

Research Article

Seroprevalence of HIV, HBV, and HCV & Behavioral Factors among Truckers in Lahore, Pakistan: Implications for Targeted Public Health Intervention

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ABSTRACT

Background: Factors such as occupational mobility, long work hours, and less access to health services can affect transport workers' exposure to blood-borne infections and access to prevention services. The evidence based on serological screening, molecular detection, and behavioral traits in this occupational group is still sparse in Pakistan.

Objective: To assess the prevalence of HIV, hepatitis B virus (HBV) and hepatitis C virus (HCV) markers and their associations with sociodemographic, occupational, behavioural and health service related factors among the transport workers of Lahore, Pakistan.

Methods: A cross sectional study was conducted among 220 truck drivers and assistants at transport terminals Lahore, these were selected by convenience sampling. A structured questionnaire was used to obtain sociodemographic data and possible exposures history. HIV, hepatitis B surface antigen (HBs Ag), and anti-hepatitis C (anti-HCV) were tested in blood specimens, with polymerase chain reaction (PCR) performed on those that were screening-reactive. 95% Wilson confidence intervals were calculated for prevalence estimates. Associations with detectable viral nucleic acid were examined using crude odds ratios, Fisher's exact tests and Pearson chi-square tests.

Results: HIV RNA was found in nine (4.1%), HBV DNA in six (2.7%) and HCV RNA in 17 (7.7%) participants. Anti-HCV antibodies were detected in 23 participants (10.5%), including six without detectable HCV RNA. A total of 23 participants (10.5%) had at least one viral PCR positive, with 7 (3.2%) participants with multiple viral PCR positives. Factors associated with composite PCR detection included recent STI-related symptoms (OR: 7.25; $p < 0.001$), and being absent from work for more than 4 months (OR: 5.36; $p < 0.001$), and IDU (OR: 6.36; $p < 0.001$).

Conclusion: Transport workers were found to have detectable markers of HIV, HBV and HCV infection, overlapping viral detection occurred and a number of factors were associated with the detection of these viruses. The findings support targeted evaluation of confidential screening, HBV vaccination, infection-prevention education, and linkage-to-care services.

Keywords: Truck Drivers, Hiv, Hepatitis B Virus, Hepatitis C Virus, Molecular Detection, Occupational Health, Behavioral Risk Factors, Pakistan.

INTRODUCTION

Human immunodeficiency virus (HIV), hepatitis B virus (HBV), and hepatitis C virus (HCV) remain major public health concerns worldwide, contributing substantially to preventable morbidity, chronic liver disease, and premature mortality. Despite the great progress made in diagnostic tools, antiviral therapy, vaccination, and measures to prevent infection, there is a disproportionately heavy burden of these infections among populations facing difficulty accessing and maintaining care. Continued

transmission and poor clinical outcomes are due to delayed diagnosis, sub-optimal screening coverage, poor prevention approaches, and treatment interruptions. HIV and viral hepatitis continue to be major concerns for integrated disease surveillance, early diagnosis and effective referral to clinical care globally [1-3]. HIV, HBV and HCV have some of the same modes of transmission, but very different epidemiology, biological activity and interpretation for diagnosis. HIV is mainly transmitted sexually from contact with the

blood of HIV infected people and vertically through HIV transmission and HBV is sexually transmitted, perenterally, even perinatally. HCV is mainly transmitted by contact with infected blood, mainly by contaminated injection equipment and inadequately sterilized medical instruments [4,5]. These differences impact screening and for preventive interventions. A person with an active hepatitis B infection will likely have hepatitis B surface antigen (HBsAg) detectable and HBV DNA will give an indication of the amount of viral nucleic acid that is circulating. On the other hand, positivity for anti-HCV antibody is indicative of past or current exposure to HCV, and further molecular investigation is needed to determine the presence or absence of HCV viral RNA. It is important to differentiate between screening reactivity and molecular detection accordingly, and this is key to the interpretation of infection frequency and clinical management [6–9].

HIV prevention, surveillance and timely diagnosis is still a challenge and there is a substantial burden of viral hepatitis (specifically HCV) in Pakistan. The prevalence of infection could be different in population groups because of differential use of screening services, unsafe injection practices, knowledge of how infection is transmitted, and differences in health care use. Epidemiological studies from Pakistan have highlighted the need for quality laboratory testing, and adequate molecular testing and referral systems for everyone who comes back positive. The distribution of these infections can differ, however, depending on work-related factors, mobility, socio-economic factors and access to health care services [10,11].

Truck drivers and driver assistants are an occupational group with unique working conditions that could impact access to preventive care. Extended travel, unpredictable schedules, extended time away from home, and fewer opportunities for routine care can be obstacles to prompt screening, vaccination, counselling, and follow-up care. Occupational-specific health interventions have been studied in the past with transport workers in relation to their HIV knowledge, sexual behaviors, substance use, and access to preventive services. However, occupational mobility doesn't prove higher risk of infection or suggest an occupational route of infection. The epidemiology of observed infection frequencies needs to be detailed to differentiate them from possible behavioural and environmental correlates [12–15].

Previous research among long-distance truck drivers (LDT) has revealed that there are differences in knowledge of STIs, condom use, sexual relations, and access to preventive health services. Exposures to injection-related events, health care practices, and other situations that could be related to blood-borne infections have also been shown to have potential significance in research done in Pakistan as well as similar occupational environments [16,17]. However, there is limited research that has assessed HIV, HBV, and HCV test results together, molecular detection, viral coinfections, and a wide spectrum of behaviors and occupational risk factors among truck drivers in Lahore. Combining laboratory results with participant-reported history of exposures can help better understand the distribution of markers of infection and inform context-specific prevention strategies [18].

The current study was conducted to find out the frequency of HIV, HBV, and HCV screening reactivity and viral nucleic acid PCR positive in truck drivers and assistants enrolled from transport terminals in Lahore, Pakistan. Other aims were to describe single and multiple viral detection patterns, sociodemographic and occupational characteristics, self-reported sexual behaviors, substance use and healthcare-related exposures, and their crude associations with a composite viral PCR detection outcome. The results were to guide the evaluation of specific screening, immunization, infection prevention education, and linkage to care interventions for transport workers.

