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RISK FACTORS AND CLINICAL ASPECTS OF HEPATITIS B AND C IN PATIENTS OF NASIRABAD DISTRICT, BALOCHISTAN

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Abstract

***Objective**: This study aimed to assess the prevalence of hepatitis B (HBV) and hepatitis C (HCV) infections in the local population of Nasirabad District in Balochistan, Pakistan. It also investigated sociodemographic characteristics, associated risk factors, and clinical as well as subclinical manifestations of these infections.*

***Methodology:** This observational research was carried out between August 2022 and November 2022. Data collection took place at the Departments of General Surgery and Pediatrics in Quetta's Bolan Medical Complex Hospital (BMCH) and District Headquarters Hospitals in Nasirabad Division, Balochistan, Pakistan. A total of 523 participants (291 males and 232 females) underwent screening tests for HBV and HCV using ELISA and ICT methods. Participants were selected sequentially. Diagnosis of chronic liver disease was based on a combination of medical history, clinical evaluation, ultrasound findings, and abnormal liver function tests.*

***Results:** HCV exhibited a significantly higher prevalence at 14.3% compared to the 6.7% prevalence of HBsAg. The prevalence of HBsAg varied by age groups, with the highest prevalence of 13.7% observed in individuals under 30. Among those aged 31 to 40, prevalence rates of 7%, 5%, 13%, and 13% were noted. For individuals aged 51 to 60, the prevalence remained at 13%. In the case of HCV, the prevalence also showed age-related variations. The under-30 age group had a prevalence of 14.3%. The 31–40 age group showed a prevalence of 10%, and for individuals aged 51 to 60, prevalence rates of 18.7%, 18.5%, and 18.7% were observed. These disparities in prevalence across different age groups underscore the significance of age-related factors and risk exposure in the transmission of these hepatitis viruses. Public health interventions and medical management strategies should consider these variations.*

***Conclusion:** To enhance public awareness of the routes of viral hepatitis transmission and prevention strategies, comprehensive community-based health education campaigns are imperative. Collaboration with healthcare professionals and the relevant authorities responsible for combating infectious diseases is essential to mitigate risk factors, such as sharing razors and exposure to unsafe medical injections.*

Keywords: *Clinical aspects, Hepatitis B, Hepatitis C, Nasirabad, Risk factors*

INTRODUCTION

Hepatitis is a pathological condition characterized by liver inflammation, often leading to liver damage with a spectrum of potential health complications, some of which can be severe and fatal. Notably, Hepatitis B virus (HBV) is a partially double-stranded DNA virus with a circular genome, whereas Hepatitis C virus (HCV) is a single-stranded RNA virus. Despite their shared status as enveloped viruses, these agents

exhibit distinct genomic structures and are classified into separate viral families. A fundamental understanding of these distinctions holds significant importance in the management and treatment of hepatitis infections (1).

Hepatitis, caused by RNA viruses, represents a global public health challenge and may culminate in either acute or chronic liver conditions, including cirrhosis (2). The worldwide prevalence of HBV infection has led to geographical categorization into three zones, denoting high, moderate, and low endemicity based on HBV carrier rates (3). Given the similarities in the modes of transmission for HBV and HCV, the idea of concurrent infection with these viruses is a plausible scenario (4). The three primary avenues of HCV transmission encompass the use of injectable drugs, the receipt of contaminated injections during medical procedures, and contact with infectious blood through transfusions of HCV-infected blood and blood products. Additionally, sexual transmission is recognized as a potential route of infection (5).

Globally, approximately 170 million individuals are afflicted with chronic HCV infection, while an estimated 350 million people grapple with chronic HBV infection, as reported by the World Health Organization in 2022 (6). The seroprevalence of hepatitis B surface antigen (HBsAg) is acknowledged to exhibit substantial geographic variability, with estimates ranging from 0.1% to 20% in different regions (7). It is well-established that individuals co-infected with HBV and HCV face an elevated risk of developing cirrhosis and decompensated liver disease (8). Pertinently, Hepatitis B has been documented to persist on environmental surfaces, such as desks and workbenches, for a duration of up to seven days after leaving the human body (9).

