# Seroprevalence and Determinants of Hepatitis-C Virus Infection in **Blood Donors of Lahore, Pakistan**

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Abstract.- The current study was designed to investigate the seroprevalence of hepatitis C in blood donors and also to find out the risk factors associated with its occurrence. An analytical cross sectional study was conducted at blood banks of various public and private hospitals of Lahore during the year 2009. A total of 245 blood donors were selected through systematic random sampling technique. Blood samples were collected and processed to confirm the anti-HCV status and data was collected on pre tested questionnaires to identify the risk factors. The seroprevalence of HCV was estimated to be 17.78%. Female to male ratio was 1:16.5. It was found that the occupational status (p=0.002, OR=3.789, 95% CI=1.515-9.473), place of surgical treatment (p=0.035), history of blood transfusion (p=0.000, OR=6.364, 95% CI=2.38-17.02), ever pricked by sharps (p=0.045, OR=2.893, 95% CI=0.979-8.543), habit of injecting drugs (p=0.04, OR=2.45, 95% CI=0.995-6.062) and glass sharing (p=0.017, OR=2.327, 95% CI=1.145-4.731) were significantly associated with occurrence of hepatitis C in blood donors. The overall prevalence of HCV in blood donors was very high that could increase the spread of disease which demands the use of a highly sensitive screening test before blood transfusion.

Keywords: Prevalence, hepatitis C, risk factors, screening, blood donors.

## **INTRODUCTION**

The etiological agent of hepatitis C was identified in late 80's (Ryan and Ray, 2004) and was classified under the domain of family, Flaviviridae. It is a single stranded RNA virus, consists of 9600 nucleotide bases (Kato, 2000). The major source of hepatitis C virus (HCV) infection includes the infected blood, its products and the other body fluids. Risk factors like intravenous drug injecting, reuse of syringes, dental procedures, use of infected razor, pricking by sharp objects, infected sexual partner and tattooing also play an important role in occurrence of HCV infection (Ajacio and Sandra, 2002). World Health Organization (WHO) estimated a prevalence of 3 percent with a total disease load of 200 million people worldwide (WHO, 2011). Before the development of screening

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tests blood transfusion was strongly associated with HCV infection. Diagnosis of hepatitis C is usually made through ELISAs.

Some of the countries have eliminated hepatitis C through routine screening of blood donors (Busch et al., 2005), whereas in other countries transfusion of blood is a major source of transmission of the disease. Furthermore, some poor countries have not prioritized the blood safety to stop HCV transmission due to lack of funds (Hladik et al., 2006).

Consecutive screening data of HCV markers in voluntary blood donors are useful to estimate the incidence of HCV infection among the general population, which are considered to be healthy in general. There have been some cohort studies on blood donors in the United States, Japan and Italy, but the number of cases in these reports is too small to draw conclusions regarding the demographic characteristics and risk factors associated with HCV infection. Furthermore, studies regarding the seroprevalence of HCV infection have also been conducted in Pakistan but risk factors remained

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ignored, so the current study was designed to find out the risk factors associated with HCV infection.

## **MATERIALS AND METHODS**

Present project was designed to estimate the seroprevalence and risk factors associated with hepatitis-C virus infection in blood donors of Lahore metropolitan.

An analytical cross sectional study was conducted during 2009. All the blood donors who donated blood in the blood banks of five public (Mayo Hospital, Sir Ganga Ram Hospital, Services Hospital, Jinnah Hospital and Lahore General Hospital) and two private hospitals (Farooq Hospital and Ghurky Trust Hospital) for their relatives or other members of their community were included in the study. A total of 245 blood donors were selected through systematic random sampling technique. Blood samples were collected aseptically with informed consent from the antecubital vein either of the inside elbow or from the back of hand with the help of phlebotomist to confirm the anti-HCV status through ELISA. Third generation ELISA Kit (ETI-AB-HCVK-4, DiaSorin S.P.A. Italy) containing 96 wells was used for Enzyme Linked Immunosorbent Assay (Rebuzzini, 2008). The concentration of anti-HCV present in the specimen was measured by photometer (Labsytem Multiskan Biochromatic). A specially designed and pre tested questionnaire was used to collect information regarding the demographic characteristics and other related risk factors associated with blood borne viral transmission.

