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PREVALENCE OF HEPATITIS B VIRUS INFECTION AMONG BLOOD DONORS IN DISTRICT BAJAUR, KHYBER PAKHTUNKHWA, PAKISTAN

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Abstract

Hepatitis B virus (HBV) is a significant cause of chronic liver disease and hepatocellular carcinoma, particularly in low- and middle-income countries. The burden of HBV in Pakistan is among the highest in the world, and the recently merged tribal districts have little data. The aim of the study was to identify the prevalence of HBV in blood donors at the District Headquarters Hospital in the Pakistani province of Bajaur, Pakistan, and to determine its prevalence by age, sex, and tehsil (sub-district). The data of blood donors visiting the DHQ Hospital Bajaur blood bank during January and December 2024 was utilized to conduct a retrospective cross-sectional study. There were 260 donors (190 male 70 female). The screening of hepatitis B surface antigen (HBsAg) by enzyme-linked immunosorbent assay (ELISA) confirmed the presence of HBV infection, which was further confirmed by polymerase chain reaction (PCR). The prevalence was computed in general and according to demographic factors. Comparisons were done using chi-square and fisher exact tests. The overall prevalence of HBV was 6.92% (18/260; 95% CI: 4.2–10.7%). Males had significantly higher prevalence (8.43%, 16/190) than females (5.71%, 4/70) (p = 0.04; odds ratio = 1.52, 95% CI: 1.02–2.27). The age group with the greatest infection rate was the 30 to 40 years group among the males (11.1%), and the 20 to 30 years group among the females (10.5%). There was a considerable amount of geographic variation (2 = 14.8, 6, p = 0.02): Barang tehsil was the highest (12.0%), then Chamarkand and Utmankhel (10.0% each), with the lowest being Khar (2.5%). There was no significant correlation between the donor type (voluntary or replacement) and HBV positivity (p = 0.34). The prevalence of HBV in blood donors in District Bajaur (6.92%) is significantly greater than provincial and national prevalence, which shows in-between-high and high endemicity. The high geographic heterogeneity and higher rates in males and younger adults justify an immediate intervention in the form of public health, which encompasses increased donor screening, increased vaccine coverage, infection control, and community awareness campaigns that are specific to high-risk tehsils.

1. INTRODUCTION

Hepatitis B virus (HBV) is a partially double-stranded DNA virus, which is a member of the family Hepadnaviridae. It is a significant worldwide epidemic as it may lead to acute and chronic liver problems, such as hepatitis, cirrhosis, and hepatocellular carcinoma (HCC) [1]. The virus mostly targets hepatocytes resulting

in inflammation and gradual liver damage. Whereas acute infection disappears in a few people, a significant number of people become chronically infected, which can pass without any symptoms over decades, slowly leading to life-threatening complications. This silent carriage aids in the continued transmission and postpones the diagnosis and treatment.

HBV infection is a severe problem in the world despite the presence of an efficient vaccine. There are 257 million chronically infected HBV individuals worldwide, and some 887000 deaths in 2015 alone were due to HBV related liver diseases [2,3]. The distribution of the burden is not even. The Western Pacific Region (6.2%) and African Region (6.1%) have the highest prevalence of HBsAg according to the World Health Organization (WHO). It has a medium prevalence of 3.3% in the Eastern Mediterranean Region that encompasses Pakistan [4,5]. Such inequities are indicative of disparities in the socioeconomic status, health infrastructure, vaccination levels, and health literacy.

HBV is spread by contacting infected blood and body fluid. Unsafe medical practices (e.g., reuse of contaminated needles and syringes), transfusion of untested or poorly tested blood, mother-to-child infection at childbirth, unprotected sex, and exposure to contaminated equipment in non-medical places such as barbershops, dental clinics, and tattoo salons are common modes of transmission [6,7]. The weak enforcement of regulations, poor public awareness and poor infection control practices are some of the factors that escalate these risk factors in most of the developing countries such as Pakistan.