For many economically challenged countries, the prevalence of HCV infections poses a significant threat to public health due to inadequacies in healthcare systems to implement necessary infection prevention measures and limited public awareness regarding transmission modalities (10). Of note, barbers in Pakistan who engage in frequent face and underarm shaving practices have been identified as a risk factor for the transmission of both hepatitis B and hepatitis C (11). Co-infection with both HBV and HCV is closely associated with the development of more severe liver conditions and a heightened risk of liver cancer (12).

The situation is particularly dire in countries like Bangladesh, where eight million individuals are HBV carriers and over a million carry HCV. In Pakistan, HBV affects approximately 4.55 million people, while HCV afflicts an estimated 8.74 million individuals (13). Regrettably, the majority of fatalities stemming from these infections are attributed to cirrhosis and primary liver cancer (14). The prevalence of chronic liver disease linked to HCV has increased in Pakistan (15). HCV infection has been shown to progress slowly in a substantial portion of infected individuals, rendering the estimation of its frequency and prevalence a challenging endeavor (16). Parenteral routes account for the majority of HCV transmission (17, 18). Furthermore, liver grafts used in transplantation become infected within days, with continued infection leading to graft hepatitis and, in certain instances, failure (19, 20).

MATERIALS AND METHODS

SAMPLE COLLECTION

Between August 2022 and November 2022, data and blood samples were collected from patients with Hepatitis B (HBV) and Hepatitis C (HCV) through a survey conducted at the Bolan Medical Complex Hospital (BMCH) in Quetta and the District Headquarters Hospitals (DHQs) in Nasirabad, Balochistan. A total of 523 participants, consisting of 291 males and 232 females, underwent screening for HBV and HCV. Patients were selected sequentially, and their diagnoses of chronic liver disease were established based on their medical histories, clinical data, ultrasound results, and abnormal liver function tests.

QUESTIONNAIRE ADMINISTRATION

Sociodemographic information and potential risk factors for the patients were collected using a pretested structured questionnaire. The questionnaire underwent pretesting at the health facility, ensuring its accuracy and consistency by daily data review.

BLOOD COLLECTION AND HBV AND HCV DETERMINATION

Venous blood samples (5 mL) were collected in sterile conditions and placed in plastic tubes without anticoagulants. After allowing the blood to clot at room temperature, serum was obtained by centrifuging the samples at 3000 RPM for 15 minutes at 5 °C. The presence of HBV and HCV was determined through an initial screening test.

INITIAL SCREENING BY HBsAg ICT (IMMUNO-CHROMATOGRAPHIC TEST)

All blood samples were initially subjected to an immuno-chromatographic test (ICT) to detect the presence of HBsAg.

ELISA TEST FOR HBV

A commercially available ELISA kit (96 well plates) (BIOKIT, S.A, Barcelona-Spain) was used to perform the HBV ELISA test on positive sera, following the manufacturer's instructions.

INITIAL SCREENING BY HCV ICT (IMMUNO-CHROMATOGRAPHIC TEST)

An immuno-chromatographic test (ICT) was conducted on all blood samples as the initial screening for HCV.

ELISA TEST FOR HCV

The HCV ELISA test on positive sera was carried out using a commercially available kit, following the manufacturer's guidelines (BIOKIT, S.A, Barcelona-Spain).

BIOCHEMICAL ANALYSIS

Biochemical assays for Alanine Aminotransferase (ALT) and Aspartate Aminotransferase (AST) were performed using commercially available kits (Human, Germany) in accordance with the manufacturer's protocol.

ASSESSMENT OF LIVER FUNCTIONAL TESTS

The following methods were employed to assess AST and ALT:

ALANINE AMINOTRANSFERASE (ALT)

The kinetic technique for ALT was performed following the procedure outlined by Schumann & Klauke (2003), without the activation of pyridoxal phosphate. Calculations were made using the formula:

U/I = 'A/min X 16.67'

ASPARTATE AMINOTRANSFERASE (AST)

The AST catalyzed the transfer of the amino group from aspartate to oxoglutarate, resulting in the production of glutamate and oxaloacetate. Oxaloacetate was then converted to malate by malate dehydrogenase (MDH). These reactions were carried out at either 30°C or 37°C, with a decrease in nicotinamide adenine dinucleotide (NADH) levels. Calculations were made using the formula:

U/I = ΔA/min X 3333

STATISTICAL ANALYSIS

Biochemical analysis results were processed using Graph Pad Prism Version 6. Mean Standard Error of the Mean (Mean SEM) was employed for result presentation. Statistical tests, including Fisher's test with PLSD and two-way ANOVA, were conducted using software (IBMP Corp, USA).

