfully respond to hepatitis B immunization. Hepatitis B core antigen (HBcAg) is an intracellular antigen that is not detectable in serum. Antibodies against HBcAg (anti-HBc), indicate a prior exposure to HBV, irrespective of the current HBsAg status. IgM anti-HBc is the first antibody detectable in an acute HBV infection. Usually it becomes detectable within one month after the appearance of HBsAg and disappears within six months. IgG anti-HBc is not a neutralizing antibody and remains detectable throughout the patient's life [Wolfgang et al., 2000].

The first generation HCV- antibody ELISA test commercially available in 1990 and was widely used. As more reactive recombinant antigens were identified from conserved regions of the HCV genome, newer serologic assays (second and third generation) were introduced with improved sensitivity and specificity. Compared to this, first generation HCV ELISA using single recombinant antigen, multiple antigens using recombinant protein and / or synthetic peptides have been added in new serologic tests to avoid non- specific cross- reactivity and to increase the sensitivity of the HCV antibody tests [Kao, 2008].

Two major types of tests are available for the lab diagnosis of HCV infection: detection of antibody to various HCV antigens and molecular methods to detect and quantitate the nucleic acid of the virus. HCV antibody testing is analogous to antibody testing for human HIV infection. The detection of HCV antibodies (anti-HCV) proves exposure to HCV, but does not distinguish between resolved and chronic hepatitis C. The ELISA test also cannot determine whether or not someone is a carrier or if disease has run its course and the antigen is no longer present [Gretch et al., 1992].

2.6. Antiviral therapy

Major advances have been made in the treatment of chronic HBV infection during the past several years

At the present time, three drugs are licensed for the treatment of HBV infection: interferon alfa, lamivudine, and the nucleotide analogue adefovir dipivoxil.

Several newer nucleoside analogues, such as entecavir, emtricitabine, and telbivudine, are in various phases of study. Adefovir, tenofovir, and entecavir have been shown to have antiviral activity against lamivudine resistant as well as wild-type HBV both in vitro and in vivo (Robert, 2004)

Although HBV infection can be prevented by vaccination, it is important to treat persons with CHB at high risk of progression to reduce the considerable morbidity associated with CHB.

Currently, seven antiviral agents lamivudine, adefovir, entecavir, telbivudine, tenofovir, emtricitabine, standard and PEG-IFN are approved for the treatment of CHB in high-income countries, and have been shown to delay the progression of cirrhosis, reduce the incidence of HCC and improve long-term survival (WHO, 2015)

HCV is a now a curable disease, and advances in HCV therapy have resulted in steadily higher cure rates

At the time of writing (December 2013), six drugs are licensed for the treatment of HCV –

1.standard interferon (IFN) or pegylated interferon alpha (PEG-IFN),

2. Ribavirin (RBV),

3. The protease inhibitors (PIs) boceprevir,

4. Simeprevir

5. Telaprevir,

6. The nucleotide analog polymerase inhibitor sofosbuvir. (WHO, 2014)

2.7. Prevention and Control

Adherence to universal or standard precautions remains the primary means of preventing

Occupational exposures and thus of reducing occupational risk of acquiring infection with blood

Borne pathogens. CDC recommends that all health-care providers at risk for HBV infection be tested and that all those found to be susceptible should receive vaccine. Post exposure prophylaxis (PEP) remains the second line of defense in instances in which primary prevention fails to prevent occupational exposures [CDC,2011].

There are still many barriers to the development of an HCV vaccine. Among these challenges are the heterogeneity of decrease with isolates, the virus ability to modify envelope and other proteins rapidly in the faces of immunlogical pressure, incomplete understanding of the pathogenesis and immunogenesis of HCV infection, the likely need to develop a vaccine that

stimulates both humoral and cellular immunity against HCV, and the inability to culture the virus [Beekman.et al., 2005]. The key to reducing the incidence of HCV infection is decreasing exposure to contaminated blood. The incidence of post-transfusion HCV infection has been reduced to very low levels by screening blood donation for HCV anti body as well as surrogate makers of HCV infection, nosocomial HCV transmission in developing countries should decrease with worldwide adherence to universal precautions [Forns et al, 2002].

