Original Article

Seroprevalence of transfusion transmitted infections among voluntary blood donors at a tertiary care teaching hospital in Kolhapur, India

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Abstract

Background: Transfusion of blood is life saving but also has life-threatening hazards and far-reaching consequences for the recipients themselves as well as for their families and their communities. The present study is based on the seroprevalence of human immunodeficiency virus, hepatitis b virus, hepatitis c virus, syphilis and malaria in voluntary donors. This gives information regarding safety associated with blood transfusion and an accurate measurement of risk versus benefits of blood transfusion. The data generated will help clinicians for judicious use of blood as well as awareness regarding the transfusion transmitted infections (TTIS). **Objectives:** The objective of the present study was to assess the seroprevalence and trend of TTIS among blood donors in the blood bank of D. Y. Patil Medical College, hospital and research centre, Kolhapur, India. **Methods:** A total of 5178 samples were screened during a period of seven years from January 2010 to December 2016. Data analysis was done by quickcal software by chi square test. $p<0.05$ is considered significant. The prevalence of TTIS was expressed by donation years and donors' characteristics (age, gender, geographical distribution). **Results:** The overall prevalence of HBV, HIV, HCV, syphilis and malaria was 1.31%, 0.54%, 0.54% and 0.08% respectively, with no reported case of malaria.

Key Words: Voluntary blood donors, seroprevalence, transfusion- transmitted infections.

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INTRODUCTION

Blood transfusion is the biggest treatment modality to save lives of thalassemic children, Disseminated Intravascular Coagulation and Post Partum haemorrhage in women, surgeries, accidents etc. There is a 1% chance of Transfusion Transmitted Infections (TTIs) with each unit of transfusion[10]. A majority of known cases of post transfusion diseases have been caused by human immunodeficiency virus (HIV), hepatitis B virus, (HBV), hepatitis C virus, (HCV), Treponema pallidum and malarial parasites[7]. Currently TTI can be divided into:- A)Viral-HIV, Hepatitis B virus, Hepatitis C virus, Hepatitis A virus, Hepatitis G virus, Human T–cell lymphotrophic virus, Parvovirus B19, Severe Acute Respiratory Syndrome caused by Corona Virus, Epstein Barr virus, Cytomegalovius, West Nile Chikungunya and Zika viruses. B) Bacterial-Treponema pallidum, Yersinia enterocolitica, Escherichia coli, Streptococcus sp. C) Parasitic- Plasmodium sp, Leishmania, Babesia microti. D) Emerging:- Prions. However, NACO (National Aids Control Organization) recommends the testing of 5 Transfusion transmitted Infections (TTIs)- HIV, HBV, HCV, Malaria and Syphilis [22,29]. An unsafe blood transfusion is very costly from both economic and human points of view, resulting in long term morbidity and mortality, delayed viremia and hidden states[25]. This leads to increased requirement of medical care, higher levels of dependency and the loss of productive labour force, placing heavy burdens on already outstretched health, social services and on the national economy. The majority

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of the problems are due to prevalence of asymptomatic carriers in the society as well as blood donations during the window period of infections. Concealing of medical history by captive, paid or professional donors, who widely exist in developing countries, also poses a great threat to safe blood supply[13]. Preventing transmission of infectious diseases through blood transfusion is one of the greatest challenges of transfusion medicine[32]. To understand the magnitude and dynamics of transfusion of a disease in a community and for its control and prevention, the assessment or study of its prevalence is very important. This knowledge might give us the idea of disease burden of the society and the basic epidemiology of these diseases in the community[9]. Hence this study was carried out to assess the seroprevalence and trends of TTIs over a period of 7 years from January 2010 to December 2016 in D. Y. Patil Medical college, hospital and research centre Kolhapur.