Study Participants: Between August 2022 and November 2022, a total of 523 cases of hepatitis were documented at the (BMCH) in Quetta and the Nasirabad DHQs Hospitals in Balochistan. The study included 291 male and 232 female participants who were screened for hepatitis B (HBV) and hepatitis C (HCV).

RESULTS

BASELINE CHARACTERISTICS

In the study group, the prevalence of HCV was notably higher at 14.3% compared to the prevalence of HBsAg, which was 6.7%. Gender-specific distribution revealed that HBsAg was found in 6.5% of female participants (n = 15/232) and 6.9% of male participants (n = 20/291). Additionally, 15.6% (n = 60/384) of married participants and 10.8% (n = 15/139) of unmarried participants were found to be positive for HCV.

The prevalence of HBsAg also varied concerning sexual partners. Specifically, it was 5.4% (n = 20/371) in individuals with a single sexual partner, 4% (n = 5/125) in participants with multiple sexual partners, and 7.9% (n = 10/127) in individuals who reported no sexual activity (Table I).

Table I. The prevalence of HBsAg & HCV in Nasirabad (Balochistan), Pakistan's general population, and the associations between sociodemographic factors

Factors		HBsAg-+ve cases (n = 35)	HBsAg--ve Cases (n = 488)	χ2/P-value	HCV-+ve cases (n = 75)	HCV +ve cases (n = 448)	Statistical test/P-value
Age (years)	< 30	10 (7%)	137	/	20 (13.7%)	126	/
	31–40	10 (5%)	185	/	20 (10%)	175	/
	41–50	5 (4.8%)	101	6.7	20 (18.9%)	86	6.3
	51–60	10 (13%)	65	P = 0.08	15 (19.8%)	61	P = 0.099
Gender	Female	15 (6.5%)	217	2.1	30 (9%)	202	0.7
	Male	20 (6.9%)	271	P = 0.1	45 (15.5%)	246	P = 0.4
Relational status	Married	27 (7%)	357	0.27	60 (15.6%)	324	1.94
	Single	8 (5.8%)	131	P = 0.7	15 (10.8)	124	P = 0.2
Education	Uneducated	30 (8.9%)	208	24.54	30 (12.6%)	208	1.07
	Educated	5 (1.8%)	280	P < 0.00001	45 (15.8%)	240	P = 0.3
Religion	Hindu	5 (9.8%)	46	0.9	9 (18%)	41	0.6
	Muslim	30 (6%)	442	P = 0.3	66 (14%)	407	P = 0.5
Sex partners	1	20 (5.4%)	351	/	45 (12%)	326	/
	> 1	5 (20%)	20	8.4	15 (60%)	10	44.5
	Nil	10 (7.9%)	117	P =0.01	15 (11.8%)	112	P < 0.00001
Drug abuse	Yes	5 (50%)	5	30.6	5 (45.5%)	6	8.8
	No	30 (5.8%)	483	P < 0.00001	70 (13.7%)	442	P = 0.002

FACTORS ASSOCIATED WITH HCV AND HBSAG TRANSMISSION

Various factors were assessed for their association with HCV and HBsAg transmission. The odds ratio (OR) and corresponding confidence intervals (CI) are detailed below:

HOSPITALIZATION

Individuals with a history of hospitalization exhibited a higher risk of HCV and HBsAg transmission (OR = 2.40; CI: 1.51-6.13). HCV transmission risk was substantially increased for those with a history of hospitalization (OR = 10.50; CI: 17.12-17.01) (Table II).

SURGERY

Surgical procedures were associated with an increased risk of HCV and HBsAg transmission (OR = 2.26; CI: 2.23-7.46). Surgical procedures posed a higher risk of HCV transmission (OR = 6.34; CI: 2.81-12.37) (Table II).

