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Research Article

Active Human T- lymphotropic Virus Type-1 Antigenemia in a Sample of Iraqi Patients on Maintenance Hemodialysis

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ABSTRACT

Background: Human T-lymphotropic virus type-1 (HTLV-1) is an oncogenic retrovirus that causes lifelong and asymptomatic infection, with a high incidence globally. Patients undergoing hemodialysis are more likely to acquire blood-borne viral infections, including HTLV-1. Therefore, they are considered one of the highest-risk groups for the acquisition of this virus.
Objectives: This study aims to identify the HTLV-1 antigenemia among hemodialysis patients, and to investigate the potential risk factors and co-viral infections with Hepatitis B and C viruses.
Subjects and Methods: This cross-sectional study involved 130 hemodialysis patients attending the dialysis center at Imamein Kadhimein Medical City in Baghdad, Iraq in the period between November 2023 and January 2024. Using enzyme-linked immunosorbent assay was applied for the detection of HTLV-1 specific capsid antigen (p24) in the serum of the studied group. The data was analyzed using specific statistical methods.
Results: Out of 130 participants, 60(46.2%) actively contracted HTLV-1 infection, comprising 37 (28.5%) males and 23 (17.7%) females. The detection of HTLV-1antigen was significantly correlated with the patient's age (0.0037), but not with marital status, frequency of blood transfusion, and hemodialysis duration. Moreover, active HTLV-1 infection was related to co-infections with HCV 18(13.9%) and HBV 5(3.8%) of patients.
Conclusions: In this study, hemodialysis patients were predominantly affected by HTLV-1 infection. Therefore, it is of utmost importance to implement screening protocols to curb the transmission of this infection within hemodialysis units. Further research involving a significant cohort encompassing diverse regions of Iraq is recommended to ascertain the accurate prevalence of HTLV-1 infection.

Introduction

Human T-lymphotropic virus type 1 (HTLV-1) is a human retrovirus that primarily infects CD4+ T cells, leading to persistent infection of the immune system. It is a member of the genus Deltaretrovirus within the subfamily Orthoretrovirinae (1, 2). HTLV-1 consists of a positive single-stranded RNA genome that is integrated into the host DNA as a provirus upon infection (3), its transmission can occur through intravenous drug use, sexual contact, and mother-to-child breastfeeding (4). HTLV-1 is a latent retrovirus

that carries an ongoing risk of adult T-cell leukemia (ATL), myelopathy/tropical spastic paraparesis (HAM/TSP), and other diseases (5, 6). It is estimated that approximately 10 million people worldwide are infected with HTLV-1, which is endemic in Latin America (7), West and South Africa (8), Japan, Iran, Romania, and Australia (9). The frequency of HTLV-1 infections is increasing in parts of the world. The prevalence of HTLV varies worldwide by region. HTLV-1 infection is increasing outside of endemic areas(10). Hemodialysis(HD) is the most prevalent form of renal replacement therapy for individuals with end-stage kidney disease (11)and, like any other intervention, has several side effects, including the risk of acquiring viral infections such as cytomegalovirus(CMV), Hepatitis G virus(HGV), Hepatitis C virus(HCV) and Hepatitis B virus (HBV) that are a leading cause of death in HD patients following cardiovascular disorders (12-15). HTLV-1 may be a risk factor for End Stage Renal Disease (ESRD) due to its urological manifestations, such as neurogenic bladder, which can lead to subsequent post-renal kidney failure (16). It has been observed that patients with end-stage renal disease have a high incidence of HTLV infection (17). Furthermore, it has long been suspected that people with HTLV-1 have an increased risk of chronic kidney diseases (CKD). Several cross-sectional studies have reported a high prevalence of HTLV-1 seropositivity rates (ranging from 14.5% up to 21.2%) among dialysis patients(18, 19). This could be due to anemia (20, 21) and regular blood transfusions in HD patients, or it may result from the risk of acquiring the virus during the hemodialysis procedure, similar to how HBV and HCV infections can occur (22, 23).