Pakistan has intermediate to high HBV endemicity. According to a recent systematic review and meta-analysis, the prevalence of HBV in the general population is about 6% (95% CI: 395% CI: 395% CI: 395% CI: 395percent CI: 395percent CI: 395percent CI: 395percent CI: 395percent C It is estimated that approximately 3.88 million Pakistanis have chronic hepatitis B [9]. Research based on healthy blood donors has provided evidence of HBV prevalence in the province of Khyber Pakhtunkhwa (KP) ranging between 1.95% and 2.51% [10,11]. These numbers, however, might be lower than the actual burden since blood donors are usually healthier than the general population [12].

District Bajaur is a tribal district that has recently been merged with KP and borders Afghanistan with a population of around 1.09 million (2017 census). The district is divided into seven tehsils; Mamund, Nawagai, Salarzai, Utmankhel, Khar, Chamarkand, and Barang. Historically, Bajaur has been socioeconomically marginalized, with inadequate development of infrastructure and access to quality healthcare. These issues have led to the existing gaps in disease surveillance and absence of sound epidemiological information on infectious diseases such as HBV. Evidence based health planning and blood safety require reliable local information. This paper thus sought to establish the rate of HBV infection among blood donors in the District Headquarters Hospital in Bajaur and to investigate the prevalence of HBV infection in relation to age, sex, and tehsil to make informed decisions regarding the appropriate strategies to be implemented by the local population in terms of their health.

2. Materials and Methods

2.1. Design and Setting of the studies.

It was a cross-sectional and retrospective study that was carried out in 2025 based on the data of the blood donors visiting the District Headquarters (DHQ) Hospital Bajaur blood bank during the period 1 January and 31 December 2025. The main government hospital in the district, which covers all seven tehsils, is the DHQ Hospital Bajaur. Its blood bank has well-organized data on the demographics of donors and screening outcomes, which is why it can be used in epidemiological analysis.

2.2. Population and Sampling of the study.

The study population comprised of all the blood donors who attended the DHQ Hospital Bajaur blood bank within the study period of 12 months. There were both voluntary and replacement donors. Completely demographic and lab data of 260 donors were enrolled. The sample size was not calculated by a priori power

concerns but the convenience (all eligible donors with full records during the study period) which is recognized as a limitation. Assuming the prevalence of HBV is expected to be 3% (average of blood donors in the province), a sample of 260 would give a margin of error about 2.1 at a 95% confidence level. Normal donor eligibility criteria were used: age 18–60 years, minimum body weight 50 kg, no known chronic or infectious diseases and negative history of high-risk behaviors according to the hospital policy.

2.3. Data Collection

To extract the data in the blood bank registry and laboratory records, a structured data collection form was used. The variables that were measured were as follows: age (1820, 2030, 3040, 4050, 5060 years), sex (male/female), tehsil of residence (Mamund, Nawagai, Salarzai, Utmankhel, Khar, Chamarkand, Barang), donor type (voluntary or replacement), and H Records with incomplete information and those with poorly-recorded laboratory results were only included. Missing or duplicated or ambiguous data in records were discarded.

2.4. Laboratory Methods

HBV infection was detected by hepatitis B surface antigen (HBsAg) and all blood donors were screened regularly. Early screening was conducted with a commercially prepared enzyme linked immunosorbent assay (ELISA; Wantai HBsAg ELISA Kit, Beijing Wantai Biological Pharmacy Enterprise Co., Ltd., China) as per the manufacturer guidelines. ELISA-repeated reactive samples were confirmed by real time polymerase chain reaction (PCR) of the HBV S gene (COBAS TaqMan HBV Test, Roche Molecular Systems, Inc., NJ, USA). This study classified HBV positive as only positive cases that were confirmed by PCR. High specificity and low false positives were achieved by this algorithm.

2.5. Inclusion and Exclusion Criteria

Criterion of inclusion included all the blood donors with full demographic details (age, sex, tehsil) and a clear HBV screening outcome (positive or negative by PCR confirmation) within the study period. The exclusion criteria were not complete records, duplication, records that did not have lab results, and cases where PCR confirmation was not done despite reactive ELISA (n = 2, excluded).

2.6. Statistical Analysis

The data were entered into Microsoft Excel (version 2019) and analyzed with the help of SPSS version 26.0 (IBM Corp., Armonk, NY, USA). HBV prevalence as percentages with 95% confidence interval (Clopper Pearson exact method) was calculated using descriptive statistics. The chi square test was used to compare categorical variables (sex, tehsil, donor type); the Fisher exact test when any of the expected cell counts was less than 5. Odds ratios (OR) with 95% confidence intervals were calculated for sex differences. A two-sided p value <0.05 was considered statistically significant. GraphPad Prism 9.0 was used to generate figures.