Table II. The prevalence of HBsAg and HCV in general population of Nasirabad (Balochistan), Pakistan and its relations to risk factors

Factor		HBsAg-+ve cases (n = 35)	HBsAg--ve cases (n = 488)	95% CI P-value	HCV-+ve cases (n = 75)	HCV +ve cases (n = 448)	95% CI P-value
Hospitalized	Yes	15 (14.9%)	86	OR = 2.40 CI = 1.51 6.13	45 (44.6%)	56	OR = 10.50 CI = 17.12–17.01
	No	20 (4.7%)	402	P = 0.0007	30 (7%)	392	P < 0.0001
Surgery	Yes	7 (13%)	46	OR = 2.26 CI = 2.23-7.46	25 (48%)	27	OR = 6.34 CI = 2.81–12.37
	No	28 (6%)	442	P = 0.01	50 (10.6%)	421	P < 0.0001
Transfusion of Blood	Yes	15 (23.8%)	47	OR = 6.02 CI = 4.41–13.55	35 (57.4%)	26	OR = 13.77 CI = 7.13–26.15
	No	20 (4.3%)	441	P < 0.0001	40 (8.7%)	422	P < 0.0001
Injury byneedles	Yes	3 (23%)	10	OR = 3.57 CI = 2.16–16.08	10 (71.4%)	4	OR = 16.07 CI = 4.30–45.05
	No	32 (6.3%)	478	P = 0.04	65 (12.8%)	444	P < 0.0001
Used syringe	Yes	20 (23.3%)	66	OR = 7.43 CI = 3.14–16.56	60 (69.8%)	26	OR = 57.22 CI = 23.25–125.3
	No	15 (3.4%)	442	P < 0.0001	15 (3.4%)	422	P < 0.0001
Shaving	Yes	20 (13.2%)	132	OR = 122.2 CI = 6.80–3,183	45 (30%)	105	OR = 119.2 CI =6.23–18.66
	No	0	140	P < 0.0001	0	139	P < 0.0001
Tooth removal	Yes	15 (14%)	91	OR = 2.61 CI = 2.52–5.54	50 (47.2%)	56	OR = 13 CI = 7.02–23.32
	No	20 (4.8%)	397	P = 0.001	25 (6%)	392	P < 0.0001

BLOOD TRANSFUSION

Those who had received blood transfusions were at a substantially higher risk (OR = 6.02; CI: 4.41-13.55) of contracting HCV and HBsAg. Individuals who had received blood transfusions faced a significantly elevated risk of HCV transmission (OR = 13.77; CI: 7.13-26.15) (Table II).

NEEDLE INJURY

The occurrence of needle injuries was linked to an elevated risk of transmission (OR = 3.57; CI: 2.16-16.08). Needle injuries were strongly associated with an increased risk of HCV transmission (OR = 16.07; CI: 4.30-45.05) (Table II).

SYRINGE REUSE

Reusing syringes was identified as a significant risk factor for both HCV and HBsAg transmission (OR = 7.43; CI: 3.14-16.56). The reuse of syringes was a predominant risk factor for HCV transmission, demonstrating a substantial risk increase (OR = 57.22) (Table II). Moreover, a robust correlation between these factors and the risk of contracting HCV was observed.

RISK FACTORS BY PREVALENCE

Table III presents an array of risk factors associated with the transmission of hepatitis C, encompassing practices like drug injection, syringe utilization, unprotected sexual intercourse, blood transfusion, tattooing, dialysis, and medication administration. These risk factors are key contributors to the prevalence of hepatitis C. The prevalence of HBsAg and HCV in Nasirabad, Balochistan, Pakistan's general population, and the associations between sociodemographic factors. , risk factors linked to HBsAg and HCV and hepatitis C infection risk factors by population in Nasirabad, Balochistan, expressed as a percentage has been illustrated in Fig. 1 (a, b & c).

