INTRODUCTION

Human blood transfusion is one of the most important components of therapeutic and surgical treatments due to the lack of artificial blood replacements (Fessehaye et al., 2011). Blood transfusions are directed to save people's lives but if this supply is unsafe then it may transfer many blood-borne infectious agents to the recipients resulting in transfusion transmissible infections (TTIs). The persons infected with these agents present an increased risk of transmission of TTIs to the surrounding population (Allain et al., 2004; Ahmed et al., 2009). There has been a rapid increase in TTIs mainly including Human immunodeficiency virus (HIV), Hepatitis B, Hepatitis C, Syphilis, Human T cell lymphotropic virus, Cytomegalovirus, Malaria, Brucellosis and Toxoplasmosis due to unsafe blood supplies (Ahmed et al., 2009; Fessehaye et al., 2011). Therefore, careful selection of donors, thorough screening of the blood supply, along with blood grouping and storage maintenance are key constituents to provide safe, healthy and cost-effective treatments to the patients (Allain et al., 2004; Ahmed et al., 2009).

Pakistan faces a continuously increasing demand of blood transfusions especially in the case of thalassemia, hemodialysis and hemophilia patients along with road side injuries. In Pakistan, there are approximately 100,000 patients of thalassemia major with their lives totally dependent on blood transfusions (Ishfaq et al., 2013). While, hemophilia is another important disease requiring frequent blood transfu-
sions with frequency of 1 hemophilic patient in every 7000 individuals (Iqbal et al., 2013). This high frequency of multiple-transfusion blood recipients is particularly prone to develop TTIs and limiting the duration of their life expectancy.

The routine laboratory diagnostic testing is very costly in Pakistan therefore general population is reluctant to go for repetitive health check-ups and diagnostic procedures. Moreover the lack of awareness and unavailability of health insurance policies among general population results in the failure of routine blood screening practices. So many people carry infectious agents of potential life-threatening diseases such as Hepatitis B virus (HBV), Hepatitis C virus (HCV) and Human Immunodeficiency virus (HIV) until signs and symptoms appear at very serious stages of ailment. Such type of careers are persistent threat to the community when donate blood without strict screening.

The present study was conducted to determine the prevalence of HIV, HBV, HCV, *Treponema pallidum* (TP) and malarial parasite (MP) among apparently healthy blood donors of Multan, Pakistan during the year 2013. This type of study in resource constrained areas is not only helpful in limiting TTIs but also provide information about the prevalence of infections in the blood donors and reveal the epidemiology of these infections in the resident population which help in adapting the preventive strategies and making government policies.

**MATERIALS AND METHODS**

**STUDY POPULATION**
The study population constituted the subjects who donated blood at Nishtar Medical College and Hospital (NMCH), Multan during the year 2013. The donors were volunteer, honorary for donations and in most of the cases relatives of the patients under treatments. During 2013 a total of 48020 subjects donated blood at the blood bank of hospital. Donors were of both sexes, different age groups from different nearby districts of Multan, Pakistan.

**PRIMARY SCREENING**
Donors were screened primarily by asking question about current health status, any previous infection, and blood donation or transfusion history. Any individual suffering from disease, any previous history and donation within three month were rejected primarily.

**BLOOD SAMPLE Analysis**
Blood samples were collected from the donors following the guidelines of standard venipuncture by National Committee for Clinical Laboratory Standards (NCCLS, 2007). Blood samples were then screened for the presence of antibodies/antigens against HCV, HIV, TP, HBV and MP through rapid immunochromatographic (ICT) assays, as described in Table 1. These ICT assays are easy, quick and visually observable qualitative membrane-based tests that can rapidly detect antibodies or antigens present in the whole blood, serum or plasma through color development on the ICT strips. Sensitivity of these immune strips declared as 1 ng/ml.

**STATISTICAL ANALYSIS**
The percentage frequency of various viral, bacterial and parasitic blood-borne diseases among healthy blood donors of Multan were evaluated through the use of Microsoft Excel 2010. Furthermore, 95% confidence interval for the mean of detected infectious agents among blood donors was also calculated.

