



## RESEARCH PAPER

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## Prevalence and their possible risk factors of hepatitis B Virus Infection in Bannu District of Khyber Pakhtunkhwa, Pakistan

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### Abstract

Hepatitis B infection is the leading health problem throughout the globe. It is estimated that worldwide, the prevalence of HBV is approximately 5 percent, but the percentage varies globally. During this study 300 blood sample were collected from district Bannu and analyzed in District Headquarter Hospital, Bannu. The entire blood samples were initially tested by Immunochromatographic (ICT) kits, and the ICT positive samples were further assayed by polymerase chain reaction (PCR). Out of total three hundred blood samples, three major areas were selected and analyzed for HBV infection; the highest prevalence was recorded in Bannu city on ICT kits was 14.44% followed by Kakki 10.78% and Domail 8.88%. By PCR, the greater percentage i.e. 9.44% of HBV Infection was reported in Bannu followed by other study areas, and as for as the age wise distribution was concerned, the highest percentage was 8.49% in age between 21-40. In general population of Bannu, the male community showed the highest prevalence (8.8%) and lowest percentage was recorded in females. The evaluation and examination of the Hepatitis B infection from Bannu revealed that the proportion of the disease increases with the increase in age.

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## Introduction

Hepatitis B infection is one of the main health problems throughout the globe (Rauf *et al.*, 2011; Afzal *et al.*, 2014). It is estimated that nearly two billion persons are infected HBV Globally (Zhu and Dong, 2009; Paraskevis *et al.*, 2010; Li *et al.*, 2010). Three hundred and fifty million people are the reported carrier of chronic HBV (Ali *et al.*, 2011; Mata *et al.*, 2012). Acute hepatitis is caused by this virus causing variable severity (Heermann *et al.*, 1999).

The percentage of HBV in children is 95% as compared to adults 10% (Bowyer and Sim, 2000). In the chronic stage, the presence of the virus leads to cirrhosis, hepatocellular carcinoma and lastly causes chronic liver disease (Mahoney and Kane, 1999). Pakistan Medical Research Council conducted a country-wide study from 2007-2008 that reports that incident of HBV is about 2.5 % in the overall population of Pakistan. Gender wise study indicated that majority of the males are affected with hepatitis B virus in general population of Pakistan (PMRC, 2009). In Baluchistan region of Pakistan, the frequency of HBV was reported very high which is 4.3%, whereas in Sindh it was 2.5%, in Punjab, it was reported 2.4%, and in Khyber-Pakhtunkhwa, it was 1.3% (Choudhary and Khan, 2005). In some districts of Punjab province such as Islamabad, Jhang, Okara, Vehari, Mandi Bha ud din, Attock, Gujranwala, Mianwali and Rahim Yar Khan, the prevalence of hepatitis B virus were found highest; where 2.9% prevalence of HBV was reported in district Gujranwala (PMRC, 2009).

The primary cause of transmission of Hepatitis B virus is mainly through serum, semen, saliva, blood and body fluids. As the virus can stay and live on the surfaces of razors, syringes, and tables (Workowski and Berman, 2002; Workowski, 2006).

The other known risk factors for HBV infection are the use of unsterilized surgical and dental tools, shaving from Barber, reuse of disposable injection and needles with drug follower, reuse of needle for ear piercing and nose are also. The HBV can be

transmitted by sharing personal kits like nail cutters, blades, and toothbrushes (Bukhari *et al.*, 1999). Generally, HBV does not need any cure, and in maximum individual, the infection is naturally clear without treatment (Hollinger and Lau., 2006). However, about one percent of patients need initial antiviral therapy (Lai and Yuen., 2007). The main study aim was to find out the prevalence of hepatitis B virus infection in the general population of district Bannu of KPK (Pakistan) and to suggest different precautionary measure for the control and management of HBV Infection.

## Materials and methods

All the experimental procedures were approved by the Ethics Committees of Department of Microbiology, Hazara university Mansehra Khyber Pakhtunkhwa. The informed consent was signed by the patients for participating in the study.

In this research study majority of the patients included males, females, and children.

History and demographic details of the patients were noted by using a structured questionnaire. The structured questionnaire consists of the major questions that include age, sex, location, socio-economic status any past history of the hepatitis and blood transfusion history, ear piercing, tattooing and frequent use of (IV) intravenous or (IM) intra muscular injections. Furthermore, past history of hepatitis B, any previous knowledge of Hepatitis B, its mode of transmission and measures taken were also included and recorded.