Table III. Risk factors for hepatitis C infection in Nasirabad, Balochistan, by population, presented as a percentage

Risk factors	Number of infected individuals	HCV infection rate %
Injection of drugs	60	36.6
Syringes	10	9.09
Razors	8	7.27
Unauthorized sexual encounters	12	10.9
Blood transfusion	10	9.09
Tattoo	2	1.8
Dialysis	3	2.7
Medication	5	4.5

Clinical Factors and HCV infection patients' p-value is presented in Table IV which displays biochemical parameters with statistically significant p-values, neutrophils, lymphocytes, platelets, total bilirubin, WBC count, alanine aminotransferase (ALT), alkaline phosphates, and hemoglobin (%).

Table IV. Biochemical parameter with normal range and their p-value

Biochemical Parameter	Normal Range	p-value
Neutrophils	45-70%	0.001
Lymphocytes	20-40%	0.001
Platelets	150,000-400,000 cells /uL of blood	0.001
Total Bilirubin	0.3-1.2 mg/dL	0.001
WBC Count	4,500-11,000 cells /uL of blood	0.001
Alanine Aminotransferase (ALT)	7-56 (U/L)	0.001
Alkaline Phosphatase	44-147 U/L	0.001
Hemoglobin (%)	13.5-17.5 g/dL	0.001

DISCUSSION

In this pioneering study, we conducted a comprehensive examination of the general population in Nasirabad, Balochistan, to determine the prevalence of HBsAg, HCV, and associated risk factors. Our findings revealed a notable prevalence of HCV at 14.4% and HBsAg at 6.7%. These figures align with previous reports from various cities in the Sindh Province, such as Karachi, Nousheroferoz, Larkana, and Thatta, which have shown HCV prevalence rates ranging from 3.2% to 25.1%. Remarkably, the HCV prevalence observed in our study is consistent with findings from other Pakistani cities, like Mardan, where HCV incidence was reported at 11.7%, and South Asian urban areas, which exhibited an HCV prevalence of 12.9% (21).

Our study's observations regarding the prevalence of HBsAg align with previous research conducted among high-risk populations, including hemodialysis patients, pregnant women, and barbers. These studies have reported HBsAg prevalence rates ranging from 2% to 7.5%. Notably, our investigation delved into the occurrence of hepatitis B and C infections concerning age and gender. It is plausible that older individuals may exhibit a higher disease frequency due to frequent visits to risky barbershops and beauty salons for haircuts and shaving (22).

Several studies in Pakistan have indicated that HBsAg and HCV exposure is more likely to occur in males. However, these findings require validation in larger, more diverse cohorts (23). It is essential to note that HBV is endemic in Pakistan, with a particularly high prevalence of 4.3% in Balochistan, as revealed by our study.