It is imperative to implement preventive strategies to control HTLV-1 infections since antiretroviral drugs are ineffective in confirmed infections, and managing the consequences can be challenging. Currently, there is no vaccine available to prevent HTLV infection. It is necessary to gather empirical evidence to develop policies for preventing HTLV-1 transmission (24). This study was conducted to assess the occurrence of HTLV-1 infection and associated risk factors, along with the risk of co-infection with other blood-borne viruses, taking into account the global spread of HTLV-1.

Subjects and Methods

Study design and Sample size: This is a cross-sectional study of 130 hemodialysis patients randomly selected according to inclusion and exclusion criteria (77 males and 53 females) who were admitted to the dialysis center at Imamein Kadhimein Medical City between November 2023 and January 2024. The patients ranged in age from 18 to 83 years. Patients who underwent dialysis for less than 6 months, those who were taking anti-retroviral drugs, and those who refused to participate in the study were excluded as per the criteria.

This research has received approval from the Ethics Committee of the Department of Microbiology College of Medicine, University of Baghdad (Institutional Review Board-Reference Number: 0230A1-11-2023). All patients participating in the study submitted a consent form and agreed to participate before collecting their blood samples. Baseline data including duration and frequency of dialysis, as well as other sociodemographic information, were gathered for each patient.

Blood samples of 5-10 ml were collected from all patients using accepted medical techniques. Each blood sample was then collected in a gel separation tube, and centrifuged at 3000 rpm for 20 minutes. The resulting serum was stored at -20°C until the examination. An Enzyme-linked immunosorbent assay two kits from (Cat.No: MBS9309936 MyBioSource, USA) was used to qualitatively determine Human T-Lymphotropic Virus Type-I capsid antigen (p24) in serum samples. The manufacturer's instructions were fully followed throughout the testing process. Fifty microliters of Positive, Negative Control and 10µl of each testing sample were pipetted to the corresponding wells. 100 µl of HRP-conjugated Human T-Lymphotropic Virus Type I antibody was added at the same time to bind the analyte, followed by incubation (for 60 minutes at 37°C) and washing procedures to remove the unbound substance. Finally, 50µl of HRP substrates were added, and incubated for 15 minutes till detection, and a blue color was developed. Adding a 50µl of stop solution terminated the enzyme-substrate reaction and the color turned from blue to yellow. The optical density (O.D) of each well was measured within 15 minutes using a microtiter plate reader set at 450 nm. The diagnosis of HBV and HCV was done as part of routine screening tests by the detection of HBsAg and anti-HBc for HBV infection, anti-HCV IgG confirmed by reverse transcriptase-polymerase chain reaction for HCV infection.

Data analysis was conducted using the Statistical Package for the Social Sciences software, version 21 (SPSS, IBM). The Pearson chi-square (χ2-test) was employed to compare the percentage of data for two groups, while the *t*-test was used to compare data from more than two variables. All tests were considered significant when the p-values were $\leq$0.05.

Results

The study included 130 patients, of which 77 (59.2%) were males and 53 (40.8%) were females. The ratio of male-to-female was 1.4:1. The age group ranged from 18 to 83 years, with a mean age of 51.8 ± 13.7 years. Sixty patients (46.2%) tested positive for HTLV-1 p24 antigen. Males had a higher infection rate than females, with 37 (28.5%) males and 23 (17.7%) females testing positive for HTLV-1 antigen. The age groups between 50-59 and ≥60 years had higher rates of HTLV-1 infection, with 17(13.1%) and 19(14.6%) respectively(p-value=0.0037) . The HTLV-1 antigen seropositive status among hemodialysis patients was statistically not correlated with other sociodemographic characteristics except for age group. The risk factors for HTLV-1 transmission are categorized into three categories: history of blood transfusion, duration of hemodialysis, and history of tattooing. Among the patients who underwent more than two blood transfusions, 33 (25.4%) tested positive for HTLV-1 p24. The majority of active infections were observed in patients who underwent hemodialysis for a duration between 1 to 5 years, with no significant positive correlation observed, as illustrated in Table 1.