3. Objectives of the Study

3.1 General Objective

To determine the seroprevalence and risk factors of hepatitis B and C viruses among healthy adult blood donors from May-July 2014 at Wolaita Zone, Ethiopia

3.2 Specific Objectives

- To determine the prevalence of HBV and HCV among blood donors
- To identify associated risk factors of HBV and HCV.

4. Method and Materials

4.1. Study Design and Period

A cross-sectional study design was conducted to determine the prevalence of hepatitis B and C viruses' infection and risk factors among healthy adult blood donor in Wolaita zone from May – Jul, 2014.

4.2. Study Area

The study was conducted in Wolaita zone. It is one of the 13 zones in Southern Nations Nationalities Peoples region (SNNPR) in Ethiopia. Wolaita zone has 12 woreda and 3 town administrations. The capital city of the zone is Sodo town which is about 360 km away south to Addis Ababa. Based on the most recent census, the projected population of zone is 1, 750, 079, in 2013. By the year 2013 there are one government referral hospital; one non-government hospital, one private hospital, 66 health centers and 335 health posts providing health services for the community. There are also 2 higher clinics, 8 medium clinics, 121 lower clinics and 42 pharmacies are among the private sectors serving the community according to the Zonal Department. (Wolaita Development Association.)

4.3. Source Populations

The source population was all health adult blood donors in Wolaita zone. Based on registration book in Wolaita zone has 350 blood donors per month.from 350 blood donors 300 were males (Hospital registration book).

4.4. Study Populations and Sample Population

The study population was including all healthy adult blood donors who donate blood from May – July, 2014 at Wolaita zone hospital based blood bank.

148 blood donors were selected from all health blood donors attending in selected hospital during the study period.

4.5. Sampling Techniques and Sample Size Determination

4.5.1 Sampling Technique

The probability sampling method employed with a sample size calculated 148 bloods donors. Using systematic random sampling the calculated sample size were allocated to each bloods donors by probability proportional to size method. Using K value the interval between each sample unit identified before and sample unit (a blood donor) was selected by simple random sampling system. Sampling interval (K): is the sum of three consecutively month (May-July) of the least year (2013) divided to desired sample size. Which was 920/148=6. Randomly selected number between 1and 6 was 2.

4.5.2 Sample size determination

The estimated sample size was 148 participants assuming a prevalence of HBsAg of 10.8%. [Kefenie et al., 1988] Sample size derived using the following formula:

$$N = \frac{Z^2_{\alpha/2} P Q}{d_2}$$

- Where: N = sample size; $Z_{\alpha/2}$ = standard normal distribution abscissa corresponding to 95% confidence interval (1.96); P = proportion of HBsAg reported in similar study noted above (10.8); $Q = (1-P)$; and d = desired level of precision (5%).

$$n=\frac{(1.96)^2 x 0.108(1-0.108)}{(0.05)^2} = \mathbf{148}$$

4.6. Inclusion and Exclusion Criteria

4.6.1 Inclusion Criteria:- All blood donors Weight >50kg, age 18- 6o years

4.6.2 Exclusion Criteria:- age <18 and >60 years, weight <50kg, pregnant and lactating women, current history of medication, recent history of operation, serious illeness and blood donation for 3 months prior to the current study.

4.7. Variables

4.7.1. Independent Variables

Age, sex, marital status, sexual partner, occupation category, History of sharp injury (cut), History of blood transfusion, tooth extraction, and blood transfusion profile.

4.7.2. Dependent variables

Sero-epidemiology of HBV and HCV among the blood donor

4.8. Data collections tools

Data was collected by pre-tasted questionnaire and laboratory diagnosis

4.9. Data collection methods and laboratory diagnosis

4.9.1. Data collection methods

A structured and standardized questionnaire to collect information about the exposure status of potential risk factors and to assess the socio-demographic characteristics of the study participants was used. The questionnaire was prepared in English language by principal investigator in simple understandable language. Three laboratory personnel was selected and Training was given on the objective, benefit of the study, individual's right, informed consent and techniques of the interview for one days. Before the actual data collection pursued a pre-test of the instrument and the procedure was conducted and corrective measures were taken.