MATERIALS AND METHODS

Study Design and Setting

This cross-sectional study was conducted at major transport terminals in Lahore, Pakistan, from January 2025 to January 2026 and included 220 truck drivers and driver assistants. A non-probability convenience sampling approach was used to determine the prevalence of markers of HIV, HBV, and HCV infections and their association with selected sociodemographic, occupational, behavioral, and health care-related characteristics. At the time of enrolment, participants were asked questions in an interview and biological samples were collected. A standardized questionnaire was used to collect data, and laboratory investigations were conducted to detect serological reactivity for screening and the presence of viral nucleic acid. Reporting was done according to specific suggestions of the

Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) guidelines.

Study Population and Sampling

The study population consisted of 220 transport workers, selected from transport terminals of Lahore comprising 179 professional truck drivers and 41 driver assistants. Participants were selected using convenience sampling techniques, and they were informed to give their consent before participating in the study. Subjects were occupationally involved in truck driving activities and/or driver assistance and agreed to have questionnaire data and blood samples collected for laboratory investigations. The subjects ranged in age from 18 to 50 years, with a mean age of 34.9 ± 8.3 years. The analytical population included 220 individuals who had both questionnaire and viral screening data. When interpreting the applicability of the findings to the recruited occupational population, the non-probability sampling strategy was considered.

Sociodemographic and Behavioral Assessment

The information was collected through a structured questionnaire that included sociodemographic details, occupational history, sexual behaviours, drug and alcohol use, past infection history, health care practices and information on BBV transmission. Sociodemographic variables were age, gender, marital status, educational level, origin ethnicity, and known family history of HIV, HBV, HCV infection. Professional role, work-related travel, and absence from home were included as occupational information.

Sexual and reproductive health characteristics were defined as age at sexual debut, past exposure and number of sexual partners, exposure to commercial sex workers, condom use, sexual partnership dynamics, and sexual behaviors in the past 30 days, 12 months, and lifetime. Substance use information comprised cigarette smoking, tobacco use (chewing or naswar), alcohol consumption, any use of recreational drugs, specific drugs used, and any recreational injecting drug use. The exposures were related to health care and included previous therapeutic injections, intravenous drips, surgeries, tattooing, piercing, going to community barbers, and using unregistered health care providers. Participants were also asked about requesting sterile shaving blades and fresh syringes or needles.

The questionnaire also examined knowledge about HIV, HBV, HCV, and sexually transmitted infections (STIs); perceived routes of acquisition; history of viral testing; history of HBV vaccination; genitourinary symptoms; and STI treatment history. The behavioral and clinical history were collected using a self-reporting method. Items in the questionnaires that had conditional applicability were analyzed with the denominators of the applicable items, while other categorical variables were summarized with the total population of the study. If participants were able to indicate more than one behavior or exposure category, these responses were not mutually exclusive.

Biological Sample Collection and Laboratory Investigations

The enrolled participants had their blood samples taken and processed in the laboratory with a reflex testing algorithm for HIV, HBV, and HCV infection markers. The initial serological studies performed were for HIV, HBsAg, and anti-HCV. For those that were screening reactive, a follow-up molecular evaluation was performed with the polymerase chain reaction (PCR) test for the same viral nucleic acid. Laboratory records were obtained and utilized for classification of individual and overlapping viral detection, including HIV RNA, HBV DNA, and HCV RNA.

HIV RNA was documented in 9 subjects who were HIV screening reactive and HBV DNA was documented in 6 HBsAg reactive subjects. Of 23 individuals who were reactive to anti-HCV screening, 17 had detectable HCV RNA and 6 had undetectable HCV RNA. Screening reactivity and molecular detection were examined separately, especially with regard to HCV, which may have antibodies detected but no current viremia.

The primary molecular event was the detection of ≥ 1 positive HIV RNA, HBV DNA, or HCV RNA test. The participants were further divided into mutually exclusive groups by molecular detection (individual viral detection, dual viral detection, simultaneous detection of all three viruses, and no detectable viral nucleic acid). To prevent double counting persons with more than one detectable virus, the virus-specific detection categories were analyzed separately from the composite outcome. Laboratory results were recorded, and molecular detection was interpreted based on these results only and molecular detection was not used independently to determine the timing or route of infection acquisition.

Outcome Measures

The main results were the percentage of individuals who had markers for screening-reactive HIV, HBsAg and anti-HCV and the frequency of detectable HIV RNA, HBV DNA and HCV RNA. The composite primary molecular outcome was deemed to be the presence of at least one of the three viruses by nucleic acid detection. Single and multiple viral detections were assessed regarding their distribution for the purpose of characterizing overlapping infections.

Secondary outcomes were the prevalence of reported sexual and occupational behaviors, recreational substance use, injection drug use, STI-related symptoms, prior exposures to health care providers, awareness of an STI infection, vaccination history, and preventive practices. Exploratory analyses examined the relationship of these characteristics to the composite molecular detection result.

Statistical Analysis

IBM SPSS Statistics (version 26.0) was used for statistical analyses. Descriptive data for continuous data were presented as mean, standard deviation, and range, and categorical data were presented as frequencies and percentages. The prevalence of each viral screening and PCR- detection outcome was determined by dividing the number of participants who had the corresponding positive outcome by the 220 participants. Confidence intervals (CIs) for proportions were calculated using the Wilson score method and they are presented at 95% level.

Inferential analyses were conducted using the composite molecular outcome where participants had at least one positive viral PCR or no viral nucleic acid was detected. Cross-tabulations were performed to determine associations between categorical exposures and the composite PCR detection. Crude odds ratios (ORs) and 95% exact confidence intervals (CI) were computed for dichotomous comparisons, and two-sided Fisher's exact tests were used to test for statistical significance. Selected multicategory comparisons were made using Pearson Chi-square tests. Nominally statistically significant two-sided p-values were set at <0.05.

The reported associations were all unadjusted and exploratory. However, the composite PCR-positive group included 23 subjects, and several exposures were correlated thus precluding multivariable modeling. No attempt was made

to correct for multiple comparisons. Thus, the resulting ORs were considered as crude association measures and not as independent predictors of infection. Results of the statistical analyses were interpreted taking into account the cross-sectional nature of the study, the exposure variables at hand and the accuracy of the exposure estimates.

