

COMPARATIVE EVALUATION OF THREE SEROLOGICAL TESTS FOR HIV DETECTION IN HARIPUR, PAKISTAN

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Abstract: *Since many years ago and with rapid population growth, the human immunodeficiency virus (HIV) has been acknowledged as a serious public health concern worldwide. However, there does not appear to be enough information available on the prevalence of HIV infection in Haripur, Pakistan, as a whole. The current study's goals were to assess the effectiveness of three different HIV screening methods and to ascertain the seroprevalence of HIV infection in Haripur, Pakistan, among the general population. An immunochromatographic test (ICT), enzyme-linked immunosorbent assay (ELISA), and reverse transcription polymerase chain reaction (RT-PCR) were used to test a total of 5370 blood samples for the presence of anti-HIV antibodies and HIV-RNA at the antiretroviral therapy (ART) center in Hayatabad Medical Complex (HMC), Haripur. 756 (14.07%) of the 5370 blood samples tested positive for ICT, 639 (11.89%) for ELISA, and 606 (11.28%) for RT-PCR. Males were more likely to have an active HIV infection than females, with 468 having it compared to 138 who did (7.14%). The age group of 01–10 years showed no signs of infection, whereas the age group of 21–30 years had the highest infection rate (237, or 16.80%). However, the HIV seroprevalence in the married population was 423 (13.83%), compared to 183 (7.91%) in the single population. ICT should not be used to detect anti-HIV antibodies, and RT-PCR instead of ELISA should be used to detect HIV RNA. In order to avoid this illness, medical care centers in rural regions should guarantee the availability of treatment and screening facilities against this virus.*

Keywords: Human Immune Deficiency Virus, Serological Testing, Viral Markers, RNA virus, RT-PCR

Introduction

An enveloped lentivirus belonging to the retrovirus family is the human immunodeficiency virus. Its genetic material consists of a ribonucleic acid virus, a single nucleic acid structure (Bieniasz and Telesnitsky, 2018). About 40 million infections are brought on by this devastating epidemic each year worldwide (Huremović, 2019). HIV can spread from one person to another through vaginal, anal, or oral sex, blood transfusion, childbirth, contaminated hypodermic needles, breastfeeding, and contact between mother and child while pregnant (Nordqvist, 2016; Sheikh, 2015). Acquired immunodeficiency syndrome (AIDS), a chronic, life-threatening condition, is brought on by HIV's damage to a person's immune system. Every year, HIV claims the lives of 25 million individuals worldwide (Kandula and Wake, 2022). Currently, 3.1 million people die yearly from AIDS-related causes, affecting more than 40 million globally

(Yoshimura, 2017). The global spread of HIV infection. The extermination of millions of people is a real possibility. 90% of AIDS patients in underdeveloped nations experience illiteracy, poverty, underdevelopment, a lack of healthcare resources, and hunger (Hipgrave, 2011). HIV infection progresses via three stages: acute, delayed, and AIDS. Each stage is distinct from the others in terms of timing and symptoms. The signs of acute infection can include esophageal mouth sores, lethargy, muscle discomfort, swollen lymph nodes, a low fever, and lymphadenopathy. A few weeks may pass before the virus clears. With few or no symptoms, the late stage might endure for two weeks to twenty years.

When the CD4 level is less than 200, the ultimate stage, known as AIDS, is dependent on the individual immune system. According to Dieffenbach and Fauci (2011), the main signs and symptoms of AIDS are shortness of breath, coughing, diarrhea, fast weight



loss, vomiting, chronic exhaustion, night sweats, nausea, recurrent fever, sores in the mouth or nose, under the skin, or on the genitalia. Unprotected sexual interactions and using unsterilized needles for drug injection are two of the main sources of HIV in Pakistan. Furthermore, a contributing factor to the incidence of this illness is the lack of knowledge regarding HIV and AIDS (Dieffenbach and Fauci, 2011). According to other scientific, HIV is a contagious infection that spreads rapidly and constantly (Akseer et al., 2020; Maimaiti and Andersson, 2008). In this study, the general population of Haripur, Pakistan, was tested for anti-HIV antibodies and HIV- RNA. Our study examined the effectiveness of screening strategies for detecting active HIV infection and the incidence of actual HIV infection.

Methodology

Sample selection and study design The Antiretroviral Therapy (ART) Centre (HMC), Haripur, conducted this retrospective analysis from March 2017 to January 2018. In the current investigation, 5370 samples were taken from people in Haripur who had been advised to get tested for HIV by medical practitioners. The district of Khyber Pakhtunkhwa, Haripur, is the sixth largest city in KPK and the most significant city in the region. With 1.003 million people living there, it has a 1,725 km² area. Each subject was provided three cubic centimeters of blood in a vacutainer tube, and HIV testing was done with ICT, ELISA, and RT-PCR-based techniques. The age ranges included in this study were 03–79.