2.7. Ethical Considerations

The study was approved by the Institutional Review Board of the Riphah International University, Malakand Campus on 10 January 2025). The Medical Superintendent of DHQ Hospital Bajaur gave permission to access the anonymized patient records. All data were anonymized and individual patient consent was waived because the study was retrospective.

3. Results

3.1. Demographic Characteristics of the Study Population

A total of 260 blood donors were included in the analysis. Table 1 summarizes the demographic characteristics of the study population. The majority were male (73.1%, n = 190) and replacement donors (69.6%, n = 181). The highest proportion of donors was in the 20–30 years age group (37.3%, n = 97), followed by the 30–40 years group (27.7%, n = 72). The age-sex pyramid (Figure 1) shows that males outnumbered females in all age categories, with the widest gap in the 20–30 and 30–40-year groups.

Table 1. Demographic characteristics of blood donors in District Bajaur (N = 260).

Characteristic	Category	Frequency (n)	Percentage (%)
Sex	Male	190	73.1
	Female	70	26.9
Age group (years)	18–20	25	9.6
	20–30	97	37.3
	30–40	72	27.7
	40–50	42	16.2
	50–60	24	9.2
Donor type	Voluntary	79	30.4
	Replacement	181	69.6
Tehsil	Barang	50	19.2
	Chamarkand	30	11.5
	Utmankhel	20	7.7
	Mamund	35	13.5
	Nawagai	25	9.6
	Salarzai	30	11.5
	Khar	40	15.4

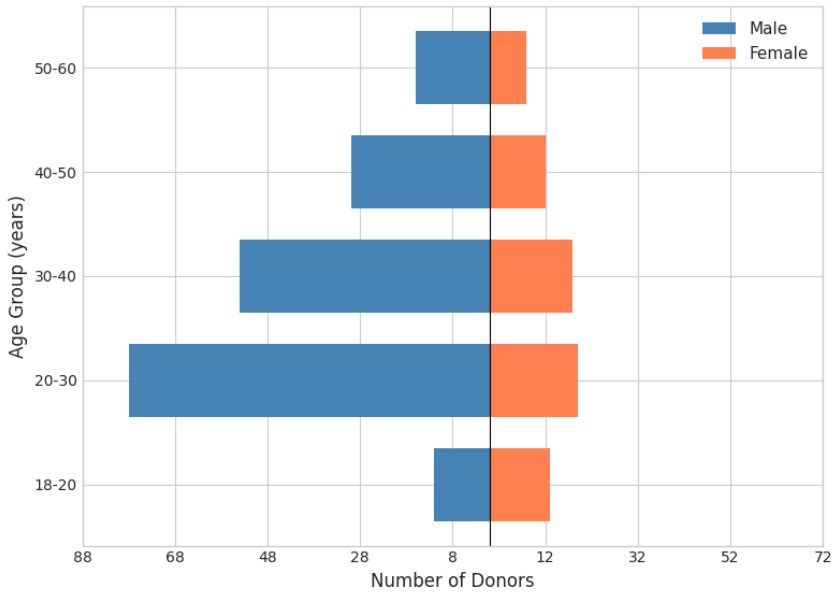

Figure 1 Age sex pyramid of blood donors in District Bajaur

3.2. Overall Prevalence of HBV

Among 260 blood donors, 18 tested positive for HBV by PCR confirmation, yielding an overall prevalence of **6.92**% (95% CI: 4.2–10.7%). The majority (93.08%, n = 242) were negative (Figure 2).

