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False Positive HIV "Combo" Screening Test in a Hemodialysis Patient in Saudi Arabia: A Case Report and Review of the Literature

Ngozi Virginia Aikpokpo[1,2*], Ahmed Yayha Omaysh[1]

[1]Diaverum AB Saudi Arabia, Riyadh, KSA
[2]Department of Health, University of Bath, Claverton Down, Bath, UK
Email: *ginnyaik@yahoo.com

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Abstract

Introduction: HIV screening tests are routinely conducted on dialysis patients as the constant exposure of their blood during the dialysis process makes them a reasonable risk for blood-borne infections. However, in low prevalence settings, where HIV rates are <0.1% of the population, false positive results are more likely. This results in apprehension in the dialysis unit as breaches in infectious disease protocols could be presumed. This is illustrated in the case report below. Case Summary: A 62-year-old male Saudi end-stage kidney disease patient secondary to DM nephropathy began dialysis a year before presentation in a hemodialysis center in Saudi Arabia. Routine screening tests done at the start of dialysis revealed negative Hepatitis C, HIV 1 and 2 screening but a positive Hepatitis B surface antigen screen. The patient went for holiday dialysis at another facility and had a routine fourth-generation HIV test done which was positive. A confirmatory HIV PCR test was negative. Conclusion: This case highlights the need for caution in interpreting highly sensitive and specific HIV screening tests in a low-prevalence setting. Routine screening beyond the national recommendation may not be necessary in low-prevalence areas.

Keywords

Saudi Arabia, False Positive HIV Test, Hemodialysis

1. Case Report

A 62-year-old Saudi male patient was diagnosed with stage 5 chronic kidney disease in 2020 but commenced hemodialysis (HD) in a government facility in Saudi

Arabia in June 2023 via a right internal jugular permanent catheter following pulmonary edema. Past medical history was significant for type 2 diabetes, diagnosed in 2003, and hypertension, diagnosed in 2020. He had a history of bilateral retinopathy and had laser surgery in 2019 and 2020. Routine screening tests at the start of HD revealed hepatitis B surface antigen positive, HIV 1 and 2 negative, and hepatitis C antibody negative. He was consequently dialyzed in a segregated area as per unit protocol.

He transferred to this facility in January 2024. On presentation, the patient had no complaints, the physical examination was unremarkable, and repeat serology testing was done per the company's protocol. Again, he tested Hepatitis B surface antigen positive, while Hepatitis C and HIV 1 and 2 were negative. He continued dialysis in the designated hepatitis B station as per guidelines, with universal, blood, and body fluid precautions strictly observed. The patient had no history of blood transfusion, the patient was married to one wife, and sexual history was unremarkable. There were no cases of HIV-infected patients or staff in the facility.

The patient desired to have holiday dialysis in another private facility in Saudi Arabia. As part of the facility's protocol, all new patients must have a fourth-generation HIV immunoassay P24 antigen and antibody test (HIV Combo test) for HIV screening. The patient tested positive for the P24 antigen test. He had no signs or symptoms of acute illness or seroconversion. An HIV PCR test was done to ascertain the patien's HIV status, and this was negative. A repeat test done 3 months later, as part of the annual serology screening protocol, was also negative.

The utility of P24 antigen/antibody testing as screening for HIV in asymptomatic patients in a low prevalence setting is therefore being questioned.

2. Literature Review

The literature review was done in parts to enable an understanding of the historical hazards of positive HIV tests, recommendations for HIV screening, available HIV immunoassays, and factors associated with false positive HIV tests.

To accomplish this, a literature search was conducted using several databases assessed via the University of Bath library catalogue. These include the Web of Science, Embase, and PubMed databases, which are the most appropriate for healthcare searches. Google and Google Scholar search engines were also used. Only peer-reviewed articles were considered.

3. Historical Hazards of Positive HIV Tests

Blood-borne virus infections have been a recognized hazard for patients and staff in renal units for decades [1]. As a result, many guidelines require patients and staff to have baseline serology testing for blood-borne viruses (BBV), notably hepatitis B, C, and HIV viruses. This is typically done for patients before commencing dialysis in a new center and repeated annually thereafter [2]-[5].