MATERIAL AND METHODS

Study Design: The present study was an observational cross-sectional study. Settings: The study was carried out at the blood bank of 'Department of Immunohaematology and blood transfusion' of Dr. D. Y. Patil medical college, Hospital and Research Centre, Kolhapur, Maharashtra state, India. Study Participants: The present study was carried out on the data of voluntary blood donors from January 2010 to December 2016 after the approval of the institutional ethics committee. The blood samples were collected from all male and female voluntary donors at outdoor blood donation camp and in- house blood bank. Replacement donors and those donors who were not fulfilling the "NACO" criteria for blood donation were excluded from the study[23]. The donors first filled up a form which carried the information of their demographic details, medical history regarding risk factors like history of previous surgery, hospitalization, blood transfusion, high risk behavior, pregnancy and lactation, tattoo marks etc. This was followed by pre-blood donation counseling, which included explanation of the procedure of blood donation, post- donation care and the outcome of the TTI tests. The donor consent form and questionnaire forms as per WHO guidelines/ Govt of India[13] with details of donors, quality control registers, TTI registers, issue registers and the results are maintained in the blood bank. There are regular FDA inspections. The venous blood was collected from all eligible male and female donors aged 18 to 60 years after complete physical examination by the blood bank medical officer. Two ml of blood sample was collected in a labeled pilot tube, at the time of collection of blood, from the donor tubing of the blood bag. The sample was further centrifuged at 3500rpm for 5 minutes to obtain a clear, non- haemolysed serum. The samples were tested for HIV, HBV, HCV, Syphilis and Malaria. Screening for HIV was done by fourth generation ELISA, Qualisa HIV 4 for detection of antibodies to HIV ½ and 'O subtypes' and HIV-1 p24 antigen in human serum or plasma (Qualpro group diagnostics Pvt ltd. www.tulip group.com, verna, Goa). Screening for Hepatitis B virus (HBV) was done by Third generation ELISA, HBsAg ELISA 3.0 for detecting HBsAg in human serum or plasma (Qualpro group diagnostics) Screening for Hepatitis C virus (HCV) was done by Third generation ELISA for detecting antibodies to HCV in human serum or plasma (Qualpro group Diagnostics). Syphilis screening was carried out by 'SYPHICHEK(R)-modified TPHA (Treponem Pallidum Haemagglutination)', which qualitatively detects the presence of IgM and IgG class of Treponema specific antibodies in the serum or plasma (Qualpro group Diagnostics) It is a rapid dipstick test, which utilizes the principle of immunochromatogrphy. It is a screening as well as confirmatory test[1]. Rapid diagnostic kit- Malascan plus (zephyr biomedicals, Goa) which detects the presence of Pan malaria specific pLDH released from the parasitized blood cells was used to screen for Malariasl parasite. This is a visual, rapid and sensitive immunoassay for the qualitative diagnosis of infection with P. falciparum and other plasmodium species (P. vivax/ P.malariae/ P. ovale) in human whole blood. Data analysis was done by Chi square test. RESULTS: Out of the total 5178 voluntary blood donors, 4943 were males (96.94%) and only 235 (4.72%) were females (Table 1). The seroprevalence of positive donors in our study was 2.47%. (Table 2).Table 3 denotes that the highest prevalence was observed for HBV (1.31%). Both HIV and Syphilis showed a prevalence of 0.54%; whereas the lowest seroprevalence was of HCV (0.08%). None of the seropositive donors were co-infected with any additional TTIs. Table 4 shows that the highest prevalence of TTIs (53.13%) was within the age group 21-30 years, followed by (21.88%) within the age group 31-40 years. None of the donors aged above 51 years showed prevalence of TTTIs. It is observed that the prevalence of HBV was highest (38.24%) age group 21-30 years followed by 35.29% in age group <20 years. The difference of prevalence of TTIs among different age groups was statistically significant. ($p < 0.05$). As mentioned in Table 5, the prevalence of all the TTIs was higher among males (91.4%) than females (8.59%).The difference of prevalence by gender was statistically significant ($p < 0.05$). There was a significantly ($P<0.05$) higher prevalence of TTIs among rural population (77.34%) as compared to urban population (22.66%).

RESULTS

Table 1: Distribution of volunitary blood donors in the study population

Year	Total	Male	Female	P Value
2010	589	564(95.76%)	25 (4.24%)	
2011	677	613(90.55%)	64(9.45%)	
2012	542	522(96.31%)	20(3.69%)	
2013	689	669(97.10%)	20(2.90%)	
2014	628	604(96.18%)	24(3.82%)	
2015	842	797(94.66%)	45(5.34%)	
2016	1211	1174(96.94%)	37(3.06%)	
Total	**5178**	**4943(95.46%)**	**235%(4.54%)**	**.0001*** **