Immuno-chromatographic tests (ICT)

Blood samples were collected in a vacutainer tube, and serum was separated from it by centrifuging them at high speed for five minutes, roughly 15000 rpm. Following the manufacturer's directions, the ICT

technique was used to identify anti-HIV antibodies. The immunological chromatographic strips (ICS) used were from the American companies Accon and Accurate. ICT-positive samples were tested for anti-HIV antibodies using an ELISA with a Werfen Company (Spain) Biokit in accordance with the manufacturer's instructions.

Nucleic acid extraction (NAE) and RT-PCR

Following RT-PCR, HIV-RNA was extracted from the samples that had been confirmed ELISA-positive using an Anagen RNA extraction kit (Anagen, USA). The manufacturer's instructions performed RT-PCR using the Norgen Biotek Corp (Canada) kit.

Statistic evaluation

The Statistical Package for the Social Sciences (SPSS), version 20, was used to run descriptive statistics on all the data entered into a Microsoft Excel sheet.

Results

Five thousand three hundred seventy blood samples, 3438 (64.02%) male and 1932 (35.97%) female were drawn for this investigation. All the collected samples were also divided into seven age groups. 756 (14.07%) of the 5370 blood samples tested positive for anti-HIV antibodies using the ICT technique. Additionally, ELISA was used to validate ICT-positive samples, and 639 (11.89%) of these samples tested positive for anti-HIV antibodies. Additionally, the ELISA-positive samples were subjected to RT-PCR processing for HIV-RNA extraction, and 606 (11.28%) were found to be HIV-RNA positive. According to Figure 1, the age groups 21 to 30 years had the highest seroprevalence of active HIV infection, with 237 (16.80%) individuals testing positive. Age groups 41 to 50 years had the lowest seroprevalence, with 39 (4.46%) individuals testing positive.

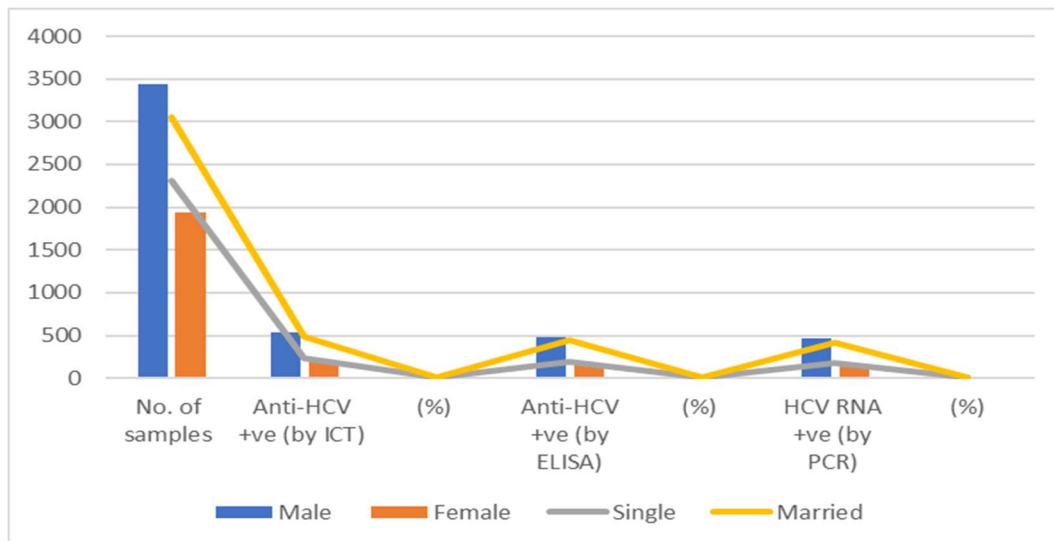


Figure 1. Seroprevalence of HIV positive samples in relation to age groups.

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Males were more likely to have HIV infection than females, with 468 (13.61%) versus 138 (7.14%). As indicated in Figure 2, the results also showed a

significant seroprevalence of HIV infection in the married population, which was 423 (13.83%), while the single population was 183 (7.91%).

