PREVALENCE OF HEPATITIS B VIRAL INFECTION AMONG PATIENTS WITH HEPATOCELLULAR CARCINOMA ATTENDING MUHIMBILI NATIONAL HOSPITAL DAR ES SALAAM TANZANIA

Kaduri Ombeni, MD

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By

Kaduri Ombeni, MD

A dissertation in (Partial) Fulfillment of the Requirements for the Degree of Masters of Medicine (Internal Medicine) of Muhimbili University of Health and Allied Sciences

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CERTIFICATION

The undersigned certify that he has read and hereby recommend for acceptance by Muhimbili University of Health and Allied science a dissertation entitled '**Prevalence of Hepatitis B virus among patients with Hepatocellular carcinoma attending Muhimbili National Hospital, Dar es salaam**' in the fulfillment of the requirements for the degree of Masters of Medicine (Internal Medicine) of the Muhimbili University of Health and Allied science.

_____

Dr. E. Komba

Supervisor

_____

Date:

DECLARATION

AND

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I **Dr. Kaduri Ombeni** declare that this **dissertation** work is my own original work and that it has not been presented and will not be presented to any other university for similar or any other degree award.

Signature……..... **Date**……

ACKNOWLEDGEMENT

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DEDICATION

This dissertation is dedicated to the memory of my Late Father, Prof. A. J. Kaduri for laying an excellent foundation right from childhood to become a good doctor.

ABSTRACT

Background: The Global burden of HBV related HCC is still a major challenge for public health in the 21st century, especially in the endemic regions due to unavailability of early diagnostic techniques and treatment. Worldwide 450 million individuals are reported to be chronic carriers of HBV, accounting for 55% of global cases of HCC 80% 0f which are found in the sub-saharan Africa. Despite, high magnitude of HBV related HCC in Africa and the world at large, no data from Tanzania is available to address magnitude of HBV among patients with HCC.

Objective: This study aimed to determine the prevalence of Hepatitis B viral infection among patients with hepatocellular carcinoma attending Muhimbili national hospital in Dar es Salaam, Tanzania.

Methods: A descriptive cross sectional study was done for a period of 9 months, June 2012 through February 2013. During this period patients attending inpatients and outpatients gastroenterology department with the diagnosis of HCC were enrolled and screened for Hepatitis B virus. The evaluation included risk factor assessment, clinical assessment, Abdominal ultra-sound, FNAC of the liver, determination of serology for Hepatitis B (HBsAg) by ELISA.

Results: In this study the prevalence of HBV among 90 patients with HCC was 68.9%, observed most commonly among age group 30-60 years, with male to female ratio of 2.7:1. Majority of the participant with HCC the most frequent reported clinical features included Generalize body malaise (98.9%), sweat (97.8%), weight loss (97.8%), loss of appetite (80%), yellowish discolouration of the eyes (63.3%) and abdominal pains (52.2%).

Conclusion: This study revealed high prevalence of HBV among patients with HCC and the highest prevalence of the disease was found among patients aged between 30-60 years suggesting early childhood exposure to HBV. Therefore early screening and treatment of chronic active HBV infection is recommended in order to reduce the magnitude of HCC in our setting.

DEFINITION OF TERMS

- Hepatocellular carcinoma is defined as FNAC confirmed which was described by the presence of cytological features trabecular pattern, pleomorphism, increase nuclear cytoplasmic (N/C) ratio and hyperchromatic nuclei.

- Consecutive recruitment/sampling is defined epidemiologically as the sampling procedure that seeks to include all accessible subjects as part of the sample (all individuals who are eligible at presentation are included in the study).

ABBREVIATIONS

ALT	Alanine amino transferase
ASAT	Aspartate amino transferase
AFP	Alpha feto protein
CCP	Comprihensive chemistry panel
CPL	Central Pathology Laboratory
ELISA	Enzyme linked immunosobent assay
EPI	Extended program of immunization
FNAC	Fine needle aspiration for cytology
FBP	Full Blood Picture
HBc	Hepatitis B core
HBeAg	Hepatitis B e antigen
HBsAg	Hepatitis B Surface antigen
HBV	Hepatitis B Virus
HCC	Hepatocellular carcinoma
HIV	Human Immunodeficiency Virus
IgM	Immunoglobin M
IgG	Immunoglobin G
INR	International Normalization Ratio
GLC	Glucose
LFT	Liver Function Test
MNH	Muhimbili National Hospital
MTCT	Mother to child transmission
OPD	Out patient department
PCR	Polymerase Chain Reaction
PT	Prothrombin Time
PTT	Partial Thromboplastine Time
RFT	Renal function test
WHO	World Health Organisation

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CHAPTER ONE

1.1 INTRODUCTION

Despite the availability of safe and effective vaccines for the past two decades, Hepatitis B Virus infection continues to be an important public-health problem [1].

Approximately 2 billion people world-wide have been infected with HBV and up to 450 million people are chronic carriers, accounting for 55% of global cases of HCC of which 89% are found in endemic regions where tens of millions of new cases of HBV infections occur annually, Causing approximately 2 million deaths per year globally, due to various complication the most common being HCC [2, 3].

It is estimated that about one third of the infected individuals with HBV have symptoms and/or biological evidence of hepatitis. It is well established that the earlier the contamination, the higher the risk of chronic infection which is as high as 90% or greater with peri-natal transmission, whereas exposure during adolescence or young adulthood is associated with a 95% or greater likelihood that the disease will be self-limiting. Chronic carriers have a high mortality rate due to complications the commonest and fatal being HCC.

1.1.1 Epidemiology:

The prevalence of HBV varies widely in the world. Regions are divided into areas of low, intermediate and high prevalence, as defined below:

High prevalence: Approx. 8% of the population is currently infected, with a lifetime risk of infection greater than 60%. About 45% of the world's population lives in these regions of high prevalence. Among these regions, the transmission of HBV usually occurs during the peri-natal period (MTCT). These regions include South East Asia, China, sub-Saharan Africa and the Amazon Basin [4]

Intermediate prevalence: Approx. 2% to 7% of the population is infected, with a lifetime risk of infection being 20% to 60%. In these regions infections with HBV occur in all age groups. The regions include Eastern and Southern Europe, The Middle East, Japan and part of South America which accounts for about 43% of the global population [4].

Low prevalence: < 2% of the population is infected, with the lifetime risk of infection being <20%. These regions of low prevalence include: North America, Northern and Western Europe and Australia which accounts for only 12% of the global population [4].

According to WHO > 2 billion people have been exposed to HBV, of which approx. 450 million are chronically infected and act as the HBV reservoir.

In Europe alone which is a low prevalent region, approx. 1 million people are infected with HBV annually, of which 0.9 % of the cases becoming carriers and 0.2% of the cases die due to complications of HBV infection.

In the United States which is also a low prevalent region, around 1.2 million people are infected with HBV annually. With the infections being common among Alaskan natives, Pacific Islanders and infants of first-generation immigrant mothers from highly endemic areas, where the rates of chronic HBV is up to 15% [5].
In Africa:

- A study done in Gambia to determine the prevalence of HBV revealed that up to 20% of the infected populations were chronic carriers [6].

- In sub-Saharan Africa the prevalence of HBV surface antigen (HBsAg) is approx. 20% [7].