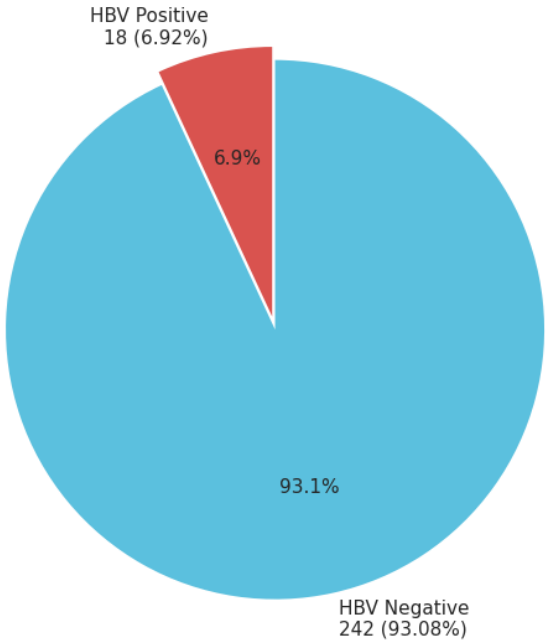

Figure 2 Overall prevalence of hepatitis B among blood donors in District Bajaur

3.3. Age- and Sex-Specific Prevalence

Table 2 presents the age- and sex-specific distribution of HBV infection. Among males, the highest prevalence was observed in the 30–40 years age group (11.1%, 6/54), followed by the 20–30 years group (9.0%, 7/78). No male donors aged 50–60 years tested positive. Among females, the peak prevalence occurred in the 20–30 years age group (10.5%, 2/19), followed by the 18–20 years group (7.7%, 1/13). No female donors over 40 years tested positive.

Overall, males had a significantly higher prevalence (8.43%, 16/190) than females (5.71%, 4/70) ($p = 0.04$, χ^2 test). The odds of HBV infection were 1.52 times higher in males (95% CI: 1.02–2.27). Figure 3 illustrates the age-specific prevalence by sex.

Table 2. Age- and sex-specific prevalence of HBV among blood donors in District Bajaur.

Age Group (Years)	Male Donors (n)	Male Positives (n)	Male Prevalence (%) [95% CI]	Female Donors (n)	Female Positives (n)	Female Prevalence (%) [95% CI]
18–20	12	0	0.0 (0.0–26.5)	13	1	7.7 (0.2–36.0)
20–30	78	7	9.0 (3.7–17.6)	19	2	10.5 (1.3–33.1)
30–40	54	6	11.1 (4.2–22.6)	18	1	5.6 (0.1–27.3)
40–50	30	3	10.0 (2.1–26.5)	12	0	0.0 (0.0–26.5)
50–60	16	0	0.0 (0.0–20.6)	8	0	0.0 (0.0–36.9)
Total	**190**	**16**	**8.43 (4.9–13.3)**	**70**	**4**	**5.71 (1.6–14.0)**

CI = confidence interval (Clopper-Pearson exact method).

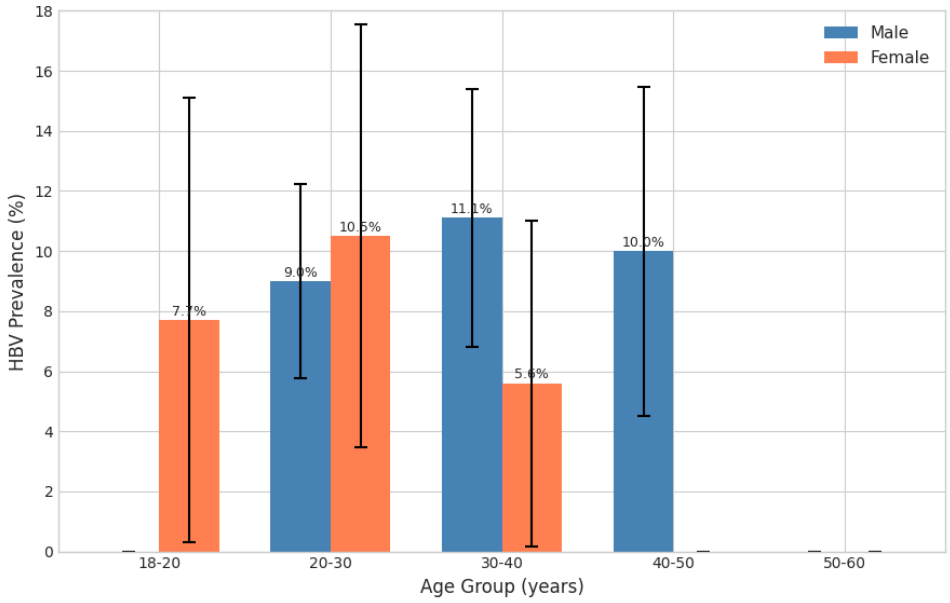

Figure 3 Age specific prevalence of hepatitis B among male and female blood donors in District Bajaur