Ethical Considerations

The study was performed after a favorable opinion of an Institutional Review Board and in line with the Declaration of Helsinki ethical principles. All subjects gave written informed consent prior to the questionnaires and biological sample collection. Data collection, laboratory investigations, and statistical analysis were confidential and participation was voluntary. Information provided by the participants and laboratory results were kept confidential and results were made available only in aggregate without identifying the participants. The people who had positive molecular results were advised to seek the relevant health services for further clinical assessment, appropriate diagnostic evaluation, and management.

RESULTS

Study Population and Baseline Characteristics

A total of 220 transport workers were included in the study, comprising 179 professional truck drivers (81.4%) and 41 driver assistants (18.6%) recruited from major transport terminals in Lahore, Pakistan. The age range of the participants was 18-50 years (mean $\pm$ SD 34.9 $\pm$ 8.3 years). The largest occupational group was professional truck drivers, representing over 80% of the study population. In terms of gender, 218 participants (99.1%) were male, while two (0.9%) were in the questionnaire gender category "male/transgender". Most participants were married (153; 69.5%), followed by single (50; 22.7%), divorced (12; 5.5%), and widowed participants (5; 2.3%).

The majority of the respondents, (161, 73.2%) had an educational attainment that was not at the intermediate level. There 123 (55.9%) participants who completed matriculation, 38 (17.3%) completed primary education, 53 (24.1%) completed intermediate education and 6 (2.7%) participants completed education beyond intermediate. The study population belonged to various ethnic groups, with the majority being Punjabi (93, 42.3%), Pathan (47, 21.4%), Hazara (31, 14.1%), Kashmiri (21,

9.5%), Sindhi (15, 6.8%) and Baloch (13, 5.9%).

As for hepatitis B virus (HBV) vaccination, 72(32.7%) participants said they had been vaccinated before, and 148(67.3%) had never been vaccinated. 61 participants (27.7%) had a

family history of HIV, HBV or HCV infection. A significant cohort feature for preventive health was the relatively low reported rate of HBV vaccination. Detailed sociodemographic, occupational, educational, and vaccination characteristics are presented in Table 1.

Table 1. Sociodemographic, occupational, and selected background characteristics of transport workers (N = 220)

Characteristic	Frequency, n (%)
Age (years)	
Mean $\pm$ standard deviation	34.9 $\pm$ 8.3
Range	18–50
Occupation	
Professional truck drivers	179 (81.4)
Driver assistants	41 (18.6)
Gender	
Male	218 (99.1)
Combined male/transgender response category	2 (0.9)
Marital status	
Married	153 (69.5)
Single	50 (22.7)
Divorced	12 (5.5)
Widowed	5 (2.3)
Educational attainment	
Primary	38 (17.3)
Matriculation	123 (55.9)
Intermediate	53 (24.1)
Above intermediate	6 (2.7)
Ethnicity	
Punjabi	93 (42.3)
Pathan	47 (21.4)
Hazara	31 (14.1)
Kashmiri	21 (9.5)
Sindhi	15 (6.8)
Baloch	13 (5.9)
HBV vaccination history	
Previously vaccinated	72 (32.7)
No previous vaccination reported	148 (67.3)
Family history	
Family history of HIV, HBV, or HCV	61 (27.7)

Data are presented as frequencies and percentages unless otherwise indicated. Age is expressed as mean $\pm$ standard deviation and range. Percentages are based on the total study population (N = 220).

Prevalence of Viral Infection Markers

Initial serological screening identified reactivity for at least one HIV, HBV, or HCV marker in 28 of the 220 participants (12.7%). Repeat molecular testing showed positive molecular test among 23 participants (10.5%, 95% CI: 7.1–15.2%) while 197 (89.5%) had no molecular test done. The distribution of screening reactivity and molecular detection was different based on the viral marker evaluated as shown in Table 2.

Nine participants (4.1%, 95% CI: 2.2–7.6%) were reactive for HIV, and had detectable HIV RNA. Six participants (2.7%, 95% CI: 1.3–5.8%) were HBsAg-reactive and in all of these, HBV DNA was detectable. Hepatitis C had the highest prevalence of screening reactivity (23; 10.5% [95% CI: 7.1–15.2%]). But 17 participants (7.7%, 95% CI 4.9–12.0%) had HCV RNA in blood, showing a discrepancy between serological evidence of HCV exposure and detectable viremia.

Of the 23 patients who were anti-HCV reactive, 17 (73.9%) had detectable HCV RNA while 6 patients (26.1%) had undetectable HCV RNA. Therefore, anti-HCV screening reactivity was 2.7 percentage points higher than HCV RNA prevalence in the overall population. This difference highlights the need to differentiate anti-HCV seropositivity from molecular evidence

of ongoing viral replication. HIV RNA and HBV DNA detections frequency was comparable to their screening reactive frequency, while HCV showed lower molecular positivity compared to serological positivity frequency. Figure 1 shows the comparison between screening and molecular outcomes.

Table 2. Screening reactivity and PCR-detectable viral markers for HIV, HBV, and HCV among transport workers (N = 220)

Viral marker	Screening reactive, n (%)	95% CI (%)	PCR detectable, n (%)	95% CI (%)
HIV / HIV RNA	9 (4.1)	2.2–7.6	9 (4.1)	2.2–7.6
HBsAg / HBV DNA	6 (2.7)	1.3–5.8	6 (2.7)	1.3–5.8
Anti-HCV / HCV RNA	23 (10.5)	7.1–15.2	17 (7.7)	4.9–12.0

Data are presented as frequencies, percentages, and 95% Wilson confidence intervals.

