

Silent Passenger: Incidental Hepatitis B in Abnormal Liver Function Workups

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Received: 17-07-2025 / Revised: 16-08-2025 / Accepted: 17-09-2025

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Conflict of interest: Nil

Abstract:

Background: Hepatitis B virus (HBV) infection affects more than 296 million people worldwide and remains a leading cause of chronic liver disease and hepatocellular carcinoma. Many individuals remain asymptomatic, leading to incidental detection during routine evaluations. Screening individuals with abnormal liver function tests (LFTs) can provide an opportunity for early diagnosis and intervention.

Methods: This descriptive study was conducted over one year (September 2023–September 2024) at a tertiary care center in Tamil Nadu. A total of 175 adult patients with abnormal LFTs detected during the routine master health checkups were screened for HBV surface antigen (HBsAg) using lateral flow immunochromatography (ICT) and chemiluminescent immunoassay (CLIA). Patients with known HBV infection, those under 18 years, and recently vaccinated individuals were excluded.

Results: Of 175 samples, 11 (6.3%) tested positive by ICT, while 14 (8.0%) were positive by CLIA. All ICT-positive samples were confirmed by CLIA. The additional three cases detected only by CLIA highlight its superior sensitivity. Among positives, 9 were male and 5 were female. Age distribution showed most cases were in the 30–50 years group.

Conclusion: Incidental detection of HBV in patients with abnormal LFTs during health checkups is valuable for identifying asymptomatic carriers. CLIA demonstrated better sensitivity compared to ICT, suggesting its preferred use in screening. Routine inclusion of HBsAg testing in health checkups can improve early diagnosis, reduce transmission, and guide appropriate follow-up and management.

Keywords: Hepatitis B virus, HBsAg, CLIA, ICT, liver function tests, asymptomatic carriers.

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Introduction

Hepatitis B virus is a blood borne DNA virus which belongs to member of the Hepadnaviridae family. It causes Hepatitis B infection in more than 300 million people worldwide and is also a common cause of various forms of liver disease with the tendency to cause carcinoma of liver [1]. Hepatitis B infection is always a global concern, as it tends to progress through various forms of illness like acute hepatitis, chronic hepatitis, severe liver failure, and death [2]. The Hepatitis B virus (HBV) is a partially double-stranded DNA virus, a species of the genus Orthohepadnavirus and a member of the Hepadnaviridae family of viruses. The viral genome of Hepatitis B - DNA is about 3.2 Kb that has four overlapping open reading frames- ORFs which is S, X, P, and C. The S/preS ORF encodes the three forms of hepatitis B surface antigen (HBsAg): long (HBsAg-L), medium (HBsAg-M) and small

(HBsAg-S). The regulatory protein X (HBx) is transcribed from X ORF; the P ORF encodes the viral DNA polymerase; and the C ORF encodes the core protein (HBcAg) and a related protein called which is the precursor of the secreted protein known as antigen e (HBeAg) [1].

The clinical features of Hepatitis B viral infection can vary in both acute and chronic diseases. During the acute Hepatitis B infection, patients can have subclinical illness or anicteric hepatitis, icteric hepatitis, or less commonly fulminant hepatitis. In chronic Hepatitis B infection, patients can have an asymptomatic carrier state, chronic hepatitis, cirrhosis, and hepatocellular carcinoma. The Initial symptoms are usually nonspecific like loss of appetite, loss of weight, nausea, vomiting, abdominal pain, and jaundice. In cases of severe

damage of liver, patients can develop jaundice, ascites, gastrointestinal bleeding secondary to oesophageal varices, coagulopathy, or infections. Patients can also present with Hepatic encephalopathy.

HBV infection is often asymptomatic in its early phase, and many individuals are unaware of their carrier status. Such undiagnosed cases contribute to ongoing transmission and delayed diagnosis of chronic liver disease [3]. Screening during master health checkups provides an opportunity for early detection, especially in individuals presenting with abnormal liver function tests (LFTs) without clear etiology. Several diagnostic methods exist for HBV detection. Lateral flow immunochromatography tests (ICT) are commonly used in field and low-resource settings due to their rapid turnaround and low cost, but their sensitivity is often lower compared to advanced assays such as chemiluminescent immunoassay (CLIA) [3-5]. This study aimed to evaluate the role of incidental HBV detection in patients with abnormal LFTs during health checkups and to compare the diagnostic yield of ICT versus CLIA.