**RESULTS**
In the present study, a total of 48020 units of blood were collected from the volunteer blood donors at NMCH, Multan during the year 2013. Among the total 48020 bleedings, 45179 (94.08%) were found healthy and safe for

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**Table 1: Screening assays used for detection of blood borne pathogens**

<table>
<thead>
<tr>
<th>Pathogen</th>
<th>Name of kit and manufacturer</th>
<th>Identification criteria (Antigen/Antibody)</th>
<th>Reference values (positive/negative results)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hepatitis B Virus (HBV)</td>
<td>HBsAg test product (latex conjugate) by ABON Biopharm (Hangzhou) Co. Ltd.</td>
<td>Antigen</td>
<td>Colored band on test region/no band on test region</td>
</tr>
<tr>
<td>Hepatitis C Virus (HCV)</td>
<td>Hepatitis C Virus HCV rapid diagnostic kits by ASSURE Tech.(Hangzhou) Co. Ltd.</td>
<td>Antibody</td>
<td>Colored band on test region/no band on test region</td>
</tr>
<tr>
<td>Human Immunodeficiency Virus (HIV)</td>
<td>HIV (1+2) home test kit/ HIV rapid test kit by ASSURE Tech.(Hangzhou) Co. Ltd.</td>
<td>Antibody</td>
<td>Colored band on test region/no band on test region</td>
</tr>
<tr>
<td><em>Treponema pallidum</em> (TP)</td>
<td>TP Eco test kit by ASSURE Tech.(Hangzhou) Co. Ltd.</td>
<td>Antibody</td>
<td>Colored band on test region/no band on test region</td>
</tr>
<tr>
<td>Malarial Parasite (MP)</td>
<td>ABON™ Plus Malaria P.f/Pan Rapid Test Device by ABON Biopharm (Hangzhou) Co. Ltd.</td>
<td>Antigen</td>
<td>Colored bands on Pf and/or pan line regions/no band on either of these two regions</td>
</tr>
</tbody>
</table>
transfusion while the remaining 2841 (5.92%) donors were infected and therefore not suitable for blood donations (Figure 1). During the study period, HCV was found to be the most frequent infection among blood donors with a total frequency of 3.44% followed by HBV (2.32%), TP (0.07%) and MP (0.06%) as shown in Figure 2. Interestingly, prevalence of HIV was reported as the least TTI with frequency of 0.01%. HBV and HCV were the most prevalent infections throughout the year. Malarial cases were significantly high in number during the months of January and December followed by February and March as depicted in Figure 3.

![Figure 1: Distribution of healthy and infected blood donors among total bleedings received at Nishter Medical College and Hospital, Multan during the year 2013](image1)

![Figure 2: Frequency of potential transfusion-transmissible infectious agents detected among blood donations at NMCH, Multan during the year 2013](image2)

![Figure 3: Month-wise incidence of MP among blood donors of Multan during the year 2013](image3)

<table>
<thead>
<tr>
<th>Infectious Agents</th>
<th>Number of infected individuals</th>
<th>Infected individuals (%)</th>
<th>Mean value</th>
<th>95% Confidence interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hepatitis B Virus (HBV)</td>
<td>1116</td>
<td>2.32</td>
<td>93</td>
<td>9.26</td>
</tr>
<tr>
<td>Hepatitis C Virus (HCV)</td>
<td>1654</td>
<td>3.44</td>
<td>137.8</td>
<td>16.98</td>
</tr>
<tr>
<td>Human Immunodeficiency Virus (HIV)</td>
<td>6</td>
<td>0.01</td>
<td>0.5</td>
<td>0.63</td>
</tr>
<tr>
<td>Treponema pallidum (TP)</td>
<td>35</td>
<td>0.07</td>
<td>2.91</td>
<td>3.17</td>
</tr>
<tr>
<td>Malarial Parasite (MP)</td>
<td>30</td>
<td>0.06</td>
<td>2.5</td>
<td>3.49</td>
</tr>
</tbody>
</table>

DISCUSSION

Transfusion transmitted diseases have an increased significance worldwide because they contribute towards the spread of infectious diseases to the population while proper screening of donors can considerably reduce the possibility of transmission (Arora et al., 2010). In the developing countries, along with various other factors, poor socioeconomic conditions favour the prevalence of infectious diseases among population such as Hepatitis B, Hepatitis C and HIV. The present study was aimed to determine the frequency of HBV, HCV, HIV, TP and MP among blood units donated at NMCH, Multan during the study period spanning from January to December, 2013.

In the present study, a total of 48020 subjects donated blood at NMCH, Multan, PK during the 12 months period of the year 2013 and among these blood donors, 45179 (94.08%) were found healthy and free from infections while the remaining 2841 (5.92%) were infected with one or another infectious agents being tested. The results of a study conducted at the Federal Government Polyclinic
Hospital, Islamabad, Pakistan indicated that among blood units received for donation, 85.66% donors were healthy while 14.34% donors were infected with any of the five WHO recommended transfusion-transmitted infectious agents (Waheed, 2012). The frequency of infections among voluntary blood donors was considerably less in the present study as compared to those of the above described study of Pakistan.