Table 2: Seroprevalence of Positive Donors

Total NO of Donors	No. of Positive Donors	Seropositvity
5178	128	2.47%

Table 3: Prevalence of HIV, HBsAg, HCV and Syphilis among voluntary blood donors in the study population

Year	Total no of Blood donors	HBsAg	HIV	Syphilis	HCV	total TTIs	P VALUE
		No (%)	No (%)	No (%)	No (%)	No (%)	
2010	589	16(2.72%)	12(2.04%)	3(0.51%)	0	31(5.26%)	
2011	677	7(1.03%)	9(1.33%)	5(0.74%)	1(0.15%)	22(3.25%)	
2012	542	6(1.11%)	2(0.37%)	6(1.10%)	0	14(2.58%)	
2013	689	5(0.73%)	2(0.29%)	4(0.58%)	1(0.29%)	12(1.74%)	
2014	628	2(0.32%)	1(0.16%)	3(0.48%)	1(0.16%)	7(1.11%)	
2015	842	14(1.66%)	1(0.12%)	3(0.36%)	0	18(2.14%)	
2016	1211	18(1.4%)	1(0.085	4(0.33%0	1(0.08%)	24(1.98%)	
Total	**5178**	**68(1.31)**	**28(0.54%)**	**28(0.54%)**	**4(0.08%)**	**128(2.47%)**	**.0001*** **

Table 4: Distribution of blood donors with transfusion transmitted infections according to age

AGE	HBsAg	HIV	Syphilis	HCV	total	p value
YEAR	No (%)	No (%)	No (%)	No (%)	No (%)	
<20	24(35.29%)	0	0	0	24(18.75%)	
21-30	26(38.24%)	19(67.86%)	21(75%)	2(50%)	68(53.13%)	
31-40	14(20.59%)	8(28.57%)	4(14.29%)	2(50%)	28(21.88%)	
41-50	4(5.88)	1(3.57%)	3(10.71)	0		
>51	0	0	0	0		
Total	**68(53.13 %)**	**28 (21.88 %)**	**28(21.88%)**	**4(3.13%)**	**128(100%)**	
P Value					**.0001*** **	

Table 5: Distribution of blood donor with transfusion transmitted infections according to sex

Sex	HBsAg	HIV	Syphilis	HCV	Total
	No (%)	No (%)	No (%)	No (%)	No (%)
Male	60(88.24%)	26(92.86%)	28(100%)	3(75%)	117(91.41%)
Female	8(11.76%)	2(7.14%)	0	1(25%)	11(8.59%)
Total	68(53.13 %)	28(21.88%)	28(21.88%)	4(3.13 %)	128(100%)
P Value	**.0001*** **	**.0001*** **	**.0001*** **	**.31 NS**	**.0001*** **

Table 6: Geographical Distribution of seropositive donors

Area	HBsAg	HIV	Syphilis	HCV	Total
	No (%)	No (%)	No (%)	No (%)	No (%)
Rural	52(76.47%)	21(75%)	22(78.57%)	4(100%)	99(77.34%)
Urban	16(23.53%)	7(25.%)	6(21.43%)	0	29(22.66%)
Total	68(53.13%)	28(21.88%)	28(21.88%)	4(3.13%)	128(100%)
P Value	**.0001*** **	**.0082** **	**.0025** **	**.045** **	**.0001*** **

DISCUSSION

Voluntary blood donors are the cornerstone of a safe and adequate supply of blood and blood products[23]. In the present study we screened only the voluntary donors for prevalence of TTIs. It is comparable to the study by Giri *et al*[9] where only voluntary donors were screened for the prevalence of TTIs. In our study the majority of donors (95.46%) were males, {TABLE 1} compared to (4.54%) females. Other authors have also reported similar finding[2,7,16,21]. Prevalence of TTIs {TABLE 5} was significantly higher ($p<0.05$) among males (91.41%) than females (8.59%). This finding is indicative of risk