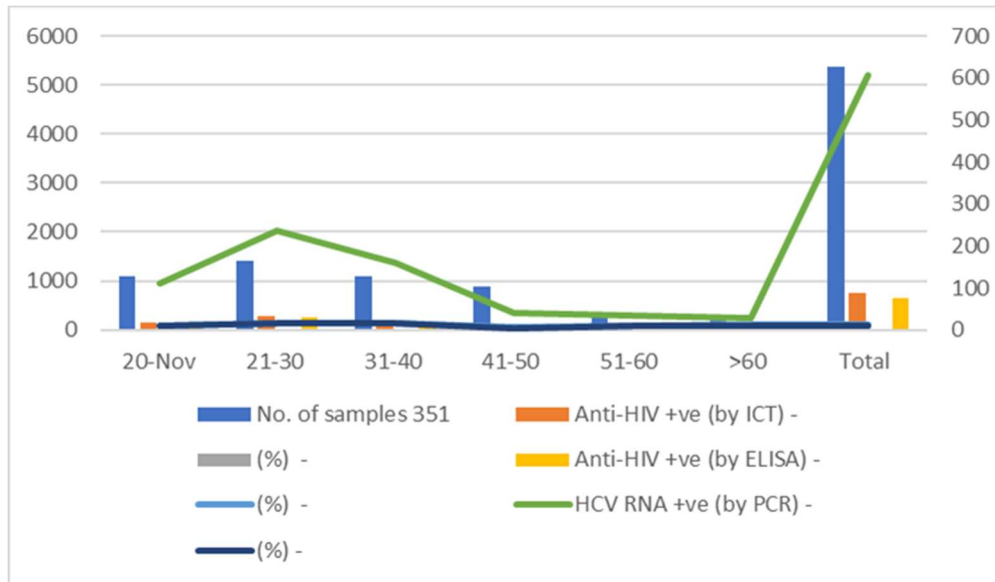


Figure 2. Marital status and gender-wise seroprevalence of HIV-positive samples.

Discussion

One of the biggest global public health problems, particularly in underdeveloped nations, is HIV. Pakistan is positioned between HIV high-risk nations like Afghanistan on the west, India on the east, and China on the north, making it a high-risk country for virus transmission. Pakistanis are susceptible to HIV infection because of this. In previous studies on HIV seroprevalence in various parts of Pakistan, patients with renal diseases and drug users had higher seroprevalences of HIV (27.27% and 27.3%, respectively) (Akram et al., 2018; Mansoor et al., 2014)(Adil et al., 2014; Altaf et al., 2016).

Other areas with HIV seroprevalence of 22.25 percent, 2.01 percent, 21.6%, 5.3%, and 4.50 percent, respectively, were Sindh, Lahore, Rawalpindi, Punjab, and Swabi (Akhtar et al., 2012; Nafees et al., 2011; Shahid et al., 2016).

64% of the 5370 HIV infection cases that were confirmed positive were in males, and 36% were in females. Similar studies from around the world, including Pakistan (Burnett et al., 2005; Khattak et al., 2014; Valadas et al., 2009), reveal that men make up most of the population. Infection with HIV is highly prevalent among married people. According to previous studies conducted worldwide, including in Pakistan, 15.99% of samples were found to be HIV-positive. Widowed people and divorcees were excluded from our study. According to the study, among the age groups, those between

the ages of 21 and 30 have the highest infection rates, followed by those between the ages of 31 and 40 (Azuoanwu et al., 2017; Zareen et al., 2016). According to (UNICEF et al., 2008), young individuals between the ages of 15 and 24 had a more than 40% prevalence. Our finding is consistent with that conclusion (Sheet, 2008).

When blood samples were examined using ICT tools for this study, the seroprevalence of anti-HIV antibodies was 14.07%, consistent with the studies' findings stated above. Due to a false positivity rate of ICT devices (Junaid et al., 2017; Mylonakis et al., 2000), we intended to improve the screening process so that positive ICT serum was tested using the third generation of ELISA. According to the ELISA test, 11.89% of blood samples tested positive for anti-HIV antibodies. This finding shows that ICT systems fall short of accurately reflecting the seroprevalence of HIV infection. It is suggested that ELISA is more trustworthy than ICT equipment. ICT tools are employed in many healthcare facilities to detect HIV infections.

HIV core antigens in the blood can be found with the aid of ELISA. ELISA devices have an advantage over ICT devices for developing nations like Afghanistan, Bangladesh, India, and Pakistan that cannot pay to test blood for nucleic acid amplification. We used a real-time PCR test to investigate the ELISA-positive samples and found that 11.28% of them have HIV-RNA that is actively infected, regardless of whether there is an active infection. The type of illness or the presence of antibodies in the patient receiving

[Citation Rehman, M.U., Zaman, S., Sajjad, A., Ali, N., Hayat, A., Khan, I., Ramzan, H.S. (2023). Comparative evaluation of three serological tests for hiv detection in haripur, pakistan. *Biol. Clin. Sci. Res. J.*, 2023: 339. doi: <https://doi.org/10.54112/bcsrj.v2023i1.339>]

comprehensive treatment may contribute to this decline. In the general population of Haripur, Pakistan, the seroprevalence of active HIV infection is 11.28%. This seroprevalence is relatively low in comparison to previously stated values. The incidence of this viral infection can be decreased by using an appropriate screening assay like ELISA instead of an ICT device and PCR in place of ELISA.

Conclusion

We conclude that 11.28% of people in Haripur, Pakistan, have an active HIV infection. If we compare this proportion to the rates that have already been reported, it is not that high. The prevalence of this viral infection can be decreased by using proper screening assays, like ELISA, instead of an ICT device.

Conflict of interest

The authors declared absence of conflict of interest.

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