- In Tanzania a study done to examine markers of hepatitis B virus infection in the population of Dar es salaam residents. 542 serum samples from health workers were analyzed and revealed 9.6% of the samples tested positive for HBsAg showing that Hepatitis B infection is very common in the country [8].

1.1.2 HCC and HBV:

HCC is the most common liver cancer, accounting for 4% of all malignant tumors worldwide,

In men is the 5th most frequently diagnosed malignancy in the world and is the 2nd leading cause of cancer-related death in the world. In women, HCC is the 7th most commonly diagnosed malignancy and the 6th leading cause of cancer death [9].

Patients with persistent HBV infection, have 100 times higher risk of HCC compared to non-infected individuals. Other risk factors of HCC apart HBV infection include; aflatoxicosis, alcoholism, smoking, and hereditary conditions such as hemochromatosis, alpha-antitrypsin deficiency, tyrosinaemia, anabolic steroids and estrogen levels [10, 11, 12, 13, 14].

For more than 3 decades, Chronic HBV infection has been associated epidemiologically to the development of HCC where-by high prevalence of HCC has been demonstrated in regions with a high sero-prevalence for HBV infection. And that HBV infection remains the major etiological factor of HCC worldwide with more than one half of HCC patients being chronic carriers [15].

Moreover patients with HCC show up to 90% sero-prevalence of HBV when compared to 10-20% HBV sero-prevalence in the entire population without HCC in the same regions [15]. Additionally, a 10 to 100-fold risk of HCC has been observed in HBsAg carriers compared to non-carriers in different ethnic and social groups [16].

In a cohort study done to establish the relationships of HBV infection and hepatocellular carcinoma risk among 306 reproductive-aged Taiwanese women, the study revealed that the risk for hepatocellular carcinoma was statistically significantly higher among women with chronic infections and among those with persistent HBV infection during follow-up than among HBV-unexposed women [17].

Strong correlation between the prevalence of HBsAg carrier state and HCC was found in Korea, where among 112 patients with HCC, almost all gave positive tests for HBV infection (97% positivity for HbsAg, 38% for HBeAg, 83% for AFP) [18].
In another study done in Thailand the prevalence of HbsAg carrier state and HCC was found in Thailand, where 56% of HCC patients were HbsAg positive [19].

Retrospective study done in Nigeria (University of Maiduguri Teaching Hospital), to assess the association between HCC and HBV, of the 114 HCC patients enrolled 86.8% were found to be positive for HBsAg [20].

In another study done in Kenya to determine the prevalence of Hepatitis B Virus (HBV) surface antigen and HBV-associated hepatocellular carcinoma in Kenyans of various ages, revealed that 75% of the HCC cases were HBsAg positive [21].

1.1.3 Etio-Pathogenesis.

The mechanism by which HBV infection causes HCC is not completely known [22]. Evolution to HCC may be the direct effect of the virus itself, or it may be an indirect effect, through the process of the inflammation, regeneration and fibrosis associated with cirrhosis due to the HBV infection.

HBV DNA has been shown to become integrated within the chromosomes of infected hepatocytes, the integration of viral genetic material occurring in a critical location within the cellular genome. For example, integration of HBV DNA has been observed within the retinoic acid receptor alpha gene and within the human cyclin A gene, both playing crucial roles in cellular growth. However, in many if not most cases, the HBV DNA integration site does not appear to be in a critical location and the process appears to be random [22].

Furthermore, the length and the components in the HBV DNA integrant vary considerably and the viral DNA may be rearranged, deleted or present in repeats [23]. These findings suggest that it is not the process of integration itself that leads to HCC.

Possible Mechanisms of HBV induced HCC

The hepatitis B x gene (HBx) product has been implicated in causing HCC because it is a transcriptional activator of various cellular genes associated with growth control [24]. The HBx gene expression is also associated with activation of the Ras-Raf-MAP kinase pathway, an important cellular pathway that has been implicated in hepatocarcinogenesis. In addition, HBx has been found to interact with p53, interfering with its function as a tumor suppressor.

In keeping with the suggestion that HCC may be directly related to HBV infection, is the observation from several studies that elevated serum levels of HBV DNA (a marker of higher levels of HBV replication) are associated with a higher risk of HCC. A recent longitudinal study of 3,653 HBsAg-positive subjects in Taiwan found that an elevated serum level of HBV DNA (>10,000 copies/mL; ~ 2000 IU/mL) at baseline was a strong predictor of subsequent development of HCC, independent of serum hepatitis B e antigen (HBeAg) status, serum aminotransferases levels or the presence of cirrhosis [25,26].

Another line of evidence suggesting a direct hepatocarcinogenic role of HBV is the association of certain genotypes with higher rates of HCC. Example in a Asian cohorts, HBV genotype C is generally thought to increase the risk of HCC above that of genotype B. Thus the exact role of HBV genotype in hepatocarcinogenesis remains to be clarified [27].

Impact of HBV vaccine on HCC incidence

Chronic infection with hepatitis B virus is closely related to hepatocarcinogenesis. The outcome of current therapies for HCC is not satisfactory. Prevention is the best way to control HCC. Among the various strategies of HCC prevention, immunization against hepatitis B virus infection is the most effective. Universal hepatitis B immunization has proved to be effective in reducing the incidence of HCC [28].

In 1991, World Health Organization's (WHO) Global Advisory Group of the Expanded Program on Immunization (EPI) recommended that hepatitis B vaccine be integrated into national immunization programs in all countries with a hepatitis B carrier prevalence of

8% or greater. Hepatitis B vaccination is now part of the National Infant Immunization Schedule in 162 countries [28].

The most immediate impact of HB vaccine is a reduction in HBsAg carriage rates in infants, children and teenagers. Several studies, particularly in HBV endemic countries and regions have shown a noticeable reduction in HBsAg prevalence in selected populations (school children, national servicemen and antenatal women) after hepatitis B vaccination [29, 30].

Taiwan was the first to demonstrate this with convincing epidemiological data showing that the annual incidence of childhood HCC has decreased from 0.52 to 0.13/100 000 children [31, 32].

Similarly, a reduction of HCC has been demonstrated among Thailand native Alaskan children who received hepatitis B vaccination at birth [33, 34].

Association of viral Hepatitis, HIV and HCC

Hepatocellular carcinoma (HCC) is mainly driven by hepatitis co-infection in the HIV-infected patient. With the introduction of highly active antiretroviral therapy (ART) and the subsequent reduction of AIDS-associated mortality, HIV infection has become a chronic illness. This has been accompanied by an increase of end-stage liver disease (ESLD)–related mortality in the world due to chronic viral hepatitis infection (mainly HCV) [35, 36, 37]

Solid evidence from the last decade has highlighted that the natural history of HCV-related ESLD is accelerated in the setting of HIV infection [38, 39], with poor rates of survival [40]. It has been suggested that the time from HCV infection to the development of HCC is shorter in the setting of HIV infection [41, 42, 43]. In a national survey on deaths among HIV-infected patients, liver diseases represented the third most frequent underlying cause of death [44].

In a study that was done in spain on HCC in HIV infected patients revealed that HCC in HIV-infected patients is mainly associated with underlying chronic hepatitis C and has a more aggressive clinical course [45].

In another study that was done in North America on Presentation and outcome of hepatocellular carcinoma in HIV-infected patients: revealed that HIV-positive HCC patients are younger and more frequently symptomatic and infected with HCV or HBV than HIV-negative patients [46].

