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Frequency of Sexually Transmitted Diseases in Pakistani Blood Donors

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Abstract

Objective: To determine the frequency of HIV and syphilis infection among Pakistani blood donors presenting to blood transfusion services of Shifa International Hospital (SIH) Islamabad through serological testing and to determine accuracy of serological testing for Human Immunodeficiency Virus (HIV) taking nucleic acid testing (NAT) as gold standard.
Methodology: Present study was a descriptive cross-sectional, conducted at SIH Islamabad over a period of six months (July-December 2016). A total of five thousand one hundred and seven (n=5,107) adult blood donors were enrolled and underwent serological testing for syphilis. For HIV, serology was performed followed by NAT screening.
Results: There were 1.3% (n=68/5107) donors came out to be serologically positive for syphilis with a yield of 1:75 or 14686 donors per million. There were 0.2% (n=12/5107) donors came out to be serologically HIV positive with a yield of 1:426 or n=2350 donors per million. All HIV patients on NAT testing proved to be negative for HIV.
Conclusions: A remarkable proportion of blood donors presented at our institute during the study period harbored STD, which can be detected through meticulous and stringent screening of blood donors. NAT has an incremental value in identifying false positive HIV cases on serological testing.
Keywords: Blood Transfusion, HIV, Nucleic Acid Testing, Syphilis

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Introduction

Sexually transmitted infections (STIs) are common causes of morbidity and are preventable by using safe and standard blood transfusion practices.[1] Several studies have demonstrated that both syphilis and HIV can be transmitted through blood transfusion and presence of STIs can facilitate HIV transmission.[2,3] In Pakistan, with a population of about 200 millions, blood required for transfusion is approximately 1.5 million bags/year.[4] During the 1990's and the first decade of current century, practices towards safe blood transfusion (SBT) have considerably evolved in Pakistan. A survey conducted at 37 blood banks in Karachi, Pakistan over a period of ten years showed a remarkable improvement in STI screening.[5] Another survey conducted by both Agha Khan university hospital & Dow university of health sciences, Karachi in 40 blood banks showed that 100% blood banks were performing HBV, HCV & HIV screening on blood donors, though most of them were performing rapid testing.[6] In northern Pakistan, Khattak MF, et al analyzed donation records of donors over the period of five years (1996-2000). They reported that the third generation ELISA technique has been in a consistent use during whole of the study period.[7] Zameer M, et al in a recent study from central Pakistan (Lahore) reported that most of the public sector hospitals in central Pakistan perform rapid and cost effective testing (mostly using immune-chromatography) for HBV, HCV, HIV and syphilis.[8]

Despite putting enormous efforts and investments into the blood transfusion services, transmission of sexually transmitted diseases remains a real possibility. Routine laboratory tests in most blood banks rely on the detection of antibodies. This leaves an immunological window period which could be minimized by using antigen detection or by nucleic acid testing (NAT).[9] The aim of this study was to

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determine the frequency of HIV and syphilis infection among Pakistani blood donors presenting to blood transfusion services of Shifa International Hospital (SIH) Islamabad through serological and NAT testing and to determine accuracy of serological testing for HIV taking NAT as gold standard.

Materials and Methods

It was a descriptive Cross-sectional Study conducted at blood Transfusion Services SIH, Islamabad after ethical approval. Informed consent was taken from all the donors and the data were collected over a period of six months (July to Dec 2016). A total of n=5107 healthy blood donors (taking prevalence of syphilis 0.03%, confidence level of 95% and absolute precision as 0.0015)[10] of either gender between 16 to 60 years of age who reported to SIH Islamabad were enrolled. For the detection of syphilis only serological screening protocol was performed on ROCHE Cobas e 601 multiplex (MP) fully automated analyzer. For HIV, serology was performed followed by NAT screening (Figure 1). Qualitative and quantitative in vitro determination of HIV was performed on COBAS E 601 fully automated analyzer for HIV immunoassay. COBAS R-TAQMAN version 2.0 quantifies the clinically significant HIV groups and subgroups with full subtype coverage and quantifies HIV-1 groups O and M allowing early detection of minute quantities of virus in the blood. The criteria for labeling a donor Positive/Negative/ borderline were according to manufacturer's instructions. All the data were entered and analyzed using SPSS version 21.0.