The present study revealed that the most prevalent infectious agent found among blood donors at NMCH, Multan, PK was HCV (3.44%) followed by HBV with a frequency of 2.32%. Results of the present study indicated an increase in the prevalence of HCV than one of the preceding studies of Pakistan reporting positive HCV test among 1.24% blood units donated at Shaukat Khanum Memorial Cancer Hospital and Research Centre, Lahore during the year 2005 (Sultan et al., 2007). This relative increase in the prevalence of HCV from previous study of Shaukat Khanum Hospital may be due to improper and excessive use of contaminated injections, re-usable vials, intravenous drug intake, sharing households with HCV patients and for sure the socio-economic status of the people favoring the viral transmission (Bari et al., 2001; Sultan et al., 2007). Likely, comparing the results with another study of Pakistan highlighted the increased prevalence of HBV among blood donors in the present study (Shah et al., 2010). Whereas, the sero prevalence of TTIs among Chinese population revealed less frequencies of HBV and HCV (0.86%, 0.51%) as compared to that of Pakistani population (Li et al., 2012). Higher prevalence of HBV and HCV as compared to other potent infectious agents among blood donors had been accredited to the viral transmission during their window period, chronic and asymptomatic hepatitis C infections, unavailability of vaccinations and inability to detect occult HBV infection (HBV DNA in blood) through routine HBsAg testings (Khattak et al., 2002; Bhattacharya et al., 2007).

The present study exhibited lowest frequency of HIV (0.01%) among blood donors of Multan, Pakistan. One of the related studies displayed 0% frequency of HIV among voluntary blood donors of Pakistan during the year 2004-2005 (Sultan et al., 2007). While, HIV prevalence among blood donors of Indian and Albanian populations presented comparatively higher 0.23% and 0.03% frequencies, respectively (Chandra et al., 2009; Durro et al., 2010). However, according to the National AIDS Control Program (NACP), 0.05% people in a country of 180 million population are afflicted with HIV, while the prevalence among general population is below 0.1% regardless of high risk group individuals (Waheed, 2012).

The frequency of TP, causative agent of syphilis was reported as 0.07% in blood donors of this study. Whereas, according to one of the studies of the population of Islamabad, Pakistan, a positive test for syphilis was reported among 0.89% of blood donor during the time period from 2010–2011 (Waheed, 2012). While a surveillance report revealed that 0.34% Australian blood donors had been found positive for syphilis (Lucky and Seed, 2011). Malarial parasite is another potent agent to be transmitted through transfused blood because it remains alive in refrigerated and frozen blood for a time period of weeks. While, screening of blood for malarial parasite is usually not carried out in developed countries but endemic areas like Pakistan should strictly screen blood specimens for malarial parasites as well. Malaria poses a greater threat to the population of Pakistan especially through blood donations because mostly blood donors in developing countries belong to low income class and are commercial donors therefore increasing the risk of transmission to the society (Ali et al., 2013). The present study revealed prevalence of 0.06% positive malarial parasite cases among blood donors at NMCH, Multan, PK whereas, the previous study conducted in Peshawar, Pakistan reported 0.57% positive MP cases among blood donations at 3 blood banks of the city (Ali et al., 2013). This study reflected a decline in malarial parasites among blood donors of Multan that might be due to primary screening and geographical as well as climatic variations that directly affect the reproduction of parasite.

As climatic and temperature variations have influence over MP so month wise prevalence was compiled and MP cases were reported in the months of January and December followed by February, March and November. The temperature, humidity and rainfall influenced the transmission of vector throughout the population of Multan, PK during these months that resulted in increased prevalence of malaria during this period. Results of this study were in accordance with a previous study conducted in the North West Frontier Province (NWF) of Pakistan that displayed higher prevalence of malaria in December and January due to the impact of temperature and humidity on mosquito breeding and transmission (Bouma et al., 1996).

There are approximately 1.5 million transfusions in Pakistan every year (Sultan et al., 2007). The risk of TTI is significantly higher among multi-transfused patients of fatal chronic illnesses like thalassemia, hemophilia and chronic dialysis (Ali et al., 2009). A large number of thalassemia patients exhibit an increased prevalence of transfusion transmitted HBV (8.4%) and HCV (56.8%) (Waheed, 2012). This threat can only be reduced by appropriately following the guidelines for the selection of blood donors and screening of blood and blood-products.

Furthermore, advanced techniques with high specificity and sensitivity such as ELISA and PCR should be practiced routinely at blood bank facilities due to the low spec-
Specificity and sensitivity of immunochromatographic methods (Abbas et al., 2008). The transfusion systems are lacking management and control that lead to the increased prevalence of TTIs in population. Therefore, there must be an active organization that keeps record of all transfusion providing bodies throughout the country and a proper check on their donor selection, blood screening procedures. Post-transfusion infections among recipient must also be followed up to improve the screening.

ACKNOWLEDGEMENT

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CONFLICT OF INTEREST

The authors declare that there is not any conflict of interest regarding this study.

REFERENCES