1.1.4 Hepatitis B Viral Infection

HBV, which may be acute or chronic, is caused by infection with human hepatitis B virus (HBV). A small hepatotropic DNA virus of the family *Hepadnaviridae,* that is distributed worldwide. Up to 5% of cases of chronic hepatitis B progress to fatal liver disease [47].
The incubation period following infection with HBV takes 40-150 days [47].
HBV serotype and genotype and their geographical distribution.
The virus is divided into four major serotypes namely adr, adw, ayr and ayw based on the antigenic epitopes present on its envelope proteins. And into eight genotypes namely A-H according to overall nucleotide sequence variation of the genome. The genotypes have a distinct geographical distribution and are used in tracing the evolution and transmission of the virus.
Difference between genotype affect the disease severity, course and likelihood of complication and response to treatment and possibly vaccination [48, 49].
Genotype A is mostly commonly found in America, Africa, India and Western Europe.
Genotype B is mostly found in Asia and the United States, furthely subdivived into B1 dominates in Japan, B2 in China and Vietnam while B3 found in Indonesia, B4 confined to Vietnam and B5 most commonly found in the Philippines.
Genotype C is mostly common in Asia and the United States. Subgenotype C1 found in Japan, Korea and China. C2 common in China and Bangladesh and C3 in oceania
Genotype D is mostly found in Southen Europe, India and the United State. Subgenotype D1-D4 mostly found in Europe, Africa and Asia [50].
Genotype E mostly found in West and Southern Africa.
Genotype F is mostly found in Central and Southern America.
Genotype G commonly found in France and the United States [51].
Genotype H is mostly commonly found in Central and Southern America and Califonia in the United States.

Africa has five genotypes A-E of these the predominant genotype are A in Kenya, B and D in Egypt, D in Tunisia, A-D in South Africa and E in Nigeria [52].
Genotype H is probably is probably split off from genotype F within the New World [53].

1.1.5 Hbv Modes of Transmission

In low-prevalent areas, most cases of HBV infection are acquired during adolescence to mid-adulthood, a period during which behaviors that increase the risk of HBV infection ie, intravenous drug abuse or unprotected sexual activity are most likely, Sex workers and homosexuals are at particular risk of sexual transmission of HBV. Intravenous drug abusers and health workers are at risk of parenteral transmission [54].

In high-prevalent areas, HBV is mostly transmitted during the peri-natal period from mother to infant(vertical transmission), conferring a high likelihood of chronicity Mothers who are HBsAg positive, particularly those who are also HBeAg positive are much more likely than others to transmit HBV to their off-springs. However horizontal transmission of HBV in this particular region is also common and it occurs through blood, blood products and sexual transmission, further more horizontal transmission through skin contact is said to be the most important though the mechanism is not known [54].

It's important to note that HBV is capable of surviving at -20˚C and heating at 60˚C for 4 hours. And it becomes in activated by heating at 100˚C for 10 min or by washing with sodium hypochlorite-bleach.

1.1.6 Factors that Influence the Course of Hbv Infection

As far as the development of HCC is concerned viral load has turned to be the most significant factor implicated. In a cohort study by Iloeje et al, which involved approximately 4000 Taiwanese it was found that viral load predicted progression to cirrhosis and eventually HCC [55].

Other factors that can influence the course of HBV infection include: Demographic characteristics (male sex and older age), comorbidities (ie, heavy alcohol consumption,

cigarette smoking, human immunodeficiency virus infection, hepatitis C virus infection, viral factors (genotype C, D, F, high level of HBV DNA, core/pre-core mutation) and clinical factors (cirrhosis, elevated alpha-fetoprotein (AFP) and alanine aminotransferase (ALT)) [55].

1.1.7 Clinical Disease

Acute Infection

Infection with acute HBV takes 6 weeks to 6 months to manifest clinically and the development of clinical manifestations is highly age dependent [56].

Infants usually do not develop any clinical signs or symptoms, but children aged between 1 to 5 years tend experience typical illness in up to 15% of HBV infections, where-as older children and adults develop signs and symptoms in up to 50% of HBV infections [56].

The consequences of acute HBV infection are highly variable, with the symptoms of infections varying in severity from mild to severe forms. Clinical signs and symptoms of acute HBV infection include fever, anorexia, nausea, malaise, vomiting, jaundice, dark urine, clay-colored or pale stools, and abdominal pain [56].

Extra-hepatic manifestations of acute HBV infections may occasionally occur and presents as skin rashes, arthralgias and arthritis. Severe hepatitis occurs in about 2% of persons with reported acute disease and has a case-fatality ratio of up to 93% [56, 57].

Chronic Infection

Chronic HBV infection is defined as the presence of HBsAg in serum for at least 6 months or the presence of HBsAg and the absence of anti-HBc immunoglobulin M (IgM).

The risk of developing chronic infection correlates inversely with age unlike in acute HBV infection. The risk is high in infants infected during peri-natal period in up to 90%. Whereas children infected between the age of 1 and 5 years develop chronic infection in up to 50% as compared to 6 to 10% of acutely infected older children and adults [56, 57].

Individuals with chronic HBV infection have a high chance of contracting chronic liver diseases as exemplified by primary HCC [58, 59, 60, 61]. The age at which chronic infection is acquired has shown to influence the risk of contracting the disease. Prospective studies have revealed that individuals who contract HBV infection as newborns or young children develop HCC in up to 25% as compared to 15% of adolescents and young adults who acquire chronic HBV infection [62, 63, 64].

Individuals with chronic active hepatitis tend develop fibrosis of the liver which eventually predisposes to the development of cirrhosis. Cirrhosis is an irreversible form of liver injury that may lead to the development of HCC through the promotional effect of hepatocyte regeneration [65].

1.1.8 Diagnosis of HBV

The definitive diagnosis of HBV infection is on the basis of serological testing for the detection of antibodies and antigens.

A number of serological tests are available to reach the diagnosis of HBV infection [66].

Acute HBV infection is diagnosed by the presence of HBsAg in serum which appears several weeks after infection and the development of IgM class of antibody.

Immunoassays for the detection of total anti-HBc involve both IgM and IgG class antibody to the core protein and indicate current or past exposure to virus and viral replication.

The detection of IgM anti-HBc in serum is diagnostic of acute HBV infection, where-as detection of IgG class of antibody in serum is a diagnostic of chronic HBV infection.

Detection of HBV DNA has limited usefulness for diagnostic purposes. HBV DNA is detectable in the serum of persons with acute and chronic HBV infection. However monitoring HBV DNA levels is useful in determining the response of chronic HBV infection to treatment [67, 68].

1.1.9 Diagnosis of HCC

Alpha fetal protein and liver ultra sound are the most current widely used screening tools for establishing the diagnosis of HCC. AFP is a serum glycoprotein that was first recognized as a marker for HCC more than 40 years ago and has since been described to detect preclinical HCC. Normal serum levels of AFP 10ng/ml-20ng/ml, elevations of AFP suggest underlying pathology which may be malignant [69].

In a study done by Trevisani F, (2001). To assess the efficacy of serum alpha-feto protein for diagnosis of hepatocellular carcinoma in patients with chronic liver disease. Revealed that AFP had a sensitivity of 39%-65%, a specificity of 76%-94%, and a positive predictive value of 9%-50% for the presence of HCC in previously published studies [70, 71].