4.9.2. Laboratory diagnosis

4.9.2.1Blood sample and Serological test

After written informed consent obtained, 5 ml of blood sample was collected using disposable EDTA tubes under aseptic condition. The tubes were numbered and processed at the time of collection. The blood sample taken was centrifuged for 5 minutes at speed of 3000rpm and the serum was separated. All serum samples were tested for HBsAg and anti-HCV using rapid screening method according to the manufacturer's instruction (Zhejiang Orient Gene Biotech Co., Ltd, China). The kit has a sensitivity of 98.9% and specificity of 98.57%.

HBsAg and anti-HCV: One step tests, rapid, self performing, qualitative, immunochromatographic techniques that detect presence of HBsAg and anti-HCV in serum/plasma specimens, qualitatively, at concentrations as low as 0.5 ng/ ml.

Principles

a) **For HBsAg:** one step test that utilizes the principle of immuno-chromathography, a unique two site immunoassay on a membrane. As the test sample flows through the membrane assembly of the dipstick, the colored monoclonal anti-HBsAg colloidal gold conjugate complexes with HBsAg in the sample. This complex moves further on the membrane to the test region where it is immobilized by another monoclonal anti-HBsAg antiserum coated on the membrane leading to formation of a pink-purple colored band which confirms a positive test result. Absence of this colored band in the test region indicates a negative test result. The unreacted conjugate and unbound complex, if any moves further on the membrane and are subsequently immobilized by the anti-rabbit antibodies coated on the membrane at the control region, forming a pink/purple colored band. This control band serves to validate the test result.

b) **For anti-HCV:** one step test that utilizes the principle of immuno-chromathography, a unique two site immunoassay on a membrane. As the test sample flows through the membrane assembly of the dipstick, the colored HCV antigen colloidal gold conjugate complexes with anti-HCV in the sample. This complex moves further on the membrane to the test region where it is immobilized by another HCV antigen coated on the membrane leading to formation of a pink-purple colored band which confirms a positive test result. Absence of this colored band in the test region indicates a negative test result. The unreacted conjugate and unbound complexes, if any moves further on the membrane and are subsequently immobilized by the anti-HCV coated on the membrane at the control region, forming a pink/purple colored band. This control band serves to validate the test result.

4.10. Data quality control

The quality of data was controlled starting from the time of questionnaires preparations. The questionnaires were developed by reviewing relevant literatures on the subject to ensure reliability. Training was given for supervisors and data collectors on the purpose of study and procedures of data collection for 1 day prior to study. After completing the training, trainees was be conducted a pre-test at non study health facility. Finally, we were discussed on problem they encountered during pre-test and corrective measures were taken. During data collection, the

supervisor was received questionnaires from data collectors and review for completeness, accuracy, and consistency. Correction measures were taken by discussing with the research team. Standard operating procedure (SOPs) was followed during laboratory analysis and known positive and negative sera for HBV and HCV results were reported.

4.11. Data management and Analysis

After the completion of the data collection, the questionnaires were checked for its completeness, unrecorded values and unlikely responses and then manually clean up on such indication. The test result was written on the laboratory data collection format sheet. Means and frequencies (%) was used to describe participant's characteristics. Univariate and multivariate logistic regression was used to determine the risk factor of HBV and HCV infection. Significance level and association of variables will be tested by using 95% confidence interval (C.I) and odd ratio. P-value less than 0.05 were taken as statistically significant. All analyses were performed using the epinfo-data version 3.1 for data entry and SPSS version 21 programs for data analysis.

4.12. Ethical Considerations

Ethical clearance was obtained from AAU, Department of Microbiology, Parasitology and Immunology ethical review and research committee. Permission was obtained from Wolaita zone Hospital where the samples were collected.

5. RESULTS

5.1 Socio-demographic Characteristics

In this study a total of 148 blood donors were participated and the socio-demographic characteristics indicated in table 1. Out of 148 participants, 130 (87.8 %) were males. With the highest age group, between 25 to31 83(56.1%) years followed by 18 to24 years of age 28(18.9 %).