3.4. Tehsil-Wise Distribution

HBV prevalence varied significantly across the seven tehsils (χ^2 = 14.8, df = 6, p = 0.02; Fisher's exact test p = 0.03). Table 3 shows the tehsil-wise distribution with 95% confidence intervals. The highest prevalence was in Barang (12.0%, 6/50), followed by Chamarkand (10.0%, 3/30) and Utmankhel (10.0%, 2/20). The lowest rates were in Khar (2.5%, 1/40) and Salarzai (3.3%, 1/30). Figure 4 displays the tehsil-wise prevalence with error bars representing 95% confidence intervals.

Table 3. Tehsil-wise prevalence of HBV among blood donors in District Bajaur.

Tehsil	Donors (n)	Positives (n)	Prevalence (%)	95% CI
Barang	50	6	12.0	4.6–24.3
Chamarkand	30	3	10.0	2.1–26.5
Utmankhel	20	2	10.0	1.2–31.7
Mamund	35	3	8.6	1.8–23.1
Nawagai	25	2	8.0	1.0–26.0
Salarzai	30	1	3.3	0.1–17.2
Khar	40	1	2.5	0.1–13.2
Total	**260**	**18**	**6.92**	**4.2–10.7**

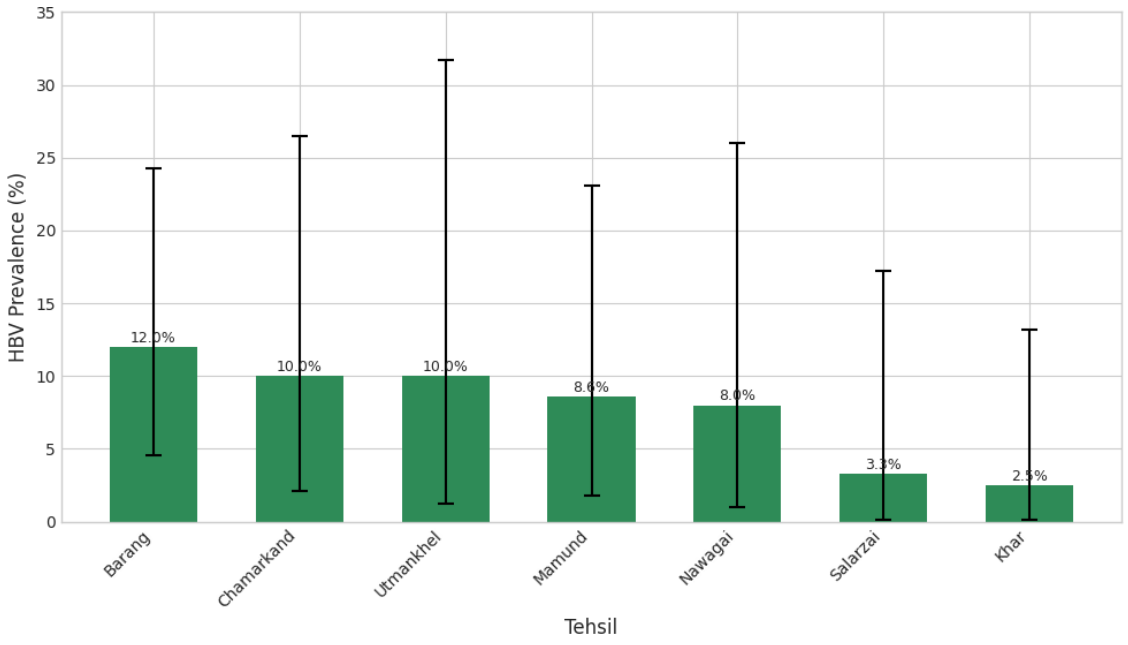

Figure 4 Tehsil wise prevalence of hepatitis B among blood donors in District Bajaur with 95% confidence intervals

3.5. Donor Type and HBV Prevalence

Replacement donors comprised 69.6% (181/260) of the sample. HBV prevalence was slightly higher among replacement donors (7.2%, 13/181) compared to voluntary donors (6.3%, 5/79); however, this difference was not statistically significant ($\chi^2 = 0.92$, $p = 0.34$).