Ultra - Sound is currently the technique of choice for screening focal hepatic lesions. On US, lesions may appear hyperechoic or show a ‘target lesion’ appearance, but none of these is specific. Any mass detected on US in a cirrhotic liver is suspicious of HCC, particularly if it is > 1 cm in size.

In a study done by **Bolondi L**, (2001).Surveillance program of cirrhotic patients for early diagnosis and treatment of hepatocellular carcinoma: revealed ultra sound had a sensitivity of 65%-80% and has a specificity of> 90% towards diagnosing HCC [72].

J. Bruix and M. Sherman, “Practice guidelines committee, American association for the study of liver diseases. Management of HCC revealed that ultra sound of the liver had a sensitivity of 65-80% and specificity of 90% in the diagnosis of HCC [73].

Other radiological imaging include CT-Scan and MR of the Iiver which provide diagnostic details superior to ultrasound but have not been validated to be used as tools for diagnosing HCC [74].

FNAC of the liver is the gold standard diagnostic tool for liver tumor (HCC) [74].
In a study by Soyuer I, Ekinci C, Kaya M, et al (2003). Assessment of the efficacy of FNAC of the liver in the diagnosis of HCC, revealed sensitivity of FNAC for diagnosing Hepatic malignancy to be 99.5% and the specificity was 100% [74].

1.1.10 Clinical Management

There is no specific treatment available for acute HBV infection, only supportive care is the mainstay of therapy [75].

In the past decade, numerous antiviral agents have been investigated as candidates for the treatment of chronic HBV infection. In 1976, two studies, one with leukocyte interferon and the other with beta interferon, suggested that interferon can affect the serologic profile of persons with chronic HBV infection by clearing serologic markers of HBV replication and improve the liver disease (by normalization of alanine aminotransferase levels and liver histological test results) [75, 76].

In a meta-analysis of 15 clinical trials to assess the efficacy of the interferon, the overall response rate (as measured by clearance of HBeAg from serum) was 33% among patients treated with interferon compared with 12% among untreated controls [77].

Alpha interferon treatment of patients with chronic hepatitis B is recommended for:

- Patients with persistent elevations in aminotransferase concentrations in serum,
- Detectable levels of HBsAg, HBeAg, and HBV DNA in serum,
- Chronic hepatitis on liver biopsy,
- Compensated liver disease [78].

The recommended regimen for Alpha interferon is either 5 million units daily/ 10 million units three times a week, given subcutaneously for 4 months.

Patient characteristics associated with a favorable response to therapy include:

- Low pre-therapy HBV DNA levels (<200 pg/ml),
- High pre-therapy alanine aminotransferase levels (>100 IU/ml),
- Short duration of infection, acquisition of disease in adulthood, Active histologic profile, and Absence of complicating diseases such as renal failure or human immunodeficiency virus infection.

But due to the fact that relatively few patients respond to interferon therapy/ unavailability, considerable researches have been conducted on antiviral agents which have shown to be well tolerated after oral administration, and these drugs agents lead to rapid decreases in HBV DNA levels, clearance of HBeAg, decreases in serum aminotransferase levels and improved liver histology. Once treatment is initiated, it should be maintained indefinitely even in patients who appear to have dramatic clinical improvement. These antiviral agents include include:

- Nucleoside analogues such as famciclovir and lamivudine
- Nucleotide such as Adefovir Dipivoxil and tenofovir
- Entecavir

1.2 PROBLEM STATEMENT

The Global burden of HBV related HCC is still a major challenge for public health in the 21st century especially in the endemic regions due to unavailability of early diagnostic techniques and treatment [79].

It is reported that individuals with chronic HBV infection have 100 times risk of developing HCC and that HBV infection takes a life every 30 seconds due to its related cirrhosis and HCC complications [80].

The global distribution of HCC correlates with the geographical prevalence of HBV which epidemiologically is categorized into three regions: high, intermediate and low prevalent regions.

In 2010 approximately 450 million individuals were reported to be chronic carriers of HBV globally, accounting for 55% of global cases of HCC. According to WHO up to 80% of the global cases HCC related HBV has been documented in the high prevalent regions, with the incidence of > 50 cases of HCC per 100,000 population [81].

In Asia and Africa particularly in the sub-Sahara Africa the burden of HCC is very high, and that HCC is the most frequent cause of cancer death among men. Chronic HBV infection is highly prevalent and the predominant risk factor for HCC in these high-incidence regions [82, 83].

In Tanzania a study that was done in Dodoma in 1979 on epidemiology of carcinoma of the liver revealed that of 939 clinically diagnosed malignancies 256 (27 %) had primary hepatocellular carcinoma [84]. However there is no published data showing the magnitude of HCC related HBV infection.

1.3 RATIONALE

Despite, high magnitude of HBV related HCC in Africa and the world at large, there is no information available on the magnitude of chronic HBV infection in patients with HCC in Tanzania and especially at Muhimbili national Hospital which is a public and tertiary Hospital receiving patient from all over the country.

Therefore, this study will provide information on the magnitude of HBV infection among patients with HCC which may be useful in creating awareness to the society that early and timely diagnosis of HBV is of paramount importance for timely provision of appropriate treatment in-order to reduce hepatitis B viral infection and thus reduce the progression or prevent HCC.

1.4 OBJECTIVES

1.4.1 Broad Objectives

- To determine the prevalence of Hepatitis B viral infection among patients with hepatocellular carcinoma attending Muhimbili national hospital.

1.4.2 Specific Objectives

- To determine the prevalence of HBV among patients with HCC attending Muhimbili national hospital
- To describe social demographic characteristics of patients with HCC related HBV attending Muhimbili national hospital
- To describe clinical characteristics of patients with HCC attending Muhimbili national hospital

CHAPTER TWO

2.0 METHODOLOGY

2.1 Study design

Hospital based descriptive cross sectional study.

2.2 Study site

The study was conducted at the Muhimbili National Hospital Gastroenterology Unity Mwaisela block, gastroenterology clinic and surgical wards in Kibasila block in Dar-es-Salaam.

2.3 Study population

Study included patients with HCC attending Muhimbili National Hospital.

2.4 Study duration

The study was conducted from June 2012 to February 2013.

2.5 Sampling and Sample size

Sample size calculation

Sample size was calculated from the following formula.

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

Whereas:

Z = statistic for a level of confidence = 1.96

P = prevalence of HBV related HCC in Nigeria (Estimated prevalence 86.8%)

d = precision = 7 %*(0.07)

n = minimum sample size= 90

*L. Naing, T. Winn2, B.N. Rusli1, Practical Issues in Calculating the Sample Size for Prevalence Studies Archives of Orofacial Sciences 2006; 1: 9-14

*In study done in Nigeria (University of Maiduguri Teaching Hospital), to assess the association between HCC and HBV, of the 114 HCC patients enrolled, 86.8% were found to be positive for HBsAg

NOTE: A precision of 7% was used in this study to calculate the sample size due to limited number of patients with HCC admitted in medical and surgical wards in a given time of data collection.

2. 6 Sampling technique

Subjects were recruited consecutively throughout the week for in-patients admitted in medical gastroenterology unit, surgical ward and at hepatology clinic for out-patients.

2. 7 Inclusion criteria

All patients with clinical suspicion of HCC attending MNH.