Materials & Methods

Study design and Setting: A descriptive study was carried out in the Department of Virology, Government Tirunelveli Medical College & Hospital, Tamil Nadu, from September 2023 to September 2024.

Study population: A total of 175 consecutive adult patients (>18 years) presenting with abnormal LFTs during master health checkup were included after obtaining informed consent.

Exclusion criteria: All patients less than 18 years of age and known case of acute and chronic Hepatitis

B infection, those with recent history of vaccination with Hepatitis B, those with normal liver function tests.

Hepatitis B Surface antigen ICT: ICT is a lateral flow immunoassay based on antigen-antibody binding. The test strip is coated with antibodies specific to HBsAg (Hepatitis B surface antigen). When a patient's serum/plasma is applied, HBsAg (if present) binds to colloidal gold-labelled antibodies and migrates via capillary action. This complex gets captured by immobilized anti-HBs antibodies on the test line, forming a visible coloured band.

Chemiluminescent Immunoassay: CLIA is an immunoassay based on chemiluminescent signal detection. It uses antigen-antibody reactions where either the antigen (HBsAg) or antibody is labelled with an enzyme or acridinium ester. When a chemiluminescent substrate is added, the reaction emits light. The intensity of emitted light is measured by a luminometer, which is proportional to the amount of HBsAg. A control line ensures the test validity.

Data Analysis: The collected data was entered in Microsoft excel Version 2021. Results were expressed as frequency and percentages.

Result

Among 175 samples tested, CLIA detected 14 positive cases of HBsAg, whereas ICT detected 11 cases. All 11 ICT-positive samples were confirmed positive by CLIA, while 3 additional cases were detected only by CLIA. Among the testing, CLIA result were taken as final due to increased sensitivity and specificity (Table 1).

Table 1: Diagnostic accuracy of ICT compared with CLIA for detection of HBsAg (n = 175)

	CLIA Positive	CLIA Negative	Total
ICT Positive	11 (True Positives, TP)	0 (False Positives, FP)	11
ICT Negative	3 (False Negatives, FN)	161 (True Negatives, TN)	164
Total	14	161	175

When compared with CLIA as the reference method, ICT demonstrated a sensitivity of 78.6%, specificity of 100%, positive predictive value (PPV) of 100%, and negative predictive value (NPV) of 98.2%. This indicates that while ICT showed

excellent specificity and PPV, its lower sensitivity resulted in missed cases, highlighting the superior diagnostic performance of CLIA in detecting HBsAg (Table 2 & Figure 1).

Table 2: Sensitivity, specificity, and predictive values of ICT using CLIA as the reference standard

Parameter	Value (%)
Sensitivity	78.60%
Specificity	100%
Positive Predictive Value (PPV)	100%
Negative Predictive Value (NPV)	98.20%