Results

Age, gender and blood groups distribution is recorded in table I. There were 1.3% (n=68/5107) donors came out to be serologically positive for syphilis with a yield of 1:75 or fourteen thousand six hundred and eighty six (n=14686) donors per million. There were 0.2% (n=12/5107) donors came out to be serologically HIV positive with an HIV yield of 1:426 or two thousand three hundred and fifty (n=2350) donors per million. All the serologically negative HIV cases came out to be negative on NAT and hence added to our inventory. Serologically positive HIV cases were discarded as per our institutional protocol but for the study purpose underwent NAT and proved to be negative for HIV.

Table I: Age, gender and blood groups distribution of donors

Age (Mean years ± SD)	
Total:	25.1±4.4
Males:	25.2±4.4
Females	24.6±4.3
Gender n(%)	
Males	5080 (99.5)
Females	27 (0.5)
Blood Groups n(%)	
B Positive	1570 (30.7)
O Positive	1492 (29.2)
A Positive	1187 (23.2)
AB Positive	444 (8.7)
B Negative	141 (2.8)
O Negative	129 (2.5)
A Negative	101 (2.0)
AB Negative	43 (0.8)

Serology Result	*Serology Interpretation*	*Action Taken*	*Serology Interpretation With 2nd Sample*	*Action Taken*	*NAT Result*	*Final Interpretation On NAT*	*Components Status*
< 0.9 S/CO	Negative	NAT	Don't need repeat Serology	NAT	Negative	Negative	In Inventory
					Reactive	Reactive	Discarded
≥0.9 - <1.0 S/CO	**Initially Borderline Reactive**	Perform Serology in Duplicate with 2nd Sample (Sample from Plts/FFPs)	If Both results are i- < 0.9 S/CO=**Negative**	After Repeat serology: **Negative** NAT	Negative	Negative	In Inventory
			If any of these 02 results is ii- ≥0.9 S/CO = **Repeatedly Reactive**		Reactive	Reactive	Discarded
				Reactive: Discard	Confirmation of discarded Components		
≥1.0 S/CO	**Initially Reactive**	Perform Serology in Duplicate with 2nd Sample (Sample from Plts/FFPs)	Both results are i- < 0.9 S/CO=**Negative**	After Repeat serology **Negative**: NAT	Negative	Negative	In Inventory
			ii- If any of these 02 results is ≥0.9 S/CO = **Repeatedly Reactive**		Reactive	Reactive	Discarded
				After Repeat serology **Reactive: Discard**	Confirmation of discarded Components		

Figure 1: Blood Transfusion Services-SIH -Donor Serology & NAT Protocol for HIV testing

Table II: Positive cases of HIV and Syphilis	
HIV n(%)	
Positive	12 (0.2)
Negative	5095 (99.8)
Syphilis n(%)	
Positive	68 (1.3)
Negative	5039 (98.7)

Discussion

Present study results showed that there were 1.3% (n=68/5107) donors came out to be serologically positive for syphilis with a yield of 1:75 or 14686 per million donors. Globally, the incidence of syphilis among blood donors showed wide variation. Bhatti et al [11] found an incidence of 0.75% among Pakistani donors, a figure close to the present study (1.3%). Saeed M et al in another study among Pakistani population reported incidence 1.55% for syphilis among 18274 blood donors.[12] Nazir S et al reported 3.1% syphilis positive cases among 14,352 tested blood donors. In a quite recent study, Amin H et al reported that out of 9152 blood donors reported at a blood bank at Lahore, 0.68% were found to be positive for sysphilis.[13]

Prevalence of syphilis in blood donors was reported as 0.34% in Yemen[2] and 0.02% in Israel[3] and 7% incidence in India.[14] The figures were remarkably high from Africa. A study from Ghana[15] showed 7.5% incidence and 12.7% from Tanzania.[16] There may be several reasons for these observed variations in the reported incidences. A study showed that incidence of syphilis was higher in replacement donors as compared to the voluntary ones.[16] We, however, in the present study did not take into account this difference due to lack of availability of required information. Another reason is the difference in the amount of organisms present in the blood and their potential of infectivity. Presence of certain risk factors in the blood donors relate closely to the risk of transfusion-transmitted syphilis. One of the most important factors is particular sexual behavior as the disease is transmitted primarily by the sexual route and higher rates were observed among homosexual men.[17] In the present study, we did not find any such association likely due to unavailability of such evidence in the patients' history.