2. 8 Data collection methods

Patients were enrolled into the study after meeting the inclusion criteria. A standard questionnaire (Appendix) was used by the author to obtain the patient's information.

2.9 Procedures

- After recruitment of the clinically suspected cases of HCC by the study investigator following brief medical history (mild to moderate upper abdominal pain (right upper quadrant), weight loss, early satiety, or a palpable mass in the upper abdomen) and physical examination (Abdominal mass, ascites, jaundice) for patients admitted in medical and surgical wards.
- Abdominal ultra sound was performed on all patient with the clinical suspicion of HCC and patients who were found to have features suggestive of HCC underwent FNAC of the liver result of which were analyzed at the CPL (cytology laboratory).
- For participants with deranged coagulation profile results, transfusion of clotting factors was done and thereafter preceded with FNAC. For patients with impalpable nodular lesions ultra sound guidance was required during FNAC procedure.

- After the diagnosis of HCC was established, each participant thereafter was screened for HBsAg by ELISA .

2.10 Laboratory blood investigations:

- Venopuncture was done from the antecubital veins in a recumbent position, 10mls of blood was be collected into appropriate bottles for Hepatitis panel, alpha-fetoprotein, LFT, FBP, serum calcium, PT, PTT and INR samples and were analyzed at the microbiology, hematology and biochemistry laboratory at the Central Pathology Laboratory (MNH) using auto-analyzers.

2.11 Classical imaging features of HCC on USS (PHILIPS HP 5000 with a broadband curvelinear transducers of 2MHz to 5MHz)

- Lession size > 2 cm (Focal lesion: single/multiple)
- Presence of arterial hypervascularity
- Hypoechoeic/Hyperechoeic/Mixed-echo appearance.

2.12 FNAC procedure

The skin at the right upper quadrant region is swabbed with antiseptic solution and draped with sterile surgical towel then followed by local anaesthetic injection. The nodular lesion on the liver is identified by palpation (for impalpable nodules ultra sound guidance is used) and then 22 gauge needle is inserted two to three times to ensure a good specimen is collected. The aspirate is then smeared on glass ground slides and fixed in 95% ethyl alcohol; the smear is then stained by the Papanicolau technique/stain.

2.13 HBsAg

The determination of hepatitis B surface antigen (HBsAg) was done using ELISA (Abbott AxSYM)

2.14 AFP

Alpha FetoProtein was quantitatively determined using Micro-practicle Enzyme Immuno Assay (MEIA Abbott AxSYM)

2.15 Pre-testing of study instrument:

This was done two weeks before starting data collection to provide time for any correction

2.16 Ethical consideration:

- Ethical clearance to conduct the study was sought from Muhimbili University of Health and Allied Sciences Ethical Review Board. Permission to do the study was obtained from the Hospital management.
- Informed consent: A written informed consent was given to all participate who were clinically suspected to have HCC.
- Disposal of the patients: After FNAC was done patients were counseled on the results and for those patients who were stable they were sectioned to attend outpatient department were they were being followed up 3 monthly, however for patients who were admitted due to HCC complications, after FNAC such patients continued with the management accordingly in the gastro and surgical wards respectively.

2.17 Data entry and analysis

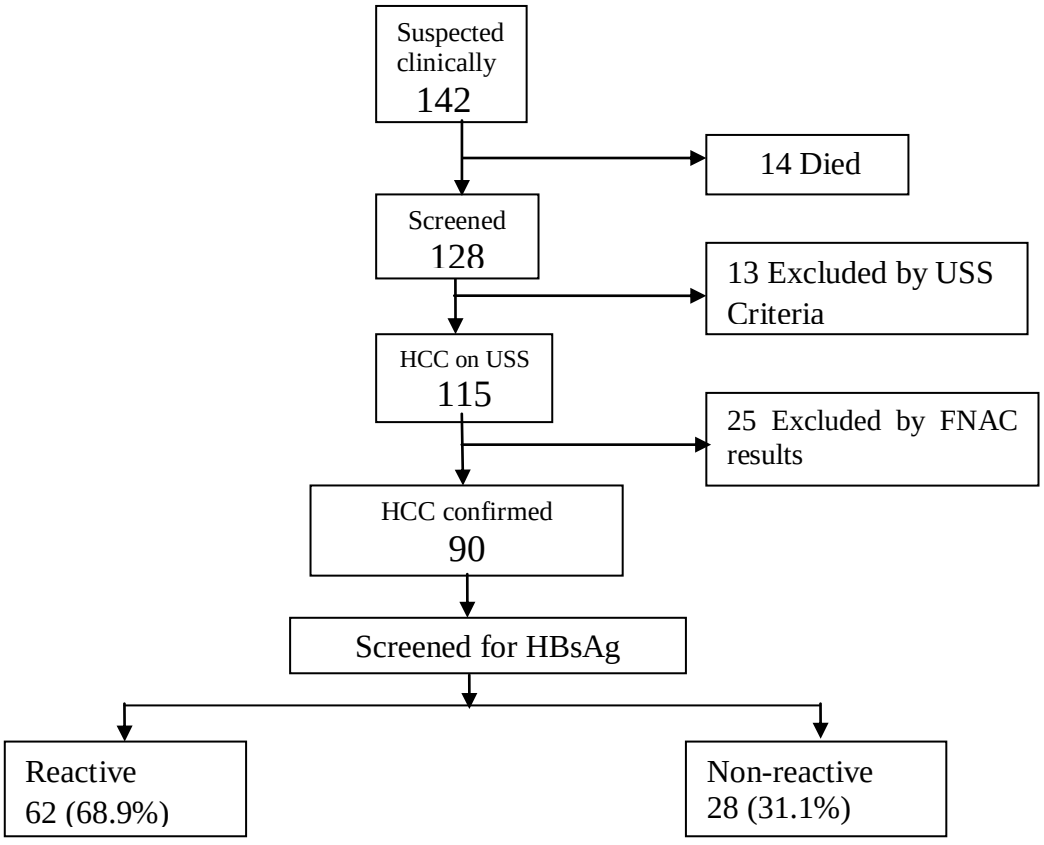
- Data was checked for completeness and consistency and errors or discrepancies found were promptly corrected.
- SPSS version 16 was used to enter and analyzed data.
- Descriptive statistics- was done by frequencies, percentages, means and proportions.
- **$\chi 2$ test** for categorical data.
- P-value of ≤ 0.05 was taken as significant.

CHAPTER THREE

3.1 RESULTS

A total of 142 clinically suspected patients with HCC were enrolled into the study between June 2012 and February 2013 of which 14 patients died due to complication of HCC (encephalopathy, hepato-renal syndrome, electrolyte imbalance, spontaneous bacterial peritonitis, hypogycemia and bleeding abdomalities) before abdominal ultra sound was done. The remaining 128 clinically suspected patients with HCC underwent abdominal ultra sound (13 were excluded by USS criteria: liver cirrhosis, liver abscess, liver cyst, fatty liver and etc) where 115 patients were found to have features suggestive of HCC and were subjected to FNAC of the liver, of which 90 patients were confirmed to have HCC (25 HCC suspected patients had other FNAC findings: Secondary liver metastases, Cholangiocarcinoma, Hepatoblastoma and micronodular cirrhosis). Hepatitis B surface Antigen (HBsAg) was detected in 62 (68.9%) of the 90 HCC patients tested, while 28 (31.1%) of the HCC patients were without HBsAg antigenemia as presented in the flow chart below (figure 1).