Table 1: HTLV-1 p24 results in relation to sociodemographic characteristics and associated risk factors of the study group

Variables		HTLV-1p24 No. (%)		OR	Correlation Coefficients	P-value
		Positive	Negative			
Age(years)	<20	2 (1.5)	1 (0.8)	1.2	R=0.9022	0.0037
	20-29	5 (3.8)	2 (1.53)			
	30-39	4 (3.1)	8 (6.2)			
	40-49	13 (10.0)	16 (12.3)			
	50-59	17 (13.1)	13 (10.0)			
	≥60	19 (14.6)	30 (23.1)			
Sex	Male	37 (28.5)	40 (30.8)	1.2	N/A	*0.6
	Female	23 (17.7)	30 (23.1)			
Marital status	Married	50 (38.5)	57 (43.8)	1.5	N/A	*0.89
	Single	6 (4.6)	10 (7.7)			
	Divorced	4 (3.1)	3 (2.3)			
Frequency of blood transfusion	Nil	17 (13.1)	18 (13.8)	0.8		
	Once	10 (7.7)	13 (10.0)		R=0.6483	0.404
	More than two	33 (25.4)	39 (30.0)			
History of tattoo	Got a tattoo	9 (6.9)	5 (3.9)	2.29	N/A	*0.2
	Without tattoo	51 (39.2)	65 (50.0)			
Duration of dialysis	6month -1 year	7 (5.4)	3 (2.3)	0.3		0.92
	< 1-5years	37 (28.5)	40 (30.7)		R=0.015	
	>5 years	16 (12.3)	27 (20.8)			

OR=Odd Ratio, R= Correlation Coefficients, N/A=not applicable, * p-value of chi-square test.

Table 2: Co-infection of HCV and HBV with HTLV-1 results

HTLV-1 p24	HCV infection NO. (%)		HBV infection (%)	NO.
	Positive	Negative	Positive	Negative
Positive	18 (13.9)	42 (32.3)	5 (3.8)	55 (42.3)
Negative	23 (17.7)	47 (36.2)	5 (3.8)	65 (50)
Total No.	41(31.6)	89(68.5)	10(7.6)	120(92.3)
P-value	0.72		0.79	

Table 2 indicates that other viral infections, such as hepatitis C (HCV) and hepatitis B (HBV) viruses, are associated with HTLV-1 p24-positive patients. Out of the patients, 18 (13.9%) were found to be infected with HCV, and 5 (3.8%) with HBV. There was only one case of combined HCV and HBV infection, and the patient had positive HTLV p24 antigen.

Discussion

As a latent oncovirus, HTLV-1 infection has become a public health concern. The potential risk of HTLV infection is higher among hemodialysis patients, who are more likely to be infected with blood-borne viruses due to impaired immunity, prolonged hospitalizations, recurrent surgical interventions, multiple blood transfusions, or the use of vascular access for dialysis (25).

Although the asymptomatic state of this virus, it can lead to life-threatening neurological and lymphoproliferative diseases like ATL. Insufficient data regarding HTLV-1 prevalence and its associated disease in Iraq, to the best of our knowledge, no previous study about HTLV-1 detection among Iraqi HD patients has been reported to date. However, in 2017, HTLV infection was detected in patients with lymphoma and leukemia with a prevalence rate of 1.2% (26) and among Iraqi blood donors, high prevalence was reported in Baghdad 24(61.5%), Karbala 5(12.8%), Al-Qadisiyyah 4(10.2%), both Al-Najaf and Al-Muthanna had 2(5.1%), and only one positive case of (2.5%) in both Basrah and Wasit province (27).