Three hundred (300) blood samples were collected from the patients who visited outpatient department (OPD) or admitted in District Head Quarter Hospital Bannu belonging to district Bannu. At least 5-6 ml of Blood was collected from the patients with the help of a sterile syringe and which was then centrifuged at the speed of 3000-4000rpm. The isolated serum samples were then stored at -20°C. For further analysis, two techniques were used i.e. ICT and PCR. ICT positive samples were further analyzed by PCR.

*Immunochromatographic technique*

With the help of dropper two (2) drops of serum (10 µl) were put on the ICT kit. After a minute duration, development of two lines on result bar of ICT device indicated an active HBV while development of a single line indicated HBV negative result.

*DNA Extraction and RT- PCR*

From the HBV positive ICT samples, the isolation of DNA and later RT-PCR were carried out with the help of DNA extraction, and RT-PCR kit from Sacace (Sacace,

Biotechnology, Italy) following the manufacturer provided procedures, inside the Cepheid SmartCycler (Nasdaq: CPHD, California, US).

*Statistical analysis*

All the data was analyzed by using Microsoft Excel 2010 to find out the percentage of different areas, and the data of questionnaire was analyzed by using Graph PadPrism version 5 portable. The p-value of less than 0.05 was considered to be significant.

**Results**

A total of 300 persons were screened for the presence of Hepatitis B virus. There were 215 (71.66%) males and 85 (28.33%) females from different areas of district Bannu Khyber Pakhtunkhwa. Out of total three hundred samples, 37 (12.33%) were ICT positive, and 21 (7%) were found positive by PCR as shown in Table 1.

**Table 1.** Prevalence of HBV.

Total Samples	ICT +ve	ICT %age	PCR +ve	PCR %age
300	37	(12.33%)	21	(7%)

*Area-wise distribution of HBV in district Bannu*

The whole district was decided into three clusters, Bannu, Kaki and Domail. The division of blood samples collected from major areas of District Bannu 180, Kaki 65 and Domail 45. The highest prevalence

on ICT Kit was observed in Bannu, i.e., 14.44%, followed by the percentage of Kaki 10.76% and Domail (8.88%) while on PCR 9.44% samples were reportedly positive from Bannu followed by Kaki 4.61% and Domail 2.22% as shown in Table 2.

**Table 2.** Area-wise distribution of HBV in district Bannu.

S. No.	Areas	Samples	ICT+ve %age	PCR+ve%age
1	Bannu	180	26 (14.44)	17 (9.44%)
2	Kaki	65	7 (10.76)	03 (4.61)
3	Domail	45	4 (8.88)	01 (2.22)
4	Total	300	37 (12.33)	21 (7)

*Age-wise distribution of HBV in district Bannu*

The individuals were categorized into three (3) age groups i.e. 10-20, 21-40 and 41-60 years. The prevalence rate of HBV was higher in age groups of

21-40 (8.49%) of total, 10-20 age groups showed comparatively low prevalence (i.e. 5.74%) whereas lowest prevalence was recorded in the age group ranging from 41-60 i.e. (5%) as shown in Table 3.

**Table 3.** Age-wise distribution of HBV in district Bannu.

Age Group	Samples	ICT +ve	ICT %age	PCR +ve	PCR %age
10-20	87	12	13.79%	05	5.74%
21-40	153	18	11.76%	13	8.49%
41-60	60	07	11.66%	03	05%
Total	300	37	12.33%	21	07%

*Sex- wise distribution of HBV in Bannu*

There were 215 (71.66%) males and 85 (28.33%) females. Out of total three hundred (300) samples, 37 (12.33%) were ICT positive, and remaining 21 (7%) were found positive by PCR as shown in Table 4.

*Assessment of different Risk factors for HBV**infection based on various factors collected/history during current study*

Table 5 signifies the circulation of HBV infected individuals in risk factors in this study.

By evaluating the distribution of cases with risk factors, our study confirmed the possible risk factors.

**Table 4.** Sex- wise distribution of HBV in Bannu.

S.No	Sex	Samples	ICT+ve	ICT%age	PCR+ve	PCR%age
1	Male	215	31	14.41%	19	8.8%
2	Female	85	6	7.05%	2	2.35%
3	Total	300	37	12.33%	21	7.33%

First the acquisition through almost crazy practices of shaving at Barber shops which account for approximately 37% of HBV in district Bannu with the P value of 0.0154 and relative risk of 0.4718 which is considered statistically significant. On the other side dental treatment which adds about 18.91%, HBV cases having the P value <0.0001 and relative risk of 0.1294. Which are statistically significant followed by blood transfusion (16.21%) which have the P value of 0.0672 and relative risk are 0.4739 which are not statistically significant. While the lowest percentage was observed in the risk groups of traveling abroad

(8.10%) P value is 0.2284, and relative risk is 0.5134 which are not statistically significant.