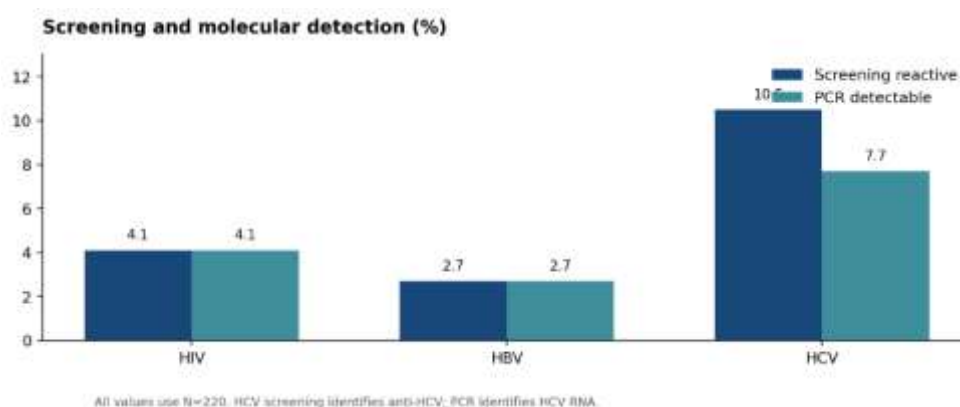


Figure 1. Comparison of serological screening reactivity and PCR-detectable HIV, HBV, and HCV among 220 transport workers. Anti-HCV reactivity was recorded in 10.5% of participants, whereas HCV RNA was detectable in 7.7%

Distribution of Single Viral Detection and Coinfections

Molecular testing demonstrated both individual viral detection and concurrent detection of multiple viruses. Among the 23 participants with at least one detectable viral PCR result, 16 (69.6%) had a single virus detected, five (21.7%) had two detectable viruses, and two (8.7%) demonstrated simultaneous detection of all three viruses. Overall, multiple viral detection was recorded in seven participants, corresponding to 30.4% of those with detectable viral nucleic acid and 3.2% of the entire study population.

HCV RNA alone represented the most frequently observed molecular detection pattern, occurring in 11 participants (47.8% of PCR-positive individuals; 5.0% of the cohort). HBV DNA alone was detected in three participants (13.0% of PCR-positive

individuals), while HIV RNA alone was detected in two (8.7%). Among participants with multiple detectable viruses, concurrent HIV RNA and HCV RNA detection occurred in four individuals (17.4% of PCR-positive participants), whereas combined HIV RNA and HBV DNA detection occurred in one individual (4.3%). Two participants (8.7%) demonstrated simultaneous HIV RNA, HBV DNA, and HCV RNA detection. No participant exhibited the HBV–HCV dual-detection pattern without concurrent HIV detection.

The detailed distribution of mutually exclusive molecular detection categories is presented in Table 3. These findings distinguish the overall frequency of participants with detectable viral nucleic acid from virus-specific detection frequencies, which overlap because of coinfections.

Table 3. Mutually Exclusive Molecular Detection Patterns Of HIV, HBV, and HCV among Transport Workers (N = 220).

PCR detection pattern	Frequency (n)	Percentage (%)
No detectable viral nucleic acid	197	89.5
HIV RNA only	2	0.9
HBV DNA only	3	1.4
HCV RNA only	11	5.0
HIV RNA + HCV RNA	4	1.8
HIV RNA + HBV DNA	1	0.5
HBV DNA + HCV RNA only	0	0.0
HIV RNA + HBV DNA + HCV RNA	2	0.9
Total	220	100.0

The molecular detection categories are mutually exclusive. Sixteen participants had exactly one virus detected, five had two viruses detected, and two had all three viruses detected. Overall, 23 participants (10.5%) had at least one detectable viral PCR result, including seven (3.2%) with multiple detectable viruses.

Sexual and Occupational Behavioral Characteristics

Evaluation of self-reported sexual behaviors showed high variability in sexual partnership, condom use, and interaction with commercial sex workers. Among 170 participants with applicable responses to the question concerning sexual partners beyond their spouse, 136 (80.0%) reported additional sexual partnerships. 146 (66.4%) participants reported a history of visiting a commercial sex worker while 145 (65.9%) had done so in the last 12 months. Moreover, 65.5% (144) of the participants were categorized in SPSS analysis as having multiple commercial sex worker contacts.

Nonuse was frequently cited with 138 participants (62.7%) saying they never used a condom during sexual intercourse. 99

participants (45.0%) reported sexual contact with both men and women. In respect of age of sexual initiation, 79 participants (35.9%) said they had had their first sexual experience at between 14 and 16 years of age. Table 4 provides the frequency of the distribution of selected sexual behaviors, which are shown in Figure 2.

The reported duration of absence from home was used to measure occupational mobility. Fifty-eight participants (26.4%) were classified as having spent more than four months away from home during the reference period. These are the occupational and behavioral characteristics of the study participants' exposure patterns. This was the case because multiple behaviors were self-reported by respondents and each individual exposure category was not mutually exclusive.

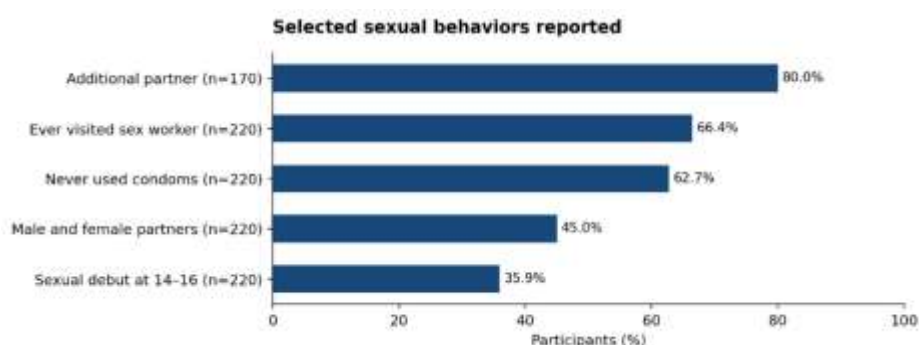


Figure 2. Distribution of selected self-reported sexual behaviors among transport workers. The additional-partner question included 170 applicable responses; the remaining displayed items used the total sample of 220 participants

Substance Use and Injection-Related Exposures

Substance-use assessment demonstrated a high frequency of reported tobacco and recreational drug consumption. Cigarette smoking was recorded in 189 participants (85.9%), making it the most frequently

reported tobacco-related behavior. Tobacco or naswar chewing was reported by 62 participants (28.2%), whereas alcohol consumption was reported by 69 (31.4%). Overall, 147 participants (66.8%) reported using at least one recreational drug.

Cannabis or hashish was the most frequently reported recreational substance, with 144 participants (65.5%) acknowledging its use. Heroin or opium use was reported by 73 participants (33.2%), followed by cocaine or crash use in 29 (13.2%) and diazepam or Valium use in three (1.4%). The reported substance-use categories overlapped because individual participants could report consuming more than one substance. The distribution of major substance-use behaviors is illustrated in Figure 3 and summarized in Table 4.