behavior of males like outside socialization, polygamy etc[17]. In Kolhapur, women are mostly housewives and this may lead them to avoid outside activities. Moreover, women have lower haemoglobin levels and a higher number of vasovagal reactions[16,25]. In our study the women were deferred for being underweight (<45kg) and having Hb level below the acceptable limit of 12.5g%. The overall seroprevalence of various TTIs among the studied donors {TABLE 2} figured out as 2.47% which intimately simulated observation by Mandal *et al*[19] (2.93%). However contrasting evidences of significantly lower or higher seropositivity also prevails at large[19]. The infectivity of HBV, HIV, HCV and Syphilis amongst Indian donors has been documented as 0.66-12%, 0.084-3.87%, 0.5-1.5% and 0.85-3% respectively[5]. As seen in {TABLE3}, HBV was the most frequently (1.31%) encountered TTI which coincided with the finding of Pallavi *et al*[27] who reported a prevalence of 1.22%. Our finding is statistically significant. ($p<0.05$). A lower seroprevalence of 0.23 % was reported by Fernandez *et al*[7] and Jain *et al* (0.72%)[10.] On the other hand, Garg *et al*[8] and Matte *et al*[20] reported a higher seroprevalence of 2.57% and 7.2% respectively. HBV is transmitted haematogenously and sexually. It is very highly infectious, far more than HIV[1]. The seropositive HBV donors were given post- test counseling and enquired about the past history of jaundice. They were advised to undergo liver function tests and serology marker for HBeAg to know the status of their infectivity. They were also advised about screening of their family members for HBsAg and immunization. Despite the fact that a safe and effective vaccine was available since 1982, India is still placed by WHO in the intermediate prevalence zone for Hepatitis B (2-7%) and estimated to be a home to over 40 million HBs Ag Carriers [28,29]. This is because Hepatitis B vaccination was not a part of our national immunization program till 2011[29] Hepatitis B vaccine is recommended for unvaccinated adults who are at a risk of HBV infection[34]. Voluntary blood donor HBV vaccination is the right approach to prevent this infection[31]. In the present study, the seroprevalence of HIV was found to be 0.54%. Other studies using fourth generation ELISA have reported a lower prevalence 0.37%.[26], 0.26%[27]. Similar finding of 0.54% was reported by Singh at al 2005[30], and 0.53% by Raut *et al*[28]; both used third generation ELISA. Lower seroprevalence was reported earlier.by Chattoraj *et al* (0.1%)[5] Fernandez *et al* (0.03%)[7] and Jain *et al* (0.02%)[10]. However higher seroprevalence of 2%[20] and 11.7%[6] have been reported. The prevalence of HIV has been decreasing in the Indian population supporting the growing awareness of this life–threatening disease[4]. We have also noticed a decreasing trend in seroprevalence of HIV from 2.04% in 2010 to 0.08% in 2016. Our finding matches with Pagaro *et al*[26] who found a declining trend in HIV positivity over a seven year period which may be due to spread of awareness and a true decline in HIV endemicity. In our study most of the HIV seropositive donors were males (92.8%) which is comparable to another study by Makroo *et al*[18] who reported 97.4% male and 2.6% female HIV positive donors. In our present study we followed the WHO strategy I for screening blood donors for HIV. According to this strategy, if the test is negative for HIV antibodies, the blood unit is considered free of HIV and if reactive the unit is discarded. The donors found reactive for HIV by initial assay are directed by blood transfusion services to linked Voluntary Counseling and Testing Centre (VCTC) for counseling and further confirmatory testing[24]. The seroreactive donors were given post– test counseling and were advised to modify high risk behavior and to self–exclude from future donations. Problems of false negative results because of 'window period, asymotomatic carrirers, high genetic variability in viral strains, non sero- converting chronic or immune silent carriers, and technical mistakes stay behind[7,10]. The risk of TTI of HBV, HIV and HCV could be curtailed by foreword of few more sensitive and specific tests for screening of donor's sample' Preface of Nucleic Acid Amplification Testing (NAT) for HIV, HCV and anti hepatitis B core antigen (HBcAg) and IgM for Hepatitis B infection is recommended to identify the infections during window period[10]. The seroprevalence of Syphilis in our study was 0.54% which is comparable to that of Mandal *et al*(0.65%)[19]. A lower prevalence of 0.01%, 0.06%, and 0.07% was observed by others[10,7,9].However Singh *et al* (2.6%)[30], Srikrishna *et al* (1.6%)[32] Matte *et al* (1.5%)[20] and Chatoraj *et al* (0.92%)[5] have reported higher prevalence rates. The risk of transfusion transmitted syphilis is particularly high in developing countries with limited blood supplies where the blood is collected from family donors and transfused within hours[11]. Treponema pallidum is inactivated by storing blood for 48-72 hours at 6^{o}C. The refrigeration of transfusion blood has therefore greatly reduced the risk of transmitting Syphilis[25]. Various strategies have been proposed by the WHO, International Society of Blood Transfusion and American Association of blood Banks to prevent transfusion-transmitted syphilis. These include: i) selection of low- risk donors and screening for the disease using efficient laboratory methods; ii) application of pathogen reduction technology; and iii) rational use of blood products. However, blood safety begins with implementation of organized blood centres, a quality system, haemovigilance programmes and adherence to Standard Operating Procedures[11]. With regards to HCV, this study shows a low prevalence rate of 0.08% which is