Figure 1: Flow chart



Of 90 patients with HCC 63 (70%) where male and 27 (30%) were female giving a ratio of 2.3:1, majority of which 81.1% were coming from urban. The overall mean (SD) age of the participants was 50.69 (±14.4) ranging from 25 to 86 years. Most participants 58 (64.4%) were found between the age group 30-60 years (P> 0.05).The social-demographic characteristics of the study population are presented in table 1.

Majority of the participants 74 (82.2%) had primary and below level of education while only 17.8% had post-secondary and above level of education with female having lower level of primary education as compared to males.

Furthermore, among HCC patients high proportion of males were married 39 (61.9%) or single 20 (31.8%) while in female a high proportion were observed among married participants 17 (63.0%). (P>0.05)

As compared to female a high proportion of males were practising self-employment 26 (41.3%) while a high proportion of females were unemployed 23 (85.2%) most of them being house wives. (P<0.05)

About 49 (54.4%) of the participants with HCC reported to be/to have ever consumed alcohol in their life while high proportion 73 (81.1%) with HCC reported not to have smoked cigarette, furthermore majority of the participants 88 (97.8%) with HCC reported not to have used recreational drugs (substance of abuse.), while high proportion of the participants 54 (60.0%) reported to have had multiple sexual partners in their life with majority of the participants 72 (80%) reported never to have used condom.

Table 1: Social demographic characteristics of patients with HCC

	Characteristics	Male (%) n=63(70)	Female (%) n=27(30)	Total n=90	P value
1	**Age group**				
	<30	3 (4.7)	1 (3.7)	4 (4.5)	
	30-60	43 (68.3)	15 (55.6)	58 (64.4)	
	>60	17 (27.0)	11 (40.7)	28 (31.1)	0.434
2	**Level of education**				
	1^0 school & below	48 (76.2)	26 (96.3)	74 (82.2)	
	2^0 school & above	15 (23.8)	1 (3.7)	16 (17.8)	**0.002**
3	**Marital status**				
	Single	20 (31.8)	4 (14.8)	24 (26.6)	
	Married	39 (61.9)	17 (63.0)	56 (62.2)	
	Divorced	1 (1.6)	1 (3.7)	2 (2.2)	
	Widowed	3 (4.8)	5 (18.5)	8 (8.9)	0.142
4	**Occupation**				
	Employed	19 (30.2)	2 (7.4)	21 (23.3)	
	Unemployed	18 (28.6)	23 (85.2)	41 (45.6)	
	Self employed	26 (41.3)	2 (7.4)	28 (31.1)	**0.001**
5	**Alcohol consumption**				
	No	26 (41.3)	15 (55.6)	41 (45.6)	
	Yes	37 (58.7)	12 (44.4)	49 (54.5)	0.143
6	**Cigarette smoking**				
	No	49 (77.8)	24 (88.9)	73 (81.1)	
	Yes	14 (22.2)	3 (11.1)	17 (18.9)	0.217
7	**Recreational drugs**				
	No	61 (96.8)	27 (100)	88 (97.8)	
	Yes	2 (3.2)	0 (0)	2 (2.2)	0.349
8	**Promiscuity**				
	Single	26 (41.3)	10 (37.0)	36 (40.0)	
	Multiple	37 (58.7)	17 (63.0)	54 (60.0)	0.466
9	**Condom use**				
	No	48 (76.2)	24 (88.9)	72 (80.0)	
	Yes	15 (23.8)	3 (11.1)	18 (20.0)	0.168

Among the HBsAg reactive HCC patients, males accounted for 45 (71.4%) and female 17 (62.9%), giving a ratio of 2.7:1 (Table 2), Table 2. Furthermore higher prevalence of HBsAg 39 (69.6%) were observed among HCC patients aged between 30-60 years, however the difference in age groups were not statistically significant (P>0.05)

There were no significant difference between the levels of education status and prevalence of HBV infection, however a higher proportion of participants with HBV infection 52 (70.3%) were observed among participants with primary school education and below, table 2.

Similarly the prevalence of HBV infection among patients with HCC co-infected with HBV was not associated with marital status; however a higher prevalence was observed among married participants 39 (69.6%), while high proportion of participants 33 (73.3%) with HBV were un-employed (P>0.05).

Majority of the participants with HCC co-infected with HBV reported neither to have smoked cigarette nor to have consumed alcohol in their life 53 (70.7%) and 35 (85.4.5%) respectively. Furthermore, majority of the participants 61 (69.3%) with HCC co-infected with HBV reported not to have used recreational drugs (substance of abuse), while high proportion of the participants 41 (70.7%) reported to have had multiple sexual partners in their life with majority of the participants 48 (68.7%) reported never to have used condom.

Table 2: Prevalence of HBV infection among patient with HCC (n=90)

	Characteristics	HBs Ag (%) Reactive	Total no Tested	P value
1	**Sex**			
	Male	45 (71.4)	63	
	Female	17 (62.9)	27	0.427
2	**Age group**			
	<30	4 (100)	4	
	30-60	39 (67.2)	58	
	>60	19 (67.9)	28	0.388
3	**Level of education**			
	1^0 school & below	52 (70.3)	74	
	2^0 school & above	10 (62.5)	16	0.623
4	**Marital status**			
	Single	18 (75)	24	
	Married	39 (69.6)	56	
	Divorced	0 (0)	2	
	Widowed	5 (62.5)	8	0.275
5	**Occupation**			
	Employed	10 (58.8)	17	
	Unemployed	33 (73.3)	45	
	Self employed	19 (67.9)	28	0.91
6	**Cigarette smoking**			
	No	53 (70.7)	75	
	Yes	9 (53.9)	17	0.115
7	**Alcohol consumption**			
	No	35 (85.4)	41	
	Yes	27 (55.1)	49	**0.002**
8	**Recreational drugs**			
	No	61 (69.3)	88	
	Yes	1 (50.0)	2	0.56
9	**Promiscuity**			
	Single	41 (70.7)	58	
	Multiple	21 (65.6)	32	0.278
10	**Condom use**			
	No	48 (66.7)	72	
	Yes	14 (77.8)	18	0.362