Regarding injection-related exposures, 31 participants (14.1%) reported recreational injection drug use during the preceding year. Therapeutic injections or intravenous drips during the same period were reported by 142 participants (64.5%). These findings characterize injection-related practices within the study population and provide the descriptive basis for subsequent analyses of their associations with viral PCR detection.

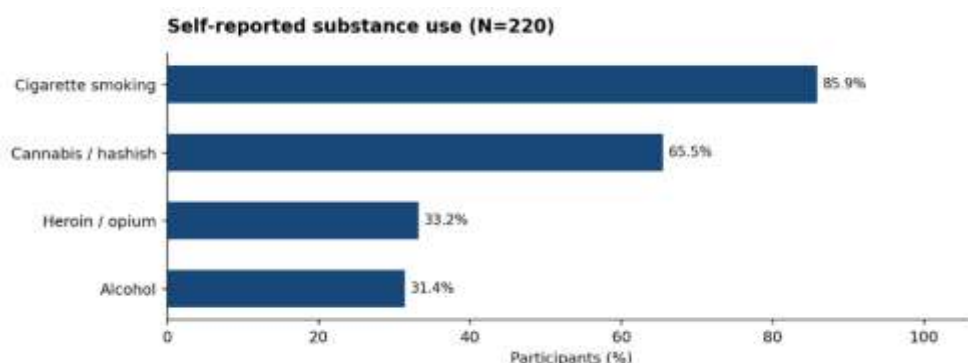


Figure 3. Frequency of selected self-reported substance-use behaviors among 220 transport workers, including cigarette smoking, cannabis/hashish, heroin/opium, and alcohol consumption. Multiple substance-use responses were possible

Sexually Transmitted Infection History and Healthcare-Related Exposures

A significant number of respondents had a history of genitourinary symptoms or STI treatment. One hundred forty (51.8%) participants reported symptoms that could have been attributable to an STI in the past year, and 98 (44.5%) reported previous treatment for an STI. The single condition reported in the questionnaire was UTI, which was the most common condition, with 89 participants (40.5%). This was followed by genital herpes in 26 (11.8%), syphilis in 18 (8.2%), chlamydia in 16 (7.3%), and gonorrhea in 15 (6.8%). These frequencies are based on self-reports and not on lab verification of the prevalence of the individual conditions. Figure 4 shows the distribution of selected STI-related histories. Potential exposures in the healthcare field and in personal grooming were also reviewed. 217

participants (98.6%) reported seeing unregistered health care providers. 46 subjects (20.9%) reported previous surgical procedures, 22 (10.0%) reported a history of tattooing or tribal marking and 32 (14.5%) reported previous piercing of ears or nose.

In terms of preventive measures, 31 (14.1%) reported that they did not ask for a new blade for shaving. Of the 217 respondents who had completed a valid questionnaire on syringe use, 33 (15.2%) said they did not ask for a new syringe/needle at health care visits. These findings are representative of the potential blood exposure situations and preventive practices as reported by those surveyed. Table 4 presents combined sexual, occupational, substance use, clinical, and healthcare-related characteristics.

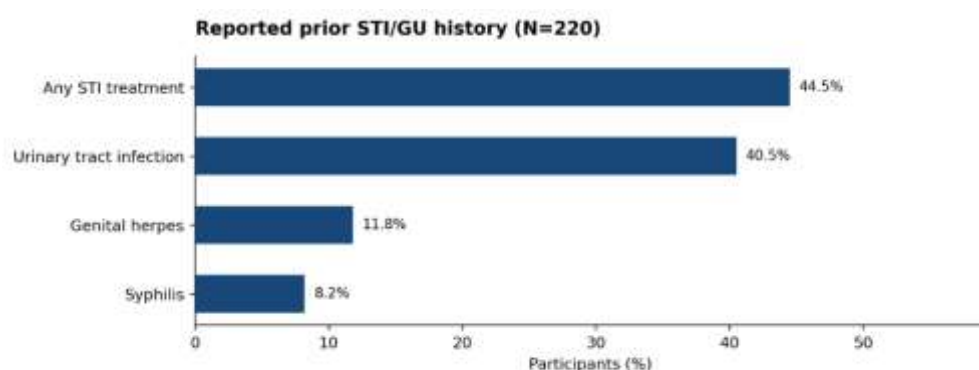


Figure 4. Frequency of participant-reported previous STI treatment and selected genitourinary conditions among 220 transport workers

Table 4. Self-reported sexual, occupational, substance-use, STI-related, and healthcare-exposure characteristics among transport workers (N = 220)

Characteristic	n/N	Percentage (%)
Sexual and occupational characteristics		
Additional sexual partner beyond spouse	136/170	80.0
Ever visited a commercial sex worker	146/220	66.4
Commercial sex worker contact in previous 12 months	145/220	65.9
Never uses condoms	138/220	62.7
Both male and female partners reported	99/220	45.0
Sexual debut between 14 and 16 years	79/220	35.9
More than one commercial sex worker (SPSS grouping)	144/220	65.5
More than four months away from home	58/220	26.4
Substance-use characteristics		
Cigarette smoking	189/220	85.9
Tobacco/naswar chewing	62/220	28.2
Alcohol consumption	69/220	31.4
Any recreational drug use	147/220	66.8
Cannabis/hashish	144/220	65.5
Heroin/opium	73/220	33.2
Cocaine/crash	29/220	13.2
Diazepam/Valium	3/220	1.4
Recreational injection drug use in previous year	31/220	14.1
Therapeutic injection or drip in previous year	142/220	64.5
STI-related characteristics		
STI-related symptoms in previous year	114/220	51.8
Previous STI treatment	98/220	44.5
Previous urinary tract infection	89/220	40.5
Previous genital herpes	26/220	11.8
Previous syphilis	18/220	8.2
Previous gonorrhea	15/220	6.8
Previous chlamydia	16/220	7.3
Healthcare-related characteristics		
Did not request a fresh shaving blade	31/220	14.1
Visited an unregistered healthcare provider	217/220	98.6
Did not request a fresh syringe/needle	33/217	15.2
Previous surgical procedure	46/220	20.9
Previous tattoo or tribal marking	22/220	10.0
Previous ear or nose piercing	32/220	14.5

Infection Awareness, Testing History, and Preventive Practices

The questionnaire assessment demonstrated widespread recognition of HIV/AIDS and viral

hepatitis. All 220 participants (100.0%) reported having heard of HIV/AIDS or HBV/HCV, while 219 (99.5%) acknowledged that certain infections could be transmitted through sexual contact. Correct condom use was identified as protective in the combined infection-awareness question by 143 participants (65.0%), whereas 76 (34.5%) selected an uncertain response.