Table 1: Socio-Demographic Characteristics of study participants at Sodo Hospital and Christina Hospital based blood bank, May-Jun 2014

Variables	Number	Percent
Sex		
Male	130	87.8
Female	18	12.2
Age		
18-24	28	18.9
25-31	83	56.1
32-38	26	17.6
39-45	9	6.0
>46	2	1.4
Marital status		
Single	90	60.8
Married	54	36.5
Divorced	4	2.7
Religion		
Christian	141	95.3
Muslim	4	2.7
Other	3	2.0

Occupation		
Daily laborer	28	18.9
Farmer	21	14.2
Merchant	29	19.6
Student	45	30.4
Other	25	16.9
Total	148	100

5.2 HBV and HCV Sero-prevalence

The prevalence of HBsAg and HCV anti-body among the blood donors in the hospital based blood bank was 10.1 %(15/148) and 8.8 %(13/148) respectively. Of these, 10 %(13/130) of HBsAg and 8.5 %(11/130) HCV anti-body positive blood donors were males. Although the difference was not statistically significant, p=0.88 (OR=0.89) a high prevalence rate was observed among females than males. In the age category of 25-31, 12 %(10/83) of blood donors were positive for HBsAg, 9.6 %(8/83) were positive for HCV anti-body and in the age group 18-24, 7.1 %(2/28) were positive for HBsAg and 14.3 %(4/28) were HCV anti-body positive(Table 2).

Table 2: Distribution of HBV and HCV prevalence among sex and different age groups of blood donors in Wolaita soddo, Christian Hospital based blood bank, May-Jul 2014

Variables	HBV		OR(95%CI)	HCV		OR(95%CI)
	Positive NO (%)	Negative NO (%)		Positive NO (%)	Negative NO (%)	
Sex						
Male	13(10)	117(90)	.89(.18-4.3)	11(8.5)	119(91.5)	.74(.15-3.6)
Female	2(11.1)	16(88.9)		2(11.1)	16(88.9)	
Age						
18-24	2(7.1)	26(92.9)		4(14.3)	24(85.7)	
25-31	10(12)	73(88)		8(9.6)	75(90.4)	
32-38	2(7.7)	24(92.3)		1(3.8)	25(96.2)	
39-45	1(11.1)	8(88.9)		0	9(100)	
> 46	0	2(100)		0	2(100)	
Total	15(10.1)	133(89.9)		13(8.8)	135(91.2)	

5.3 Marital status and occupation

married donors were more positive for HBsAg 11.1 %(6/54) compared to the Single blood donors 7.8 %(7/90) (OR=0.08) (Table 3). HCV anti-body was higher in single blood donors, 11.1 %(10/90) followed by married donors, 5.6 %(3/54).The difference was not found statistically significant, p=0.25 (OR=0.47).Regarding occupation, daily workers accounted 17.9 %(5/28), followed by merchants, 13.8% (4/29) and others, 12% (3/25) from the total HBsAg positive. HCV prevalence in students was 11.1% (5/45) followed by daily workers, 10.7 %(3/28), merchants 10.3% (3/29), farmer 9.5 %(2/21).The difference was not found statistically significant p= >0.05

Table 3: HBV and HCV prevalence among Marital status and Occupational status of blood donors in Wolaita sodo and Christian hospital based blood bank, May-Jul 2014

Variable	HBV		OR(95%CI)	HCV		OR(95%CI)
	Positive NO(%)	Negative NO(%)		Positive NO(%)	Negative NO(%)	
Marital status						
Single	7(7.8)	83(92.2)	.08(.01-.69)	10(11.1)	80(88.9)	
Married	6(11.1)	48(88.9)	.13(.2-1.9)	3(5.6)	51(94.4)	.47(.12-1.79)
Divorced	2(50)	2(50)		0	4(100)	
Occupation						
Daily laborer	5(17.9)	23(82.1)	1.6(.34-7.5)	3(10.7)	25(89.3)	1.00
Farmer	1(4.8)	20(95.2)	.77(.12-5.12)	2(9.5)	19(90.5)	.87(0.13-5.7)
Merchant	4(13.8)	25(86.2)	.85(.15-4.6)	3(10.3)	26(89.7)	.96(0.17-5.2)
Student	2(4.4)	43(95.6)	.34(0.05-2.2)	5(11.1)	40(88.9)	1.04(.23-4.7)
Others	3(12)	22(88)	1.00	0	25(100)	.000(.000)
Total	15(10.1)	133(89.9)		13(8.8)	135(91.2)	