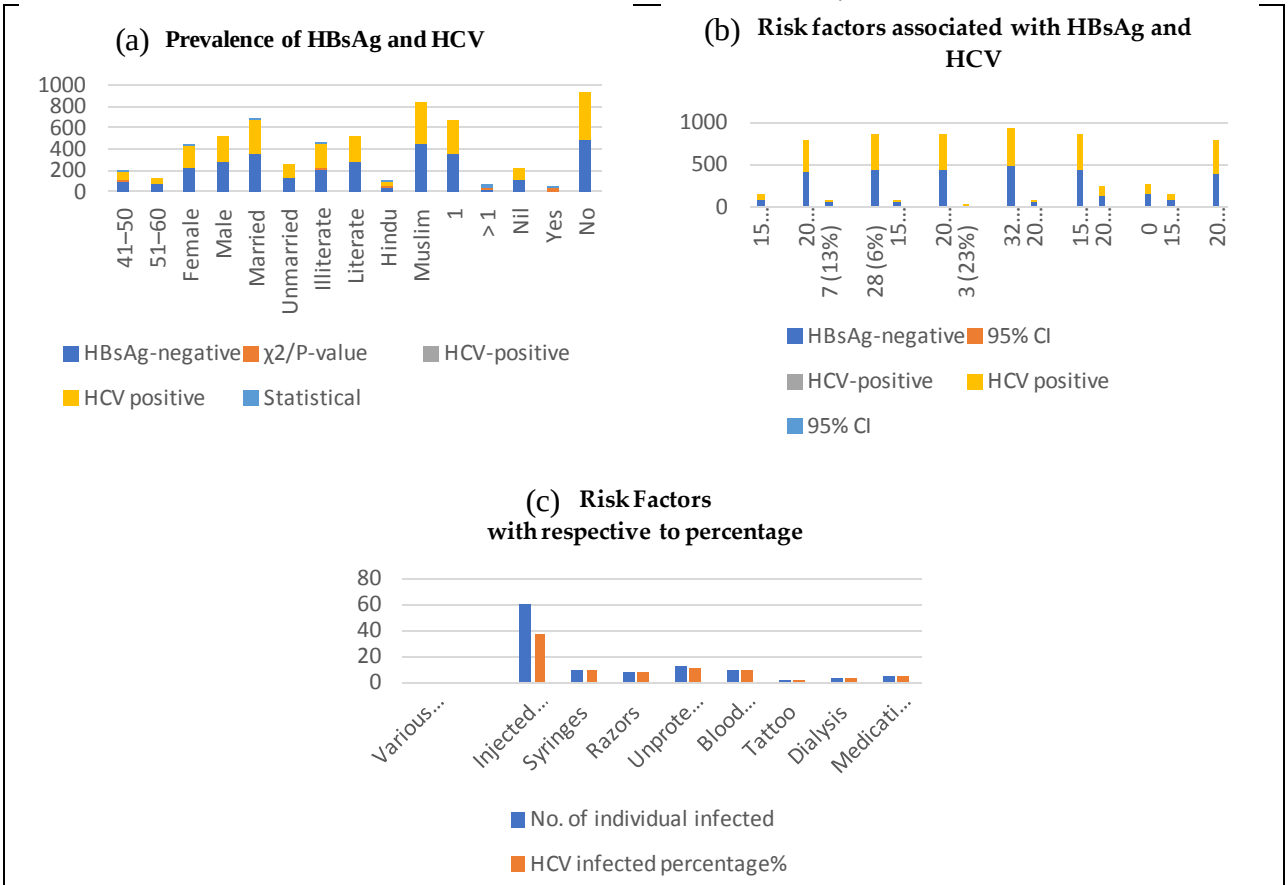

Fig. 1. (a) The prevalence of HBsAg and HCV in Nasirabad, Balochistan, Pakistan's general population, and the associations between sociodemographic factors. **(b)** Risk Factors Linked to HBsAg and HCV. **(c)** Hepatitis C infection risk factors by population in Nasirabad, Balochistan, expressed as a percentage

Worldwide, liver failure, cirrhosis, and hepatocellular carcinoma contribute to more than one million deaths annually; with 4,000 to 5,500 fatalities recorded each year in the United States. Our data highlighted a higher prevalence of HBsAg among illiterate participants compared to their literate counterparts. This suggests that hepatitis infections are more common in Pakistani adults than in children, although the younger population is not immune to its impact (24).

Our sociodemographic data indicated that married individuals were more likely to have hepatitis infections, with a proportion reporting that their spouses were also affected. This observation is in line with the findings of Razavi-Shearer et al. (2018), which suggested that three doses of the HBV vaccination might be inadequate to provide protection against hepatitis B. Despite this, Pakistan has an approved HBV vaccination with high immunogenicity and tolerability (25).

Epidemiological Implications for Nasirabad District: Based on our findings, it is conceivable that the Nasirabad district could face acute hepatitis B and hepatitis C epidemics among the adult population. This underlines the urgency of targeted public health interventions and vaccination campaigns to mitigate the impact of these infections.

CONCLUSION

In conclusion, our study sheds light on the prevalence of hepatitis B and C in Nasirabad, Balochistan, and underscores the importance of continued research, education, and intervention strategies to address this significant public health concern.

Hence, with the elevated prevalence of HBV and HCV in Nasirabad, the general population remains perpetually exposed to the risk of infection. To address this, there is a pressing need to enhance public awareness regarding the transmission routes of viral hepatitis and the available treatment modalities. Effective community-based health education campaigns must be meticulously planned and executed. Collaborative efforts involving healthcare professionals and specialized teams dedicated to combating infectious diseases can be pivotal. Moreover, fostering cooperation to develop novel and more potent anti-

infective medications and vaccines stands as a promising strategy to reduce the incidence and mortality associated with these diseases.

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