Although not as transmissible as the hepatitis B virus, hemodialysis-associated HIV transmission has been reported in the nineties, mostly due to breaches in

infectious disease practices [6] [7]. In Saudi Arabia, HIV transmission in a hemodialysis unit was documented in the southern region of the kingdom in 2012 [8]. The infection was linked to an HIV-infected patient referred to the dialysis unit from the Abha region [8]. In the era of good infectious disease practices, transmission of HIV infection in hemodialysis units is rare [9].

4. Recommendations for HIV Screening

HIV screening is defined as performing an HIV test for all persons in a defined population. The CDC recommends HIV screening be performed routinely for all patients aged 13 - 64 years except HIV infection rates in the community have been documented to be <0.1% [10]. Repeat screening is recommended at least annually for patients at high risk for HIV infection. These include injection drug users and persons with multiple sexual partners. Repeat screening of persons not likely to be at high risk for HIV should be performed based on clinical judgment [10]. It is thus sensible to screen chronic hemodialysis patients annually as the hemodialysis process involves regular exposure of patients' blood.

HIV screening is done by immunoassay testing. Immunoassays detect infections by using the principles of antigen-antibody reaction. The results of the screening test can be affected by the stage of the disease. The "window period or seroconversion period" describes the time interval before HIV-specific antibodies appear [11]. This period is marked by the absence of HIV-specific antibodies and detectable viremia (measured by RNA or p24 antigen). IgM antibodies are the first antibodies to be detected after infection and indicate early stage of infections, usually within three weeks of infection, and peaks at the 4th or 5th week [11]. IgG antibodies are produced within 3 - 4 weeks of infection, increase soon after their appearance, and show a reduction about 10 - 12 weeks after infection [11]. IgG antibodies may remain detectable for years. HIV immunoassays have evolved over the years. Broadly speaking, they are classified according to "generations". There are currently four generations of HIV immunoassays used for screening [12] [13]. These are summarised in **Table 1**.

Table 1. Types of HIV immunoassay.

Generation	Examples	Characteristics
First generation, detects HIV 1 antibodies.	HIV 1 Western blot, HIV-1 IFA	Requires significant specimen dilution to overcome cross-reactivity with cellular protein contaminants hence reduced specificity. Detect IgG antibodies (thus miss early infections).
Second generation	HIV-1 enzyme immunoassay, six rapid HIV antibody tests	Improves sensitivity for HIV-1 group O and HIV-2. Improves specificity by eliminating cross-reactivity with cellular proteins. Detects IgG antibodies hence miss early infections.

Continued

Third generation	HIV1/HIV2 enzyme immunoassay, HIV1/HIV2 chemiluminescent immunoassay	Detects IgG and IgM antibodies. Lower sample dilutions and ability to detect IgM antibodies increase sensitivity during early seroconversion.
Fourth generation	HIV1/HIV2 rapid test that uses separate indicators for antigen and antibody reactivity. HIV Ag/Ab combo assay	Detects IgM and IgG antibodies, monoclonal antibodies are also included to detect p24 antigen (which allows detection of HIV-1 infection before seroconversion. Some "combo" assays do not distinguish antibody reactivity from antigen reactivity.

Fourth-generation immunoassays are currently the recommended tests for routine use in most guidelines [12] [13]. As they test for p24 antigen and antibodies, they are better suited for detecting recent infection. They also have excellent sensitivities and specificities. However, false positive tests have been known to occur with these immunoassays [14]-[32]. Heterophilic interference, non-specific cross-reactivity to synthetic peptides, and cross-reactions to viruses, infections, vaccines, and autoimmune diseases have been proposed as associations with false positive 4th generation tests. These are summarised in Table 2.

Table 2. Factors associated with false positive fourth-generation HIV tests.

Associations	Examples
Viruses	Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) [15] [22] Cororanavirus-19 [17] Hepatitis A virus [20] Ebstein Barr virus [14]
Other infections	Prior schistosomiasis infection [18] [30]
Tumors and cancers	Ameloblastoma [18] Hodgkin Lymphoma [23] Other hematologic malignancies, some solid tumors, and metastatic cancers [14] [26]
Vaccinations	Covid-19 vaccination [16]
Autoimmune diseases	Autoimmune hepatitis type 1 [21] Systemic Lupus Erythematosus [24] Autoimmune hemolytic anemia [25] Rheumatoid arthritis [28]
Miscellaneous Immunologic causes	Non-specific reactivity to isolated synthetic peptide component of the assay with cross-reactivity to other antigens [18] Heightened CD5+ and early lymphocyte response with polyclonal cross-reactivity [19] Presence of P24 antigen and CD4 lymphocytopenia [27] Heterophilic antibody interference [31] [32]

As seen from Table 2, there are many plausible causes for a false positive fourth-generation HIV screening test. Therefore, A positive result should be treated with caution in a low-risk patient in a low-prevalence setting.