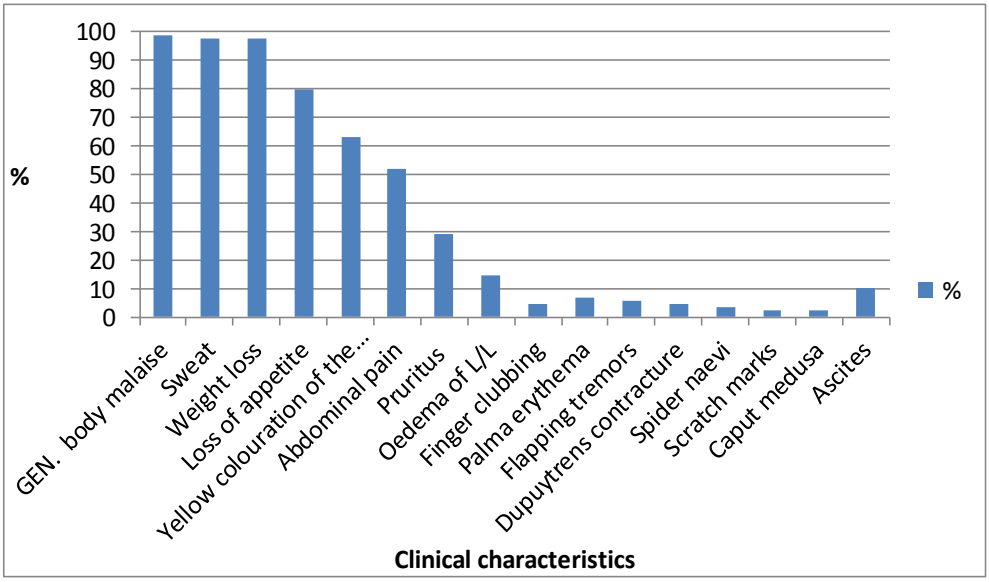


Figure 2: Distribution of the clinical features of patients with HCC

In majority of the study participants regardless of hepatitis B status the most frequent reported clinical features included generalized body malaise (98.9%), sweat (97.8%), weight loss (97.8%), loss of appetite (80.0%), yellowish discolouration of the eyes (63.3%) and abdominal pains (52.2%) as the most prevailing clinical features in the course of their illness.

CHAPTER FOUR

4.0 DISCUSSION

This study was conducted among HCC patients. The aim of the study was to determine the prevalence of HBV infection and to describe social-demographic characteristics and clinical characteristics among patients with HCC attending MNH.

The sero-prevalence of HBV infection among patients with HCC in this study was found to be 68.9%, majority of which being urban dwellers. This finding compares with similar studies carried out in Kenya 75% [85], in Gombe North East Nigeria 67% [86] and in Maiduguri Nigeria 86.8% [87], which incriminated HBV as the most common etiologic factor for HCC.

The results obtained in this study also indicate male to female ratio among HCC patients was found to be 2.3:1 which is similar to other studies done world-wide indicating male predominance in HCC, in Kenya 5:1, in Maiduguri Nigeria 2.5: 1 and Mustapha et al 2007 4: 1.

Furthermore the ratio of male to female of patients with HCC infected with HBV in this study was found to be 2.7: 1 which compares favourably with findings from similar studies carried out in Kenya ratio 5:2 and in Maiduguri Nigeria ratio 2.7: 1.

The mean age of patient with HCC in this study was 50.69 (±14.4) years with the highest incidence of the disease found among those aged 30-60 years males being mostly affected and that majority of the participants had low social economic status. This is similar to the findings in Kenya 40 years (30-60 years), in Enugu South East Nigeria 43.17±16.52 years [88] and Maiduguri Northern Nigeria 48.6±14.7 years (40-59 years).

The age difference as was observed among HCC patients associated with HBV infection in which the highest incidence of the disease was found among those aged between 30-60 years and the number of HBsAg reactive tend to decrease with advance in age suggesting either mother to child transmission or early childhood exposure to HBV, these findings

were consistent with previous similar studies carried in Maiduguri University teaching hospital Nigeria, showing that patients who are chronically infected with HBV (through vertical transmission) tend to develop HCC in their third and fourth decades of life.

Most of the frequent clinical features reported in this study were non-specific, however the few specific clinical features reported included yellowish discolouration of the eyes, Pruritus, Abdominal pain, Weight loss, and Ascites, similar presentation were reported in studies carried out in Ile-lfe Southwest Nigeria [89] and in Enugu South East Nigeria, these generally reflects the problem of late presentation which is common in patients with HCC.

Other specific clinical features that were less frequent reported included oedema of the lower limbs, finger clubbing, palmar erythema, duputriens contracture and caput medusa showing to the fact that most patients present at time when the disease is advanced, with profound hepatic failure, portal H.T.N and prospect of only palliative treatment.

4.1 STUDY LIMITATION

1. There may have been an underestimation of HCC results because FNAC is not a gold standard tool for the diagnosis of HCC.
2. The study could not take into account the use of term CHBV due to lack of standardized definition in our setting that is internationally accepted and hence used HBV which cannot be linked as causal for HCC.
3. Other methods of determining HBV infectivity (HBeAg, HBcoreAg and HBV-DNA) could not be taken into accounts in this study due to lack of finance.

CHAPTER FIVE

5.1 CONCLUSION

The study revealed high prevalence of HBV among patients with HCC which principally affecting middle-aged men, majority of them being urban dwellers and presenting with advanced disease.

5.2 RECOMMENDATION

1. Early screening and treatment of chronic active HBV infection is recommended in order to reduce the incidence of HCC in our setting.

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APPENDICES

Appendix I: Questionnaire

PREVALENCE OF HEPATITIS B INFECTION AMONG PATIENTS WITH HEPATOCELLULAR CARCINOMA ATTENDING MUHIMBILI NATIONAL HOSPITAL.

PART I: DERMOGRAPHIC CHARACTERISTICS:

1. Date of interview……
2. Serial number…….
3. Name…….
4. Tel no: ……..
5. Sex:

   Male ☐

   Female ☐
6. Date of birth: …….
7. Residency: ……...
8. Religion

   Catholic

   Muslim ☐
9. SDA
10. Assemblies of God
11. Any other (specify)……

12. Marital status:

    1. Single
    2. Cohabiting
    3. Married
    4. Divorced
    5. Widowed

13. Highest formal education achieved:

    1. None
    2. Primary school
    3. Secondary school
    4. Post secondary

14. What is your occupation

    1. Government employee
    2. Private sector employee
    3. Petty trader
    4. Housewife
    5. Not employed
    6. Other (mention)……..

15. Have you ever smoked cigarette?

    1. Never

    2. Yes but quit (if yes go to quest. 13) ☐

    3. Yes till now (if yes go to quest. 15)

16. When did you quit smoking…….. (months)

17. How many cigarette where you smoking per day…….

18. How many cigarette do you smoke per day…….

19. Do you drink alcohol?

    1. Never

    2. Yes but quit ☐

    3. Yes till now

20. When did you quit drinking alcohol…… (months)

21. Which type of alcohol do/did you drink? (multiple response)

    1. Local brew ☐

    2. Beer

    3. Konyagi

    4. Wine

    5. Whisky

    6. Any other…….

22. How many years have you been drinking alcohol

23. How much do/did you drink per week? ……..

24. Have you ever used drugs of abuse

1. Yes
2. No ☐

25. If yes when did you begin --------years

26. At what age did you begin sexual intercourse ---------years

27. Number of sexual partners in a life time----------

28. Do/did you use condoms during intercourse

1. Yes
2. No ☐

PART II: MEDICAL HISTORY AND PHYSICAL SIGNS

	Yes	No
29. Generalized body malaise	☐	☐
30. Sweats	☐	☐
31. Weight loss	☐	☐
32. Loss of appetite	☐	☐
33. Yellow colouration of the eyes	☐	☐
34. Abdominal pain	☐	☐
35. Pruritus (skin itching)	☐	☐
36. Oedema of L/L	☐	☐
37. Finger clubbing	☐	☐
38. Palma erythema	☐	☐

39. Flapping tremors ☐ ☐

40. Dupuytrens contracture ☐ ☐

41. Spider naevi ☐ ☐

42. Scratch marks ☐ ☐

43. Caput medusa

44. Liver palpable ☐ ☐

1. Liver span-------cm

2. Tendernes ☐ ☐

3.surface - smooth---- rough--- nodular----

45. Ascites ☐ ☐

46. History of HBV Vaccine

1. Yes

2. No ☐

47. if yes when were you vaccinated…….(years)

PART III: PROCEDURE

PROCEDURE	**RESULTS**
ABDOMINAL USS	
FNAC	

PART IV: LABORATORY FINDINGS

TEST	**RESULTS**
HBsAg	
FBP	
Glucose	
LFT (ASAT & ALAT)	
Calcium	
AFP	

Appendix ii: Informed Consent Form

ID no…….