There was often a knowledge of the selected blood borne transmission routes. 217 (98.6%) identified previously used needles, blades, or syringes as a possible vehicle of infection transmission, and 216 (98.2%) identified infected blood transfusion as another vehicle of transmission. However, 119 respondents (54.1%) suggested that infection could spread via the sharing of food. Furthermore, 217 (98.6%) identified that infection was not transmitted through handshaking,

demonstrating differences in knowledge across the various transmission-related questions.

Previous HIV, HBV, or HCV testing during the questionnaire-defined period was reported by 105 participants (47.7%). Awareness of sexually transmitted infections was reported by 217 participants (98.6%). But, only 72 (32.7%) participants had a history of HBV vaccination. Regarding preventive practices, 189 participants (85.9%) reported requesting a fresh shaving blade, while 184 of 217 applicable respondents (84.8%) reported requesting a fresh syringe or needle during healthcare encounters. Recognition that an apparently healthy individual could have an infection was recorded in 84 participants (38.2%). The distribution of infection awareness, testing history, reported symptoms, and preventive practices is summarized in Table 5.

Table 5. Infection awareness, previous testing, selected symptoms, and preventive practices among transport workers (N = 220).

Questionnaire response	n/N	Percentage (%)
Infection awareness and transmission knowledge		
Heard of HIV/AIDS or HBV/HCV	220/220	100.0
Aware that some infections can be sexually transmitted	219/220	99.5
Identified correct condom use as protective	143/220	65.0
Uncertain about condom protection	76/220	34.5
Believed infection could be transmitted through food sharing	119/220	54.1
Identified handshaking as a nontransmission route	217/220	98.6
Recognized transmission through previously used needles/blades/syringes	217/220	98.6
Recognized transmission through infected blood transfusion	216/220	98.2
Recognized that apparently healthy individuals may have an infection	84/220	38.2
Heard of sexually transmitted infections	217/220	98.6
Previous testing and vaccination		
Previous HIV/HBV/HCV testing within the specified questionnaire period	105/220	47.7
Previous HBV vaccination	72/220	32.7
Selected clinical and exposure history		
Previous blood transfusion	31/220	14.1
Genital ulcer symptoms in previous 12 months	30/220	13.6
Frequent painful urination	111/220	50.5
Genital itching	39/220	17.7
Groin swelling	11/220	5.0
Preventive and healthcare practices		
Visited a community barber	220/220	100.0
Visited an unregistered healthcare provider	217/220	98.6
Requested a fresh shaving blade	189/220	85.9
Requested a fresh syringe/needle	184/217	84.8

Data are presented as frequencies and percentages. Most denominators were 220; the fresh-syringe question included 217 applicable participants. Knowledge-related responses were assessed using the questionnaire's original combined HIV/HBV/HCV wording, despite differences in the transmission pathways of these viruses.

Bivariate Associations with Detectable Viral PCR Results

Exploratory bivariate analyses assessed relationships between sociodemographic, occupational, sexual, substance use, and STI-related symptoms and exposures and the composite outcome of detectable HIV RNA, HBV DNA, or HCV RNA. 220 subjects had at least one viral PCR detected (10.5%) and 197 (89.5%) had no detectable viral nucleic acid. Dichotomous comparisons were made using crude odds ratios (ORs), exact 95% confidence intervals (CIs) and two-sided Fisher's exact tests. The available Pearson chi-square tests were used for the evaluation of multicategory variables. Table 6 includes all 36 bivariate comparisons reported, including statistically significant, nonsignificant and non-estimable comparisons.

Sociodemographic and Occupational Associations

There was a higher prevalence of composite PCR detection in participants with lower than intermediate education compared to those with intermediate or higher education (21/161 [13.0%] versus 2/59 [3.4%]; OR: 4.25; 95% CI: 0.99–38.62; $p = 0.045$). The frequency of viral PCR detection was comparable between professional truck drivers and driver assistants (19/179 [10.6%] versus 4/41 [9.8%]; OR: 1.10; 95% CI: 0.34–4.70; $p = 1.000$).

There was an increased rate of composite PCR detection with those who were absent from work for more than four months, as compared to those who were absent for less than four months (14/58 [24.1%] versus 9/162 [5.6%]; OR: 5.36; 95% CI: 2.01–15.05; $p < 0.001$). However, absence exceeding two months was not significantly associated with PCR detection (19/171 [11.1%] versus 4/49 [8.2%]; OR: 1.40; 95% CI: 0.44–5.97; $p = 0.791$).

Associations with Sexual Behaviors

Composite PCR detection was statistically significantly associated with several sexual behaviours based on unadjusted associations. The proportion of PCR-positive participants in the group of participants who reported contact with more than one commercial sex worker was higher than in the reference group (21/144 [14.6%] versus 2/76 [2.6%]; OR: 6.28; 95% CI: 1.47–56.79; $p = 0.005$). Similarly, more frequent stops for commercial sex were associated with higher PCR detection (20/133 [15.0%] versus 3/87 [3.4%]; OR: 4.93; 95% CI: 1.39–26.73; $p = 0.006$).

Participants reporting never using condoms demonstrated a higher frequency of composite PCR detection than the comparison group (19/138 [13.8%] versus 4/82 [4.9%]; OR: 3.10; 95% CI: 0.98–13.00; $p = 0.041$). Sexual debut before 18 years was also associated with PCR detection (22/169 [13.0%] versus 1/51 [2.0%]; OR: 7.44; 95% CI: 1.14–314.90; $p = 0.020$). Receptive anal intercourse during the preceding year demonstrated an association with composite PCR detection (19/131 [14.5%] versus 4/89 [4.5%]; OR: 3.59; 95% CI: 1.13–15.03; $p = 0.023$). Similarly, vaginal intercourse outside marriage was associated with a higher frequency of PCR detection (21/151 [13.9%] versus 2/69 [2.9%]; OR: 5.38; 95% CI: 1.25–48.73; $p = 0.016$). The SPSS-defined variable describing all three forms of sexual partners demonstrated a marked difference in PCR detection (9/18 [50.0%] versus 14/202 [6.9%]; OR: 13.10; 95% CI: 3.94–44.45; $p < 0.001$). This is a composite variable which is a group of items from the original questionnaire and therefore interpretation is somewhat limited by the operational classification available.