Older age, multiple sexual partners, scarification of skin, intravenous drug use and a pervious history of syphilis are the other factors.[5] In the present study, older aged males (>40 years of age) showed significantly higher prevalence, however, we did not find any association between past sexual activities likely due to lack of such evidence in the patients' history. Different standards of services including quality of equipment, trained personnel and quality of reagents used may be the other reasons. It is recommended that serologic testing for the diagnosis of syphilis should include the use of both treponemal and non-treponemal tests.[18] In the present study, we used only treponemal test.

In the present study, there were 0.2% (n=12/5107) donors came out to be serologically HIV positive with an HIV yield of 1:426 or 2350 donors per million. Moiz B et al in their meta-analysis of published studies over a period of 25 years (1988-2012) reported the pooled prevalence of HIV in blood donors in Pakistan as 0.00111%.[19] Saeed M et al in another study among Pakistani population reported incidence of 0.02% for HIV among 18274 blood donors. They employed rapid immunochromatographic testing technique.[12] In the present study all HIV positive patients went on NAT testing and proved to be negative for HIV.

Similar findings were reported by other studies conducted on transfusion related HIV transmission in Pakistan, which found no HIV NAT– reactive donation. This may be attributed to low prevalence of HIV infection in the general population as well as in blood donors in Pakistan. Even the most sensitive screening technologies currently available cannot identify the presence of HIV infection during the first few days after infection, when neither HIV RNA nor HIV-specific antibodies have reached detectable levels. Based upon evaluation of recipients of blood units from donors who later developed HIV antibody, this window period (time from exposure to seroconversion) was estimated to be 56 days with the first generation antibody assays (early 1980s) and 42 days with the second generation assays (late 1980s). Addition of mini-pool nucleic acid testing (MP-NAT) reduced the window period to 11 days.[20] Despite these procedures, HIV transmission may still occur for several reasons. Some variant strains of HIV may escape detection by current screening assays and due to testing or clerical errors. However, since HIV screening currently includes two independent assays, these errors are significantly minimized. Another reason is donations may be collected during the window period of infection. In 1999, with the addition of MP-NAT, the HIV window period was further reduced. This method uses detection of plasma HIV RNA in samples that are pooled together from 6 to 16 blood donors (or a larger number of plasma donors).[21] Due to the increased sensitivity of HIV MP-NAT for detecting early window period infection, the risk of HIV infection from transfusion has further decreased and is now estimated to be one in approximately 1.5 to 2 million units.[24] In some countries with a very high HIV incidence (eg, South Africa), individual donation (ID) NAT has been implemented, but that incurs high cost. Other countries where incidence is not as high, have different

practices (eg, in China all blood is screened using HIV serology, and the addition of pooled NAT is being phased in at varying rates in different blood centers).[22] In Pakistan, there are currently very few setups where HIV NAT is employed. In SIH, we employed HIV NAT in all donors.

All serologically positive HIV cases in the present study were discarded as per our institutional protocol but for the study purpose underwent NAT screening and proved to be negative for HIV. The main reason of these false positive results is probably the detection of non-specific antibodies on serological testing. Other reasons could be sampling or technical/clerical error. False positive results could also be observed in people who recently had a shot of flu vaccine or have an autoimmune disease. These factors need to be addressed in future studies. Rifkin SB et al reported that blood group O-negative blood and history of sex with an HIV-infected person may increase the rate of false-positive HIV finger-stick results.[23] We, however, did not find any such association in our study. Ndase P et al demonstrated that rapid HIV tests result in a high number of false positive results and recommended effective quality assurance of HIV testing along with mechanisms for confirmatory HIV testing. Moreover, they also emphasized on counseling strategies for persons with positive rapid test results.[24] In summary, NAT has consistently shown an incremental value in identifying false negative and false positive HIV cases on serological testing. We recommend routine use of NAT for HIV testing of blood donors in Pakistan.

Conclusion

Our results revealed that approximately fifteen thousand per million transfusions for syphilis and more than two thousand per million transfusions for HIV may be prevented with meticulous donor screening. NAT has an incremental value in identifying false positive HIV cases on serological testing.

It is recommended that key laboratories at national and regional level should continuously monitor the quality of screening methods. Based on STD enhanced screening protocols, 376 components were interdicted from our inventory over a period of six months. It is therefore strongly recommended that STD enhanced screening protocols should be adopted all over the country to significantly reduce the transfusion induced infection spread. We also recommend large scale national-wide studies employing NAT testing as a complement to traditional serological testing to formulate a uniform policy of blood screening program in Pakistan.

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