Consent to participate in the study on the Prevalence of Hepatitis B infection among patients with Hepatocellular carcinoma attending MNH

Greetings! My name is Kaduri Ombeni, a postgraduate student at MUHAS, conducting a study on the prevalence of chronic hepatitis B infection among patients with hepatocellular carcinoma attending MNH.

The purpose of the study; To determine the prevalence of chronic Hepatitis B viral infection among patients with hepatocellular carcinoma attending Muhimbili national hospital.

What participation involves:

If you agree to participate in the study, you will be interviewed, and a detailed clinical history regarding your health will be requested as well so investigation will be taken.

Confidentiality:

All information collected on questionnaires will be entered into computer with special identification number. The questionnaires will be handled with greater secrecy in order to maintain your confidentiality.

Risks: There is no risk associated with this study

Benefits: Patients found to have HCC will be counseled and treatment of complications will be offered and for those found to have hepatitis B infection will be offered appropriate treatment.

Right to withdraw and alternatives: Participating in this study is completely a voluntary. Participant is allowed to withdraw from the study any time and yet continue to receive all services that are normally provided in the hospital.

Whom to contact: If you have any question about the study or in case of further information, you can contact Kaduri Ombeni 0713665818. If you have questions about your rights as a participant you may contact Prof M M Aboud, Chairman of MUHAS Research and Publications Committee.

P.O.BOX 65001 Dar es Salaam. Tel 2150302-6

Signature

Do you agree?

Participant agrees…….. Participant does NOT agree……

I …… have read and understood the content of this form .My questions have been answered and I voluntarily agreed to participate in this study.

Signature of participant…….

Signature of witness ……..…… Date of signed consent…….

Appendix iii: Fomu ya Ridhaa (Swahili Version)

Nambari ya usaili …….

Salaaam! Mimi naitwa Kaduri Ombeni, ni mwanafunzi wa shahada ya udhamili - Chuo Kikuu cha Sayansi za Afya na Tiba – Muhimbili. Nafanya utafiti kuhusu.....

Madhumuni ya utafiti .....

Jinsi ya kushiriki

Kama utakubali kushiriki katika utafiti huu, utaombwa kujibu maswali kadhaa na pia kufanyiwa vipimo vya damu na figo.Vipimo vitafanyika bure bila wewe kuchangia pesa.

Utunzaji wa siri

Taarifa zote zitatunzwa kwa siri kwa kutumia herufi au nambari badala ya jina la mgonjwa.

Faida

Mgonjwa atakaye gunduluka kuwa na saratani ya ini atapatiwa matibabu husika yakiambatana na ushauri nasaha, halikadhalika kwa mgonjwa atakaye gundulika kuwa na virusi ya hepatitis.

Madhara /Athari

Hakuna athari au madhara yoyote yanayotegemewa kutokana na ushiriki wako katika utafiti huu

Uhuru wa kushiriki

Una uhuru wa kushiriki au kutoshiriki kwenye utafiti huu, pia unaweza kujitoa wakati wowote. Kama utachagua kutoshiriki au kujitoa kwenye utafiti huu matibabu yako utaendelea kupata kama kawaida.

Taarifa

Endapo unahitaji kupata maelezo kuhusu haki zako au taarifa zaidi ,wasiliana na Kaduri Ombeni, Mtafiti, kupitia namba ya simu 0713665818 au Prof M M Aboud Mwenyekiti wa Kamati ya Utafiti, MUHAS S.L.P 651001,Dar es salaam, Simu 2150302-6. Je unakubali kushiriki kwenye utafiti? weka alama ya vema(√)

Ndiyo..... **Hapana.....**

Mimi ....., nimeelezwa na nimesoma maelezo haya hapo juu na kuyaelewa na Ninakubali kwa hiari yangu kushiriki kwenye utafiti huu

Sahihi ya mgonjwa .....

Sahihi ya ndugu/shahidi .....

Sahihi ya Mtafiti ..... Tarehe……

Appendix iv: Introduction Letter

MUHIMBILI UNIVERSITY OF HEALTH AND ALLIED SCIENCES

Directorate of Postgraduate Studies

P.O. BOX 65001
DAR ES SALAAM
TANZANIA.

Website: http://www.muhas.ac.tz

Tel: +255-(0)22-2150302 Ext 207.
Tel (Direct): +255-(0)22-2151378
Telefax:255-(0)22-2150465
E-mail: *dpgs@muhas.ac.tz*

Ref. No. HD/MUH/T.27/2010

31st July, 2012

Executive Director
Muhimbili National Hospital,
P.O. Box 65000,
DAR ES SALAAM.

Re: INTRODUCTION LETTER

The bearer of this letter Dr. Kaduri, Ombeni is a student at Muhimbili University of Health and Allied Sciences (MUHAS) pursuing MMed Internal Medicine.

As part of his studies he intends to do a study titled: ***"Prevalence of Chronic Hepatitis B Infection among patients with Hepatocullular Carcinoma attending Muhimbili National Hospaital".***

The research has been approved by the Chairman of University Senate.

Kindly provide him the necessary assistance to facilitate the conduct of his research.

We thank you for your cooperation.

A. Ndyeikiza
For: DIRECTOR, POSTGRADUATE STUDIES

cc: Dr. Kaduri, Ombeni
cc: Dean, School of Medicine

Appendix v: Ethical clearance

MUHIMBILI UNIVERSITY OF HEALTH AND ALLIED SCIENCES
DIRECTORATE OF POSTGRADUATE STUDIES

P.O. Box 65001
DAR-ES-SALAAM
TANZANIA
Telefax: 255-022-2150465
Telegrams: UNIVMED

E-MAIL dpgs@muhas.ac.tz
TEL: (255-022)-2150302-6 Ext. 207
Direct line: 2151378

Ref. No. MU/PGS/SAEC/Vol. VI/

20th June, 2012

Dr. Kaduri, Ombeni
M.Med. Internal Medicine,
MUHAS.

RE: APPROVAL OF ETHICAL CLEARANCE FOR A STUDY TITLED "PREVALENCE OF CHRONIC HEPATITITS B INFECTION AMONG PATIENTS WITH HEPATOCELLULAR CARCINOMA ATTENDING MUHIMBILI NATIONAL HOSPITAL"

Reference is made to the above heading.

I am pleased to inform you that, the Chairman has on behalf of the Senate approved ethical clearance for the above-mentioned study.

Thus ethical clearance is granted and you may proceed with the planned study.

Please liaise with bursar's office to get your research fund.

Prof. Z. Premji
DIRECTOR, POSTGRADUATE STUDIES

/emm

c.c. Vice Chancellor, MUHAS
c.c. Deputy Vice Chancellor – ARC, MUHAS
c.c. Dean, School of Medicine - MUHAS