Substance Use and Injection-Related Associations

Alcohol consumption was associated with a higher frequency of composite viral PCR detection (15/69 [21.7%] versus 8/151 [5.3%]; OR: 4.92; 95% CI: 1.84–14.22; $p < 0.001$). Participants reporting any recreational drug use also demonstrated higher PCR detection than those without reported drug use (20/147 [13.6%] versus 3/73 [4.1%]; OR: 3.66; 95% CI: 1.03–19.89; $p = 0.035$). Recreational injection drug use demonstrated one of the more pronounced unadjusted associations. Composite PCR detection occurred in 10 of 31 participants (32.3%) reporting injection drug use compared with 13 of 189 (6.9%) without this reported exposure (OR: 6.36; 95% CI: 2.21–18.06; $p < 0.001$). Therapeutic injection or intravenous drip exposure was also associated with higher PCR detection (21/142 [14.8%] versus 2/78 [2.6%]; OR: 6.55; 95% CI: 1.53–59.26; $p = 0.005$).

However, there was no statistically significant difference between the composite PCR detection rates for cigarette smoking (22/189 [11.6%] versus 1/31 [3.2%]; $p = 0.213$). In the unmatched comparisons, there were no statistically significant associations found for cannabis/hashish, heroin/opium,

cocaine/crash, diazepam/Valium or tobacco/naswar chewing (Table 6).

Healthcare Practices and STI-Related Symptoms

Individuals with STI symptoms over the past year had a higher composite PCR detection rate than those who did not have STI symptoms (20/114 [17.5%] versus 3/106 [2.8%], OR: 7.25; 95% CI: 2.05–39.31; $p < 0.001$). As for shaving, the PCR was more frequently detected in those who did not request to change the blade (10/31 [32.3%] compared to the

reference group [13/189 [6.9%]; OR: 6.36; 95% CI: 2.21–18.06; $p < 0.001$). In the SPSS-derived fresh-syringe comparison, PCR detection occurred in 10 of 35 participants (28.6%) in the nonrequest group compared with 13 of 185 (7.0%) in the reference group (OR: 5.23; 95% CI: 1.85–14.57; $p < 0.001$). There was no evidence of a significant link between the composite PCR detection and prior tattooing or surgical interventions or ear or nose piercing, or refusal to ask for a new piercing needle. Table 6 provides the overall comparative results.

Table 6. Bivariate associations of sociodemographic, occupational, behavioral, and healthcare-related characteristics with detectable HIV, HBV, or HCV PCR among transport workers (N = 220)

Variables	PCR-positive, exposed (n/N)	PCR-positive, reference (n/N)	Crude OR (95% CI)	p-value
Sociodemographic characteristics				
1. Education below intermediate level	21/161	2/59	4.25 (0.99–38.62)	0.045
2. Combined male/transgender category vs male	1/2	22/218	8.74 (0.11–702.69)	0.199
3. Non-Muslim vs Muslim	1/11	22/209	0.85 (0.02–6.51)	1.000
4. Punjabi vs other ethnicities	7/93	16/127	0.57 (0.19–1.53)	0.269
5. Pathan vs other ethnicities	7/47	16/173	1.71 (0.56–4.77)	0.284
Sexual and reproductive behaviors				
6. Sexual partner among unmarried (SPSS grouping)	5/34	18/186	1.61 (0.43–4.97)	0.367
7. All three partner forms (SPSS grouping)	9/18	14/202	13.10 (3.94–44.45)	<0.001
8. Sexual debut before 18 years	22/169	1/51	7.44 (1.14–314.90)	0.020
9. More than one commercial sex worker	21/144	2/76	6.28 (1.47–56.79)	0.005
10. Frequent stops for commercial sex	20/133	3/87	4.93 (1.39–26.73)	0.006
11. Never uses condoms	19/138	4/82	3.10 (0.98–13.00)	0.041
Occupational characteristics				
12. Truck driver vs driver assistant	19/179	4/41	1.10 (0.34–4.70)	1.000
13. Away from home for more than two months	19/171	4/49	1.40 (0.44–5.97)	0.791
14. Away from home for more than four months	14/58	9/162	5.36 (2.01–15.05)	<0.001
Substance use				
15. Alcohol consumption	15/69	8/151	4.92 (1.84–14.22)	<0.001
16. Cigarette smoking	22/189	1/31	3.94 (0.59–168.46)	0.213

17. Tobacco/naswar chewing	8/62	15/158	1.41 (0.49–3.79)	0.468
18. Any recreational drug use	20/147	3/73	3.66 (1.03–19.89)	0.035
19. Cannabis/hashish use	15/144	8/76	0.99 (0.37–2.83)	1.000
20. Heroin/opium use	4/73	19/147	0.39 (0.09–1.24)	0.105
21. Cocaine/crash use	1/29	22/191	0.28 (0.01–1.85)	0.326
22. Diazepam/Valium use	1/3	22/217	4.38 (0.07–87.38)	0.283
Sexual health and clinical characteristics				
23. STI-related symptoms during preceding year	20/114	3/106	7.25 (2.05–39.31)	<0.001
24. Receptive anal intercourse during preceding year	19/131	4/89	3.59 (1.13–15.03)	0.023
25. Previous tattooing or tribal marking	2/22	21/198	0.84 (0.09–3.90)	1.000
26. Previous surgical procedure	5/46	18/174	1.06 (0.29–3.19)	1.000
27. Recreational injection drug use	10/31	13/189	6.36 (2.21–18.06)	<0.001
28. Vaginal intercourse outside marriage	21/151	2/69	5.38 (1.25–48.73)	0.016
Healthcare-related and blood-exposure practices				
29. Therapeutic injection or intravenous drip	21/142	2/78	6.55 (1.53–59.26)	0.005
30. Community barber visits	23/220	—	Not estimable	—
31. Lubricant use during anal intercourse (three categories)	—	—	Not applicable	0.447
32. Did not request a fresh shaving blade	10/31	13/189	6.36 (2.21–18.06)	<0.001
33. Previous ear or nose piercing	6/32	17/188	2.31 (0.68–6.88)	0.116
34. Did not request a fresh syringe/needle (SPSS grouping)	10/35	13/185	5.23 (1.85–14.57)	<0.001
35. Piercing blade use (three categories)	—	—	Not applicable	0.908
36. Did not request a fresh piercing blade	2/23	21/197	0.80 (0.08–3.67)	1.000