Note Others: teacher, nurses, broker, house wife,NGO.

5.4 Risk factors

Out of the **148** blood donors 1.4%(2/148) had occupational exposure to blood,52.7%(78) had history of sharp injury, where28.2%(22/78),25.6%(20/78), 11.5%(9/78) and 34.6%(27/78) had at least one time, two time, three time and several time history of sharp injury respectively. The difference was found statistically significant to HBsAg p=0.036 (OR=4.06). Out 0f the total 148 blood donors 95.9 %(142) had first donation of blood from whom 10.5 %(15/142) were HBV positive and 9.2% (13/142) were HCV positive and 4.1 %(6) had repeated history of blood donation and none of them were HBV and HCV positive. Three donors (2%) had family history of liver disease. The difference was not found statistically significant, p= >0.05. According to teeth extraction 18.9 % (28) had history of teeth extraction. The difference was found statistically significant to HBsAg, p=0.035 (OR=3.36) (Table 4).

Table 4: HBV and HCV prevalence among exposure to risk factors of blood donors in Wolaita sodo, Christian Hospital based blood bank, May-Jul 2014

Variable	HBV		OR(95%C)	HCV		OR(95%C)
	Positive NO (%)	Negative NO (%)		Positive NO (%)	Negative NO (%)	
Occupational exposure						
NO	15(10.3)	131(89.7)		13(8.9)	133(91.1)	
Yes	0	2(100)	0.00	0	2(100)	0.00
His of sharp injury						
NO	3(4.3)	67(95.7)		6(8.6)	64(91.4)	
Yes	12(15.4)	66(84.6)	4.06(1.09-15)	7(9)	71(91)	1.05(0.34-3.3)
If yes to sharp injury						
one time	1(4.5)	21(95.6)	1.06(.1-10.7)	1(4.5)	21(95.6)	0.5(.05-4.46)
Two time	2(10)	18(90)	2.48(.38-15.9)	4(20)	16(80)	2.67(0.67-10.58)
Three time	1(11.1)	8(88.9)	2.79(.25-30.1)	1(11.1)	8(88.9)	1.33(0.14-12.53)
Several time	8(29.6)	19(70.4)	9.4(2.27-38.9)	1(3.7)	26(96.3)	0.41(.047-3.57)
Donation time						
First	15(10.6)	127(89.4)		13(9.2)	129(90.8)	
Repeated	0	6(100)		0	6(100)	
Liver disease						
No	14(9.7)	131(90.3)		12(8.3)	133(91.7)	
Yes	1(33.3)	2(66.7)	4.67(.39-54)	1(33.3)	2(66.7)	5.54(0.46-65.6)
Teeth extraction						
No	9(7.5)	111(92.5)		11(9.2)	109(90.8)	
Yes	6(21.4)	22(78.6)	3.36(1.1-10)	2(7.1)	26(92.9)	1.32(0.34-5.15)
Total	15	133		13	135	

Note His= History

5.5 The Sexual activity was compared with HBsAg and anti-HCV prevalence among blood donors. From the total of 148 blood donors 64.9% (96) had history of sexual partner in life, from whom 12.5 %(12/96) were HBV positive and 8.3% (8/96) were HCV positive. The difference was not found statistically significant OR=2.33 (Table 6)

Table 5: sexual activity versus HBV and HCV prevalence among blood donors in Wolaita sodo and Christian hospital based blood bank, May-Jul 2014