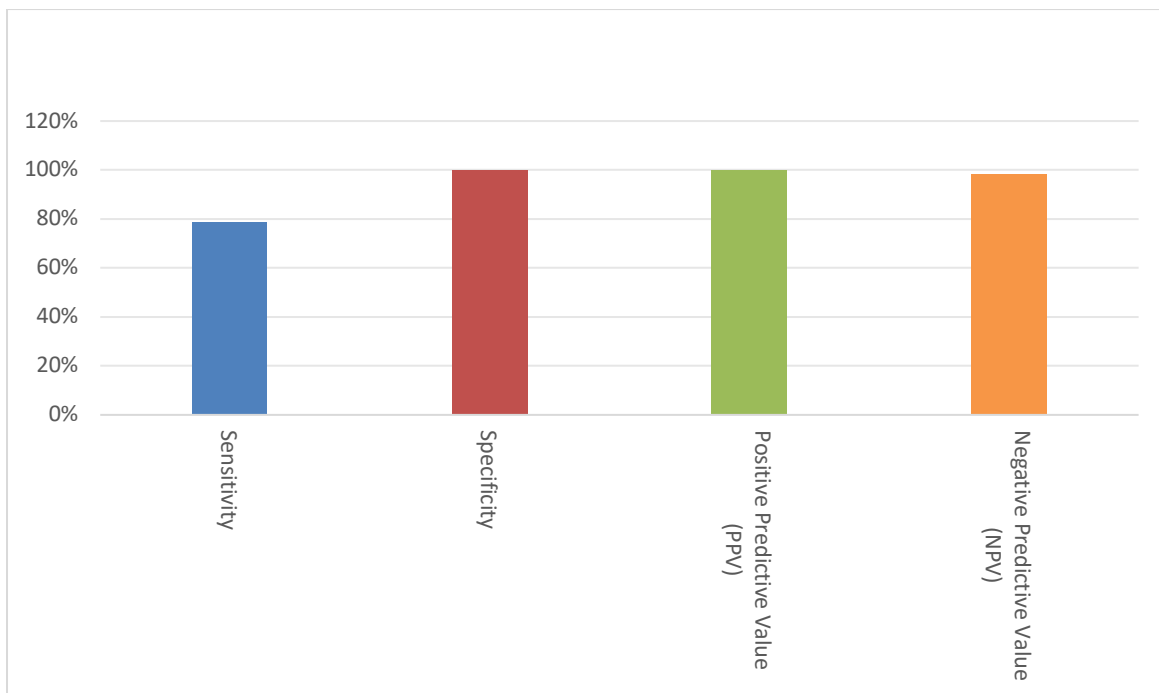


Figure 1: Sensitivity, specificity, and predictive values of ICT using CLIA as the