DISCUSSION

The present cross-sectional study evaluated HIV, hepatitis B virus (HBV), and hepatitis C virus (HCV) infection markers and associated behavioral characteristics among 220 truck drivers and assistants in Lahore, Pakistan [1,2]. Twenty-three participants (10.5%) had detectable viral nucleic acid, with seven (3.2%) having two or more detectable viruses. The most common was HCV RNA, followed by HIV RNA (4.1%) and HBV DNA (2.7%). Among the study participants, it is important to note that

the number of anti-HCV antibody positive cases (10.5%) was higher than the number of HCV RNA positive cases (7.7%), with 6 of the HCV antibody positive cases being HCV RNA negative. These findings underscore the need for molecular testing to determine whether individuals have been previously exposed to HCV or are currently viremic and indicate the presence of multiple viral infections in this occupational population [3,4].

The difference between “serological screening” and “molecular detection” has implications for

infectious disease surveillance and clinical management. Anti-HCV antibodies are a sign of past or present exposure; detectable HCV RNA is evidence of continued viremia [5]. HBsAg and HBV DNA complement each other in providing information about the HBV infection and the viral replication. Further identification of concurrent viral infections further highlights integrated screening approaches, particularly among individuals with similar exposures. However, simultaneous detection does not necessarily mean a shared route of transmission or time of acquisition of the infection. Confirmatory diagnostic algorithms and clinical assessment are still crucial for accurate classification, and management [6-8]. Multiple sexual and occupational factors were significantly associated with composite detection (by viral PCR) and were related to proximity to the animal and number of exposures. Multiple commercial sex worker contacts, frequent commercial sex, condom nonuse and earlier sexual debut increased frequency of viral detection [9]. Similarly, longer occupational absenteeism (defined as ≥ 4 months of absence) was significantly associated with composite PCR positivity (OR: 5.36; 95% CI: 2.01–15.05; $p < 0.001$). Findings indicated that occupational mobility and some sexual practices reported could serve as indicators to prioritize groups for which it is necessary to augment health care services for prevention. But these exposures can be related to each other and associations with viral detection cannot be used to conclude independent risk factors or specific transmission routes [2,3,9].

Other public health findings included substance use and exposures from injection. Use of recreational injection drugs had a significant association with the composite PCR detection (OR: 6.36; 95% CI: 2.21–18.06; $p < 0.001$). Participants who had reported therapeutic injections/IV drips and those who did not ask for new shaving blades/syringes also had higher detection frequencies [10]. All these observations highlight the need for sterile injection equipment, proper infection control practices, and education about the transmission of blood-borne viruses. However, the safety of healthcare procedures reported was not independently evaluated and the results cannot determine if specific medical or grooming contacts caused infection. Likewise, individuals who had had sexual health-related symptoms in the past 12 months had a higher composite PCR positivity, suggesting the potential for combined

sexual health and viral screening services [11,12].

The use of preventive health care seemed to be restricted, although there is widespread knowledge about HIV/AIDS and viral hepatitis. Less than half of the participants (32.7%) had been vaccinated against HBV, and 105 (47.7%) had been tested for HIV, HBV or HCV sometime in the period of the questionnaire [13]. Moreover, 143 (65.0%) respondents responded that they would be safe if they use condoms properly in the multi item awareness question. These findings suggest areas for improvement to boost vaccination, accessibility of screening, and knowledge of transmission prevention. Station-based programs with confidential testing, hepatitis B vaccination assessment, counseling and linkage to clinical care sites may be an option to reach occupationally mobile populations. But their feasibility and effectiveness need to be assessed from a prospective point of view [1,3,14].

This study's main strengths are its serological and molecular results, characterization of individual and multiple viral detections, and complete evaluation of occupation and behavior exposures. However, due to the cross sectional design of the study, it was not possible to determine temporal or causal relationship [15-17]. Generalizability was limited due to convenience sampling and self-reported behaviors may be subject to recall and social-desirability bias. Several subgroup comparisons had imprecise estimates as only a small number of PCR positive participants were included. In addition, lack of adjusted analyses, possible misclassification of exposure, and multiple exploratory comparisons limit interpretation of the observed associations [18,19]. These results thus need to be validated with larger, multicenter prospective trials with standardized exposure assessment, full diagnostic algorithms, and appropriately powered virus-specific analyses [20].

CONCLUSION

The prevalence of detectable HIV RNA, HBV DNA, and HCV RNA among 220 transport workers in Lahore was found to be 4%, 2.7%, and 7.7%, respectively. A total of 10.5% (3.2% multiple viral PCR detections) had at least one detectable viral PCR result. The antibody HCV reactivity was higher than the HCV RNA detection, highlighting the need for molecular confirmation. Several exposures were found to be significantly associated with viral detection in unadjusted analyses, but no causal

relationships could be inferred from these findings in any of the exposures. Results indicate a need for continued assessment of accessible occupational screening, hepatitis B vaccination, infection-prevention education and linkage-to-care for transport workers.

Conflict of Interest

The authors declare no conflicts of interest.

Data Availability

Data are available from the corresponding author upon reasonable request.

Acknowledgments

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Ethical Approval

The study received ethical approval from the Institutional Review Board and was conducted in accordance with the Declaration of Helsinki.

Author Contributions

M.F.H.B.: Conceptualization, methodology, data collection, analysis, and drafting. T.M.: Supervision, methodological review, and critical revision. A.A.S. and M.A.A.: Statistical analysis, interpretation, and visualization. S.R., M.U.A., and A.R.: Review, editing, and proofreading. All authors approved the final manuscript and accepted accountability for the work.

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