Sexual activity	HBV		OR(95%CI)	HCV		OR(95%CI)
	Positive No (%)	Negative No (%)		Positive No (%)	Negative No (%)	
sexual partner						
No	3(5.8)	49(94.2)		5(9.6)	47(90.4)	
Yes	12(12.5)	84(87.5)	2.33(0.62-8.67)	8(8.3)	88(91.7)	0.85(0.26-2.75)
	15	133		13	135	
If yes one	5(9.4)	48(90.6)	1.7(0.38-7.51)	3(5.6)	50(94.4)	0.36(0.06-1.99)
Two	1(5.9)	16(94.1)	1.02(0.9-10.51)	1(5.9)	16(94.1)	0.58(0.06-5.41)
Three	2(28.6)	5(71.4)	6.53(0.87-48.85)	1(14.3)	6(85.7)	1.56(0.15-15.76)
>Three	4(21)	15(79)	4.35(0.87-21.67)	3(15.8)	16(84.2)	2.50(0.59-10.55)
	12	84		8	88	

6. Discussion

HBV and HCV infections are common serious complications of blood transfusion. Prevention of transfusion-transmitted infections in developed countries has been achieved by reducing unnecessary transfusions, using only regular voluntary donors, excluding donors with specific risk factors and systematic screening of all donated blood for infection. By contrast, in many developing countries none of these interventions is applied uniformly and the risk of transfusion-transmitted infections remains high (Gurol et al.2006).

In this cross- sectional survey prevalence of HBV and HCV was 10.1% and 8.8% respectively.

This result comparatively higher than those previously conducted studies among Ethiopia blood donors. The result of this study was higher for HBsAg compared to the prevalence of HBsAg among blood donors in Gondar hospital of blood bank 4.7%, and Dessie hospital of blood bank 3%(Gelaw and Mengistu 2008).However, the prevalence of HBsAg was lower than compared to Mekele hospital of blood bank 14 %(Gelaw and Mengistu 2008), Felege Hiwot referral hospital 25%(Azene et al.,2007) and the 1984 Red Cross blood bank in Addis Ababa prevalence, which was 11%(Tsega ,2000).The differences were due to variations in geographical distribution as well as population differences in terms of lifestyle, awareness, sensitivity and specificity of tests.

The result of this study showed 8.8% anti-HCV prevalence among blood donors, which was lower than the prevalence of Cairo blood donors 14.6 % (Theodore and Mazen, 2006). However the prevalence of anti-HCV was higher than compared to Pakistan, blood donors 1.8%, Saudi Arabia 1.8 and Yemen blood donors, which was 2.1%.(Theodore and Mazen 2006).

The result of this study was higher than those previously done studies among blood donors in Gondar hospital of blood bank 2.33% (Gelaw and Mengistu 2008). However, the prevalence of HCV was lower than compared to Felege Hiwot referral hospital, North West Ethiopia which was 13.3 %(Azene etal., 2007). The differences were due to variations in geographical distribution as well as population differences in terms of lifestyle, awareness, sensitivity and specificity of tests.

In this study there was no co-infection by hepatitis viruses (HBV and HCV) observed.some authors found an inhibition of hepatitis B virus by hepatitis C virus (endword et al., 2001)

This is supported by other similar study conducted in Amhara and Tigray Regional States (Gelaw and Mengistu 2008)

The highest prevalence rate of HBsAg was seen in the daily-laborer 17.9 %, followed merchant 13.8% which is different from other studies conducted in Gondar and Addis Ababa blood bank. It has been documented that HBsAg prevalence was higher in farmer in Gondar 18.8% followed by soldiers 16.3% (Alkan et al.1993).The study conducted in Addis Ababa showed that drivers and mechanics accounted for higher prevalence28% followed by student 20%(Tsega 2000, Alkan et al 1993).

The anti-HCV anti-body prevalence was higher in student followed daily based workers, merchant and farmer, 11.1%, 10.7%, 10.3%, and 9.5% respectively. This is different from other studies conducted in Amhara and Tigray Regional States Which showed that the merchant accounted for higher prevalence followed by daily based workers, 15%and 8.5% respectively (Gelaw and Mengistu 2008).