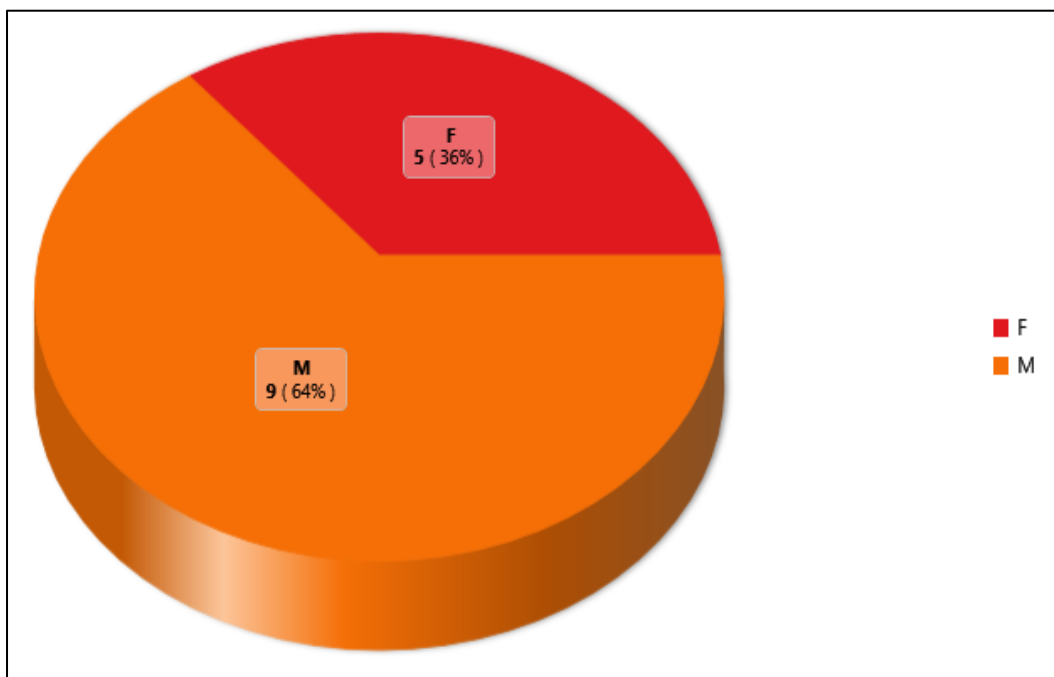


Figure 2: Gender distribution of HBsAg-positive cases

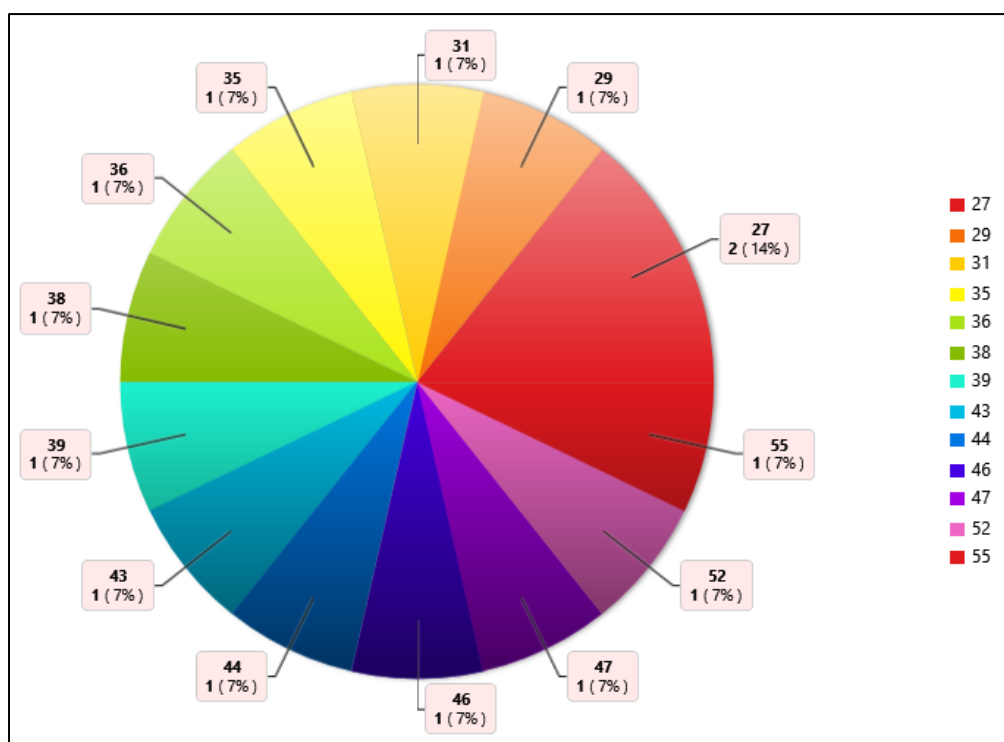


Figure 3: Age-wise distribution of HBsAg-positive cases

Of the 14 positive cases, 9 (64.3%) were male and 5 (35.7%) were female (Figure 2). Age-wise distribution showed the majority (57%) were in the 30–50 years age group (Figure 3). Follow ups and further evaluation needed to know if these cases are acute or chronic Hepatitis B.

Discussion

This study was conducted on the outpatients visiting the tertiary care institution with abnormal liver function tests detected during the master health checkup. 2mL of blood were collected in the serum activator Red BD Vacutainer tube. Serum specimens were utilized for Hepatitis B Surface antigen detection by rapid lateral flow through card and Chemiluminescent Immunoassay (CLIA).

The samples tested were 175 of which 14 (8.0%) were positive for Hepatitis B Surface antigen. The prevalence (8.0%) is consistent with reports from other Indian tertiary centers, which have documented HBV positivity ranging from 5–10% among individuals with abnormal LFTs [6-8]. The clinical proper history and physical examination, as well as the analysis of serum or viral biomarkers, are necessary for the diagnosis of hepatitis B. Hepatitis B surface antigen (HBsAg) is the main viral marker for viral serology of detection of hepatitis B, which can often be seen 1–12 weeks after the initial infection. As the initial point of contact for undiagnosed patients, primary care physicians are crucial in the diagnosis and prevention of disease. However, despite their sufficient understanding of risk factors, their limited diagnostic facilities prohibit them from diagnosing and following up on

cases [6]. Testing for Hepatitis B by HBsAg detection is not done on all routine cases with abnormal liver function tests and also during master health checkup. Inclusion of testing such as Hepatitis B Surface antigen detection in health checkups, especially those with abnormal liver function tests can be helpful in picking up Hepatitis B infection. While ICT remains valuable in resource-limited settings, its lower sensitivity leads to underdiagnosis. CLIA detected additional cases, supporting its use as a more reliable screening tool, consistent with previous comparative studies [9-10].

Conclusion

The main goals of Hepatitis B screening are to prevent transmission and reduce liver disease-related morbidity and mortality, since most of the cases of Hepatitis B are asymptomatic. Despite the availability of an effective vaccine for over three decades, the global burden of Hepatitis B has not declined substantially. Incorporating routine Hepatitis B screening among individuals with abnormal liver function tests during comprehensive health checkups can facilitate the early detection of asymptomatic cases. Early identification enables timely diagnosis, accurate classification of disease status, and initiation of appropriate monitoring and management strategies.

Limitations

The samples were not subjected to testing for Hepatitis C.

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