Screening tests are commonly utilized for the detection of antibodies against HTLV-1/2 in plasma or serum. Subsequently, these tests must be supplemented with confirmatory procedures such as line

immunoassay or molecular methods. Notably, the identification of antigens is a preliminary method used for HTLV diagnosis, as opposed to the identification of antibodies, due to the heightened risk of obtaining false negative results in the detection of HTLV antibodies. This phenomenon is attributed to the prolonged immunological window period following infection. Research conducted with individuals who were infected through blood transfusion revealed a median seroconversion period of approximately fifty-one days, with a range of 36 to 72 days (28). In the present study, HTLV-1 p24 specific capsid antigen was detected among Iraqi hemodialysis patients at a rate of (46.2%), our findings are inconsistent with several studies that observed higher HTLV-1 seropositivity rates among hemodialysis cases (18, 19, 29-31). The data obtained from this study showed that higher HTLV-1 infections were observed in older (50-59) and ≥ 60 age groups, male than female with a mean age of 52 years which coincides with the study found a gradual increase in seropositive rates with age reaching 48.5% in men aged 50 to 64 years (32), male is associated with a higher susceptibility to viral infections related to hormonal as well as sex chromosomal differences as causes for this(33, 34), the reason why HTLV-1 infection is predominantly male in this study can be explained by the high number of hemodialysis participants in this study who were males. Furthermore, our findings show that most of the HTLV-1 infected patients (38.5%) were married dissimilar to the previous studies conducted in Brazil and Japan documented that HTLV-1 detection was higher in females than males(35, 36) and single individuals (35). Explaining the sexual route of transmission may play an important role in our studied patients.

This study revealed that patients who underwent more than two blood transfusions experienced the highest frequency of infections. Numerous studies have elucidated that the transfusion of infected cellular blood serves as an efficient mode of HTLV-1 transmission (37, 38). Furthermore, The implementation of systematic screening of blood, organs, and blood components, along with the adoption of leukoreduction practices, has significantly mitigated the risk associated with the transfusion of blood and its derivatives (39).

This study revealed coinfection rates of 13.9% of HCV-HTLV-1 and 3.8% of HBV-HTLV-1. In contrast, prior research identified HBV with HCV as the most prevalent blood-borne viral coinfection at 2.35%, with a coinfection rate of 1.17% for both HBV and HTLV-1(40). Indicating a notable percentage of co-infection involving viruses transmitted through similar routes, suggesting the need for interventions to address this issue. Another study showed a coinfection rate of 0.5% for HBV/HCV and a rate of 1.1% for HBV/HTLV-1 coinfection, which is lower than what we found (41). Moreover, a survey conducted in Australia in 2017 unveiled a 14.4% seroprevalence of HTLV-1 among HBV-infected patients, a figure nearly higher than our study's results (42). In the context of HCV-HTLV coinfection, available data present conflicting results. The observed variations underscore the impact of studying diverse patient populations across different regions. Several studies indicate that environmental changes, as well as physiological, iatrogenic, and acquired immunocompromise, are the main factors contributing to the emergence and re-emergence of microbial co-infections (43). However, the occurrence and transmission of viral coinfections are significantly influenced by host ecology and interactions between different viruses (44). Some studies indicate an increase in HCV viremia and a reduced likelihood of spontaneous clearance of the infection. Conversely, other studies propose a heightened probability of HCV elimination in individuals coinfected with HIV-1 and HTLV. This propensity is attributed to the immunomodulatory effects of HTLV, resulting from the heightened production of proinflammatory cytokines in this cohort. Furthermore, compelling research indicates a decrease in hepatic damage among individuals who are triple-infected with HIV, HTLV, and HCV, as well as an enhanced likelihood of spontaneous clearance of HCV (45)

Conclusion

The detection of active HTLV-1 antigenemia among Iraqi hemodialysis patients has exhibited a notably high rate of HTLV-1 infection. Consequently, screening for HTLV-1 is imperative to mitigate its transmission within hemodialysis units. This study represents the inaugural investigation in Iraq pertaining to HTLV-1 among hemodialysis patients. Nonetheless, further comprehensive studies encompassing a larger patient cohort across diverse regions of Iraq are imperative.

Limitation Of This Study: Challenges were encountered during the process of sample collection and patient follow-up. It would be advantageous to encompass all HD centers across Iraq to attain a more comprehensive elucidation

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Conflict of Interest

The authors hereby declare the absence of any conflicts of interest.

Data availability

Data are available upon reasonable request.

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