When we see prevalence of hepatitis B virus with regard to sharp injury and teeth extraction accounted 15.4% and 21.4% respectively. There was a statistically significant association observed the P-value 0.036 and 0.035 respectively. The activity of sexual partners was compared with HBsAg and anti-HCV prevalence 12.5% and 8.3% respectively. The maximum number of positivity among blood donors with one sexual partner, however the finding was no statistically significant the P-value=0.206, which suggested that the sexual transmission of HBV in Ethiopia is not of much importance, vertical transmission as well as cultural practices such as traditional surgery, uvulectomy, ritual scarring etc.., are known to contribute considerably to the spread of HBV infection (Gebreselassie, 1986).

Limitation

Our study has some important limitations, majority of serological test were also considerable to the test kit used by this study. Those test kits may not completely detect donors with HCV infection at the window period. We did not perform additional laboratory tests of interest including anti-hepatitis B core antigen (anti-HBc), confirmatory testing for anti-HCV antibodies and HBsAg positive samples, and HCV-RNA was also not determined in any of the anti-HCV antibody positive patients in order to differentiate between active and resolved infection. The presence of anti-HBc antibody is a lifelong marker of HBV infection, irrespective of whether a patient has recovered from or has an ongoing chronic infection.

Conclusion & Recommendation

Blood donation saves millions of lives; however, although blood transfusion plays an important role in the supportive care of medical and surgical patients, unsafe transfusion practices also put millions of people at risk of transfusion-transmissible infections. Screening blood donors for both HBC and HCV is indispensable for safe blood transfusion. It is of utmost importance to continue screening donated blood with highly sensitive and specific tests and to counsel donors who are positive to any of the above infections.

A prevalence of 10.1% HBsAg and 8.8% anti-HCV might warrant the introduction of screening of all blood donors for hepatitis viral markers (HBV and HCV) and should be instituted in the wolaita zone as well as in the country. As it was already indicated by the work of others and this study, hepatitis virus infections are found at high prevalence rate in our country. This high prevalence rate is attributable to lack of adequate knowledge way of transmission and cultural practices such as traditional surgery, uvulectomy, ritual scarring etc.., are known to contribute considerably to the spread of HBV infection.

Since blood donors are part and parcel of the community, the problems of those blood donors reflect the problem of the community as a whole. So as to reduce the spread of HBV infection among blood donors and the community, the following activities are recommended.

-Giving health education different population groups in order to know the way of transmission, risk reduction counseling and epidemiology of the diseases.

- Providing education to avoid bad cultural practices such as traditional surgery, ritual scarring etc...

-Report the results of the tests after donation with follow-up counseling to prevent further transmission of the infection.

-Implementation of more sensitive tests (such as nucleic acid amplification testing [NAT] for HBV and HCV) that detect infection earlier (reduce the window period) will further decrease risks of transfusion-transmitted viral infections.

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ANNEXES

I. Information sheet (English Version)

We are studying the prevalence of HBV and HCV among health adult blood donors. Hepatitis virus is the etiological agent of chronic hepathopathies and that patients with this acute infection may not develop symptoms in most cases and serve as a source of infection. Early detection of the HBV and HCV important to prevents complication and transmission to others

I, the undersigned, confirm that, as I give consent to participate in the study with a clear Understanding of the objectives and conditions of the study and with recognition of my right to Withdraw from the study if I change my mind.

I……do hear by give consent to DR/Mr./Miss…….to Include me in the proposed research. I have been given the necessary information about the Research. I have also been assured that I can withdraw my consent at any time without penalty or loss of benefits .The proposal has been explained to me in the language I understand.