Those who get infected with needle prick were (2.70%); p-value is 0.1378, and relative risk is 0.2698 which are not statistically significant. Ear piercing (3.70%) P value is 0.0087, and relative risk is 0.1237 which are statically significant.

In the case of individuals with previous vaccination were 2.70% having a P value of 0.0467, with a relative risk of 0.1859 which are statically significant Table 5.

**Table 5.** Distributions of HBV Individuals in Risk Group.

S. No	Possible risk factors	+ve Cases	%age	P value	R.R
1	History of Blood Transfusion	6	16.21%	0.0672	0.4739
2	History of Vertical Transmission	Nil	0.00%	0.0224	0.0
3	History of Travelling Abroad	3	8.10%	0.2284	0.5134
4	History of Dental Treatment	7	18.91%	<0.0001	0.1294
5	History of Previous Vaccination	1	2.70%	0.0467	0.1859
6	History of Shaving from Barber	14	37.83%	0.0154	0.4718
7	History of Ear Piercing	1	3.70%	0.0087	0.1237
8	History of Needle Prick	1	2.70%	0.1378	0.2698
	Total	37	100%		

**Discussion**

Globally, the infection burden of HBV is increasing day by day, and its rate increases in developing countries, especially in Pakistan (Alam *et al.*, 2007a). While studying majority areas of Pakistan for HBV

infection, no such type of studies is available, most specifically related to Khyber-Pakhtunkhwa and federally administrated tribal areas. The percentage of Hepatitis B infection shows differences geographically throughout the country. Up till now,

about ten percent people were analyzed for the chronic liver disease and twenty percent of them have developed cirrhosis, and the remaining community remained as carriers.

The results showed that the prevalence of HBV infection is greater in Bannu (i.e. 7%) as compared to other district of KhyberPukhtoon Khwa Pakistan. The highest prevalence in Bannu district was assumed to be due to the low level of education and low economic status associated with the risk factors influenced this area. In our study, the male population residing in Bannu district were more infected 8.8% with Hepatitis B virus as compared to the female population 2.35%. In these areas, the involvement in blood transfusion practices, barber shaving, homosexuality and drug use are very common which strengthens the influences for such high prevalence of HBV in males. Our research showed that all of the people of different age groups residing at District Bannu were infected with Hepatitis B virus.

The recorded highest prevalence of HBV infection in this study was in the age group of 21-40 which was 13(8.49%). While in the age group 10-20, 5 (5.74%) persons, were infected with Hepatitis B virus and the lowest prevalence percentage of HBV was recorded in the age group of 41-60 (i.e. 5%). This result shows that on the incidence of Hepatitis B infection there was an age effect. The greater prevalence percentage in the age groups of 21-40 of HBV infection may be attributed to the greater contact to the risk factors and prolong Hepatitis B infection, and it may be due to the highest contact and relations with people as compared to the youngsters' and in old people. In Our study, we for the first time reported various types of risk factors for the infection of HBV infection in District Bannu.

The maximum percentage of 8.28% was observed due to the risk factor that most of the people residing in the specific area go to barber shops for shaving. Percentage of infection in individuals with the pre-history of dental treatment in our study was 5.69%. The transmission chances increase more since the

technician and the medical staff, due to lack of knowledge, don't use sterile and autoclaved apparatuses. Risk factor with a prior history of surgery and with a history of ear piercing contribute to 5.71% and 1.81% respectively. This may be possibly because of no such awareness and knowledge in the specified population about the possible risk factors from the healthcare providers. In our study, 6.89% of HBV infection occurred by the transfusion of blood. It is recommended for the government of Khyber-Pakhtunkhwa that they should install proper and state of the art diagnostics equipment at different DHQ hospitals in Khyber-Pakhtunkhwa. Appropriate and on time treatment should be available for that patient who are in the stage of chronic disease liver. Physician and health care workers must have the knowledge about injection and procedural safeties. Proper screening of blood donors must be done before transfusion to reduce the spread of HBV infection. To (or "intending to") minimizing the spread of HBV infection, the community must be educated through electronic, social and print Media, newspaper, arranging workshop and seminars throughout the country while most specifically in backward and uneducated regions.

### Conclusion

The study concludes that majority of the people living in Bannu district were affected by the infection of Hepatitis B virus, and its percentage varies among different age groups and between major areas of Bannu district. The screening of the general population in Bannu district revealed that the proportion of HBV infection increases with the increase in age, low HBV prevalence percentage was recorded in age below twenty years while a high percentage of the infection was in the individual's having age above twenty years. However, Hepatitis B Virus Infection percentage was greater in males as compared to females residing in district Bannu.

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**Competing interest**

The authors of this manuscript do not have any conflict and competing of interest.

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