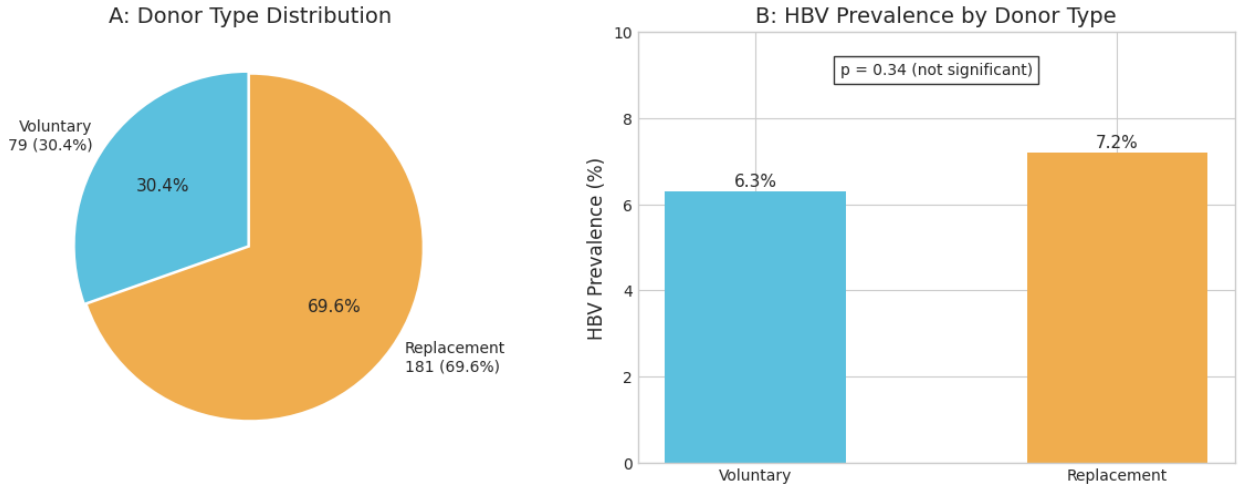

Figure 5 Donor type distribution and HBV prevalence. (A) Proportion of voluntary vs. replacement donors. (B) HBV prevalence by donor type

4. Discussion

This research presents the original estimations of HBV among blood donors in the previously under researched and socioeconomically disadvantaged areas of Pakistan (District Bajaur). The general prevalence of 6.92% (95% CI: 4.2 10.7) is the sign of intermediate to high endemicity as per WHO classification (>2% HBsAg prevalence). This is a significantly larger value than provincial reported values of Peshawar (0.861.95) [13,14] and the national blood donor prevalence of 2.4% [15] but is in line with the pooled national general population estimate of 6% (95% CI: 3-9) of a recent meta-analysis [8]. The prevalence is also similar to those recorded in high endemicity areas like sub–Saharan Africa [16] and among the Afghan refugees in Pakistan (8.3%) [17]. Such comparisons reveal the striking regional variability in the burden of HBV in Pakistan and the significance of local surveillance.

Sex differences: The prevalence is much higher in males (8.43% vs. 5.71% in females; OR = 1.52, p = 0.04) as compared to other global and regional sources [16,18]. This difference probably is a result of a mix of behavioral, professional and sociocultural aspects. The Bajaur situation is that men tend to migrate more to work, which may put them at risk of unsafe medical injections, non-sterile barbering, and other unregulated medical procedures, which are known to be risk factors of HBV in resource poor settings. Cultural barriers to women donating blood, on the other hand, could have led to selection bias; it could be that women who participated in this sample (n = 70), are no longer representative of women in the district. Therefore, the actual sex disparity in community prevalence might vary with what is found in blood donors.

Age distribution: The highest prevalence among males aged 30-40 (11.1) implies accumulating risk factors over the years. The fact that the previous high in females (2030 years, 10.5) could be explained by higher healthcare interaction in the reproductive years (e.g., antenatal care, childbirth) in which infection control is not optimal. The lack of cases in older age groups especially in females is probably caused by small samples sizes and low rates of blood donations among older people, not because they are not being infected.