Name of the participant: _____

Participant's signature: _____

Date: _____

Witness: _____Date:_____

Address of the investigator

Name: Tigistu Demisse
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II. Questionnaire (English Version)

Addis Ababa University School of Health scienceDepartment of Microbiology, Immunology and Parasitology

Questionnaire for investigation of HBV and HCV viral infections in health adult blood donors at Wolaita sodo hospital and Christian hospital, Wolaita zone, Ethiopia

Direction: - Please circle on the answer among choices or write on the space provided

Serial number: _____ date of interview: _____/____/_____
DD MM YYYY

Identification		
01.	Sex	Male.....1 Female..... …..……....2
02.	Age	____ ____
03.	Marital status	Single……..……..1 Co-habiting……..…….2 Married……..…3 Divorced……...4
04.	Religion	Christian……...1 Muslim……..2 Does not have……...3 Other specify……..
05.	Occupation	----------

Risk assessment		
6.	Occupational exposure to blood	Yes……..1 No……2
7.	History of sharp injury (cut)	Yes……..1 No……2

8.	If yes how many times	Once ……1 Twice …….……2 Three times ……..……3 Several…….……4
9.	History of previous surgery	Yes……1 No……..2
10.	History of blood transfusion	Yes……1 No……..2
11.	History of blood donation	first……1 repeated…….....2
12.	History 0f parenteral/intravenous drug use	Yes……1 No……..2
13.	History of tooth extraction	Yes…….……1 No…….…….2
14.	History of sexual partner in life	Yes…….……1 No…….…….2
15.	If yes how many sexual partner in life	one…….1 two…….2 three ……..3 >four ……4
16.	Have you ever practiced needle recapping?	Never……1 Sometimes……2 Most of the time……..…3 All the time…..……4
17.	Family history of liver disease	yes……..…….….....1 No…….2
18.	Did you take vaccination for HBV?	Yes……...1 No…….....2
19.	Blood group	A …….....1 B……..…….....……2 AB…….…..3 O…….….4 Rh +ve……..…5 _ve……...6
20	Lab. Result	1…….. 2……

III. Test procedures and interpretation

1. Before sample collection introduce yourself and identify the patient
2. Take a time to wash your hands and wear gloves
3. Prepare the material required (needles, tubes, etc)
4. Prepare the patient
5. Apply the tourniquet(do not let it on for extended period)
6. Choose a vein
7. Disinfect the draw site
8. Collect serum/plasma specimen in a clean test tubes.
    - Ensure that only sufficient quantity of the specimen is collected to allow submerging the red area of the dipstick (about 1cm high).
    - Exit the vein and apply pressure
    - Discard the needle(in appropriate biohazard container)
    - Label the specimen before leaving the patient
    - Check the patient apply a bandage if necessary
    - Allow the specimen for 30 minutes to facilitate clotting
    - Centrifuge with medium speed for 5 minutes
    - Separate serum from the blood by Pasture pipette
    - Perform the lab test and store the remaining serum at -20^0c
9. Bring the sealed pouch to room temperature, open the pouch and remove the dipstick once opened, the dipstick must be used immediately.
10. Dip the dipstick in the serum/plasma specimen submerging only the red area
11. The dipstick should be left to stand in the specimen for the entire duration of the test ensuring only the red area is submerged in the specimen.
12. At the end of 15min, read the result as follows:
    - ✓ NEGATIVE: Only one colored band appears on the dipstick
    - ✓ POSITIVE: two distinct colored bands appear on the dipstick.
13. The test should be considered invalid if no band appears. Repeat the test with a new dipstick.

14. Although, depending on the concentration of HBsAg in the specimen, positive results may start appearing as early as 2min, negative results must be confirmed only at the end of 15 min.
15. In case of a doubtful result at 15min, the test may be extended up to 30min to get a clear background.

Signed Declaration

I, the undersigned declare that this thesis is my own original work and it has not been presented for a degree or some other purpose in any universities, colleges or institutions and that all sources of material used for the thesis have been dully acknowledged.

Name of principal investigator: Tigistu Demisse

Signature: _____

Date of Submission: 2015

Addis Ababa, Ethiopia

This thesis has been submitted for examination with my approval as University Advisors.

Name Advisor: Dr. Tamrat Abebe

Signature: _____

Place: Addis Ababa University

Name Advisor: Dr. Adane Mihret

Signature: _____

Place: Addis Ababa University

Name of Examiner: _____

Signature: _____

Place: Addis Ababa University

Date of Submission: _____/_____/_____