closely similar to Fernandez *et al* (0.05%)[7]. Lower prevalence rates of 0.001%, and 0.01% have been reported in India[4,10]. However Srikrishna *et al* (1.02%)[32] and Giri *et al* 2012 (0.74%)[8] have observed higher prevalence rates. The wide variations in HCV prevalence in India might be due to the use of different generation ELISA test kits having different sensitivities and specificities as well as increased awareness among donors. The low prevalence of HCV when compared with HBV might be due to the fact that HCV is less infective when compared with HBV and HCV is transmitted mainly through transfusion of infected blood or blood products, iv drug abuse and needle sharing which may not be common in our region. HCV positive donors were informed about their disease, counseled, referred to medicine department and prevented from future donations permanent. Malaria is endemic in many tropical and subtropical regions of the world. Over 300million people worldwide are infected, with 1 million fatalities annually. Though malaria constitutes a big health problem in the general population, its prevalence among blood donors is low and ranges from 0% to 0.5%[35]. None of the donors in our study were positive for malarial parasite. Similar results were observed by other authors[7,27]. This may be due to the fact that infection with malarial parasite results in development of fever and weakness. Because of the prominent signs and symptoms majority of infected persons will not visit the blood donation camp/centre and even if they come, will be readily excluded by medical fitness examination and counseling[28]. The good news is that there was no co- infection noted between HIV and other infections in our study. Some authors have observed definite correlation between HIV and other infections probably because of the similar mode of transmission, high risk patient population and immunosuppresion[14,32,33]. TABLE 4 shows that in our study the highest prevalence of TTIs (53.13%) was in age group 21- 30 years (53.13%) and the 31-40 years age group showed the second highest clustering of TTIs (21.88%). In both the age groups HBV infection was predominant (p<.0.5). A statistically significant (p<0.05) HIV positivity (67.86%) was seen in age group 21-30 years followed by 28.57% rate in the age group 31-40 years. Our finding is coinciding with the study by Khalid *et al*[12]. This observation is worrisome since the most productive and economically viable age group from the population is worst hit. There is a need for renewed intensification of preventive programs aimed at high risk behavioural change[33]. The WHO theme for world blood donor day 2015 focuses more on recruitment of young donors as they are the main source of safe blood. Once these young individuals enter the donor population there can also be the possibility of reduction in TTI prevalence since they are counseled before blood donation[27]. Prevalence of TTIs {TABLE 5} was significantly higher (p<0.05) among males (91.41%) than females (8.59%). This finding is indicative of risk behavior of males like outside socialization, polygamy etc [17]. There was a significantly (p<0.05) higher prevalence of all the TTIs in rural populations (77.34%) as compared to urban population (22.66%). Nada *et al*[25] have reported similar finding.

CONCLUSION

A significant percentage of apparently healthy donors harbor TTIs. Our study showed an increase in Hepatitis prevalence, poor participation of females and significant number of HBV prevalence in age groups 21-30 years. All the TTIs were significantly higher in rural area as compared to urban area. Promoting strict screening with authentic and reliable donor history, using more sensitive techniques for diagnosis, treatment, counseling, follow-up and continuing education among blood donors are very important measures to control the transmission of TTIs. Safe and effective HBV vaccination can play a target role in prevention and control of HBV infection. It is important to incorporate more women in the group of regular donors by reducing the obstacles that prevent them from donating. Implementing the National Blood Donor Vigilance Program (NBDVP), which is a comprehensive, centralized and well-structured, approach to collect, collate and analyze data to continuously improve donor safety and satisfaction so that the donors feel well-treated and well taken care of, may motivate blood donors to continue as repeat donors and will have a positive impact on National Blood Supply[3].
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