Geographic heterogeneity: One of the most significant findings is the impressive difference in prevalence across tehsils (2.5% in Khar to 12.0% in Barang; p = 0.02). The most prevalent tehsil, Barang, shares a border with Afghanistan and there is common movement of populations across the borders. HBV prevalence in Afghanistan is estimated to be about 6 8% [19] and the cross-border transmission may be a contributing factor to the high prevalence of the same in Barang. Conversely, Khar - the district capital and administration hub - probably has a more developed healthcare system, increased immunization rates, and more general awareness. These geographical inequalities highly support tehsil specific interventions as opposed to district wide interventions.

Type of donor: The prevalence of replacement donors (7.2 vs. 6.3) was not significantly different than that of voluntary donors (p = 0.34). The replacement donors are historically thought to be at risk as they might be donating due to family pressure and fail to provide proper self-exclusion [20]. The non-significance of this study, however, could be due to limited statistical power (small sample size). However, the prevalence in both types of donors is high thus necessitating the need to conduct screening in a universal and stringent way regardless of the type of donor.

Limitations

There are a number of limitations to this study which must be taken into account when interpreting the results:

- Sample size: The sample of 260 donors is rather limited, which leads to broad confidence intervals of subgroup estimates (e.g., tehsil specific CIs). The research did not have the power to identify small differences and also to adjust the study multivariable.
- Single site design: Data were only collected in one hospital blood bank, which cannot be generalized on all blood donors and the population in general of the District Bajaur.

- Retrospective nature: The retrospective design made it impossible to collect individual level data on possible confounders and risk factors, including vaccination status, injections or surgeries, barber practices, incarceration and occupational exposures.
- Selection bias: Blood donors are a selected healthier sample of the population; thus, the prevalence reported here may be an underestimation of the actual burden to the community (chronically ill people are not eligible to become a blood donor) or an overestimation (high risk people may be overrepresented among the replacement donors). Also, the few female donors restrict extrapolation of sex specific results.
- Failure to differentiate stage of infection: HBsAg is not able to differentiate between acute and chronic infection or give information about viral load or infectivity.
- None of the vaccination data: we cannot determine the role of immunization in the patterns we observe without vaccination data.

Public Health Implications

Nevertheless, despite these constraints, the implications of the findings on policy and practice in District Bajaur and other similar contexts are evident. The prevalence of HBV among blood donors (almost 7 percent) means that there is an urgent need to:

Strengthen blood safety: Have all donated blood screened (preferably by nucleic acid testing) in a high sensitivity manner that is mandatory. At present, window period infections may be overlooked by relying on ELISA.

- Vaccination: Provide universal HBV infants vaccination (already included in EPI in Pakistan) and adolescents and high-risk adult catch up vaccination, especially in high prevalence tehsils such as Barang.
- Specific interventions in high-risk tehsils: Elaborate awareness, training on infection control and screening services on Barang, Chamarkand, and Utmankhel.
- Community awareness: Initiate culturally aware communal awareness campaigns on routes of transmission (barbering, injection safety, dental procedures) and stigma reduction.
- Surveillance: Have regular sentinel surveillance of blood donors and the high-risk populations to control trends and intervention effects.

Future studies ought to utilize larger, prospective studies using comprehensive risk factor questionnaires, incorporate community-based sampling to determine general population prevalence, and determine HBV genotypes and vaccination coverage.

5. Conclusions

This paper has shown that Hepatitis B virus infection has become a major public health issue among Blood donors in the District Bajaur with an overall prevalence of 6.92% - very high compared to provincial and national levels. It affects the males, younger adults (20-40 years) and those residing in border tehsils (especially Barang) disproportionately. The high geographic heterogeneity of the district highlights the fact that the district cannot be treated as a homogeneous geographic segment but instead there is need to implement tehsil specific interventions.

The immediate steps must be taken to increase blood donor screening, spread vaccination, increase infection control in healthcare and community environments, and initiate specialized awareness. This research offers the much-needed baseline information to guide such interventions, and it demands further surveillance and investigation in this underserved area.

Informed Consent Statement: Patient consent was waived because this was a retrospective study using anonymized laboratory and administrative data.

Data Availability Statement: The data presented in this study are available on reasonable request from the corresponding author.

Conflicts of Interest: The authors declare no conflicts of interest.

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