5. Discussion

Saudi Arabia is a low-HIV-prevalence country with prevalence rates of <0.1 [33]. According to the WHO Eastern Mediterranean Region report (Saudi Arabia profile), there was a rise in new HIV infections between 2010 and 2020 which was when the report was published [33]. Despite this increase, Saudi Arabia remains a low-HIV-prevalence country.

The likelihood of a false positive HIV test increases as the prevalence of HIV reduces. Kim et al. retrospectively analyzed the performance of the Architect HIV antigen/antibody (Ag/Ab) combination assay and found that the specificity and positive predictive value (PPV) were 99.78% and 31.21%, respectively [34]. Specificity is the probability of a negative result being truly negative, while the PPV is the probability of a positive result being truly positive. In simple terms, Kim *et al.* showed that in a low prevalence area (such as Saudi Arabia), a negative test has a 99.78% probability of being truly negative, while a positive test has a 31.21% chance of being truly positive. In other words, it has a greater chance, 68.79% of being negative. In a low prevalence setting, therefore, other potential explanations for a positive P24 test become as prevalent as HIV.

In the case scenario above, the patient was screened for BBV twice before initiating dialysis at the third center: once before he was commenced on hemodialysis in a government facility and another before he was commenced in a private facility. Both tests revealed HIV and Hepatitis C were negative while Hepatitis B surface antigen was positive. False positive HIV-1 ELISA tests have been reported in Hepatitis B-positive patients and patients with Hepatitis A with the older generation immunoassays [29] [35]. Although there have been no reports of false HIV positivity in hepatitis B patients using fourth-generation immunoassays, there have been reports of false positive fourth-generation HIV 'combo' tests in hepatitis A patients [20]. As accounts are limited to case series, it is not clear if this is a causal association.

At the time of collection of samples for the HIV screening test at the third center, the patient was in good health, and his chronic conditions, diabetes and CKD, were well controlled on insulin and adequate hemodialysis, respectively. He did not have any symptoms or signs of a viral illness and had not received any recent immunizations. The probable cause of the false positive test, therefore, remains obscure.

HIV infections are uncommon in hemodialysis centers in Saudi Arabia. Although there are few prevalence studies on serology status of hemodialysis patients in Saudi Arabia, the Saudi Center for Organ Transplantation (SCOT) data for 2018 revealed a prevalence rate of HCV and Hepatitis B of 9% and 2.7%, respectively out of 20,496 patients [36]. No data was given for HIV prevalence, perhaps

because of low prevalence rates. A recent study in a dialysis center in the Eastern province of Saudi Arabia the prevalence of blood-borne viruses was assessed amongst 239 patients. 3.77% and 7.53% were reactive to Hepatitis B surface antigen and hepatis C antibodies respectively but there were no cases of HIV infection [37]. An older study in a large dialysis center in Riyadh, evaluating quality of care amongst dialysis patients also noted that were no cases of HIV infection [38]. As the cases of HIV infection amongst hemodialysis patients are low, it is recommended that routine HIV testing in holiday dialysis patients would not be necessary, as false positives are more likely given the low prevalence rates.

This report has certain limitations, as it is a case report, it is difficult to ascertain causality for the false positive test. However, as a case report, it adds information about resource utilization as performing unnecessary tests leads to increase costs.

6. Conclusion

False positive HIV screening tests can occur and cause considerable anxiety in the outpatient hemodialysis setting as a positive result may indicate a breach of infectious disease protocols. However, in a low-prevalence setting with no case of HIV infection in the hemodialysis unit, these results must be interpreted with caution. A confirmatory HIV test such as HIV PCR done in this case is needed. In the "window period", viremia is often very high and can be detected by polymerase chain reaction (PCR). A negative PCR therefore indicates NO HIV infection. Although reasons for a false positive test may not always be clear, it should be considered in all cases. Staff must be aware of the high incidence of false positive tests in a low prevalence center. It may therefore be prudent to avoid conducting routine highly sensitive and specific HIV screening tests in hemodialysis centers with low prevalence rates than what is recommended to reduce the waste of resources.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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