## Statistical analysis

All the quantitative data was presented in the form of frequency, percentage and mean  $\pm$  S.D. SPSS version 16.0 was used for statistical analysis. Chi-Square was used to see the association of risk factors with respect to anti-HCV status of the blood donors. Odds ratio was calculated for assessment of risk regarding certain factors. A p-value<0.05 was considered as statistically significant.

## RESULTS

#### Prevalence

The overall prevalence of HCV in blood

donors of Lahore metropolitan was estimated to be 17.78% as given in Table I.

Table I	Prevalence	of	hepatitis	С	virus	infection	in
	blood donor	rs o	f Lahore 1	net	ropolit	an.	

	Blood donors	
Individuals tested	245	
*HCV positive (%)	37 (17.78%)	
Total population	42600	
Affected population	7574	

#### \*HCV= Hepatitis C virus

Blood donors were selected from healthy population of Lahore and tested for anti HCV. Among 245 blood donors, 37 (17.78%) were reactive. Mean age among reactive and non-reactive blood donors was 28.64±7.31 and 29.44±8.83 years respectively. According to p-value no significant difference (p-value>0.05) was present among the age of reactive and non-reactive blood donors. Among 37 reactive blood donors, 34 were male and 3 were female. Mean age of reactive male and  $28.52 \pm 7.52$ female blood donors was and 30.00±5.00 years respectively. In terms of p-value age of reactive male and female blood donors was insignificantly different (p-value>0.05) as shown in Table II.

Table II.-Distribution of reactive (Positive) & non-<br/>reactive (Negative) blood donors according to<br/>gender

Gender	Anti-HCV Status	Number	Age (Mean±SD)	p-value
Male	Reactive Non-Reactive	34 197	28.52±7.52 29.36±8.73	0.602
Female	Reactive Non-Reactive	03 11	30.00±5.00 30.90±10.70	0.893

Independent sample *t*-test was used to see the difference at 5%  $\alpha$  level

#### Risk factors

Table III elucidates the distribution of HCV reactive and non-reactive blood donors according to their demographic characteristics. It was found that the occupational status of the blood donors was significantly associated (p=0.002, OR=3.789, 95% CI=1.515-9.473) with HCV infection. The blood

Demographic characteristics		Anti-	HCV status	p-value	Odds	Confidence interval	
		Reactive	Non-Reactive		ratio		
Gender	Male	34	197	0.496	1.580	0.419-5.960	
	Female	03	11				
Geographical status	Punjabi	36	203	0.913	0.886	0.1006-7.812	
	Pashton	01	05				
Marital status	Married	22	111	0.493	0.780	0.383-1.588	
	Unmarried	15	97				
Educational status	Illiterate	30	149	0.233	1.697	0.7066-4.076	
	Educated	07	59				
Occupational status	Private job	31	120	0.002*	3.789	1.515-9.473	
•	Public job	06	88				
Socioeconomic status	5000-10000	27	116	0.106	-	-	
	11000-30000	10	84				
	>40000	00	08				

Table III	Distribution of hepatitis C	virus	reactive	and	non-reactive	blood	donors	according	to their	demographic
	characteristics									

Chi-Square test was applied and p<0.05 at 95% confidence interval was considered as significant

\*p=value is significant

donors who were doing private jobs were more prone to get the HCV infection as compared to public sector employees. In terms of p-value and OR gender, ethnicity, marital status, educational status and socioeconomic status were insignificantly associated with anti-HCV status.

Table IV shows the exposure of HCV reactive blood donors to various indicators. Among 37 reactive blood donors, 8 were having history of surgery, 9 had blood transfusion, 4 had under gone dental procedures, 1 blood donor had tattoo on his body and 25 had received injections during their treatment for some diseases. In terms of p-values factors like history of blood transfusion (p=0.000, OR=6.364, 95% CI=2.38-17.02), being pricked by sharps (p=0.045, OR=2.893, 95% CI=0.979-8.543), injecting drug user (p=0.04, OR=2.45, 95% CI=0.995-6.062) were significantly associated with occurrence of hepatitis C. Place of surgical treatment was also found to be significantly different (p=0.035) in blood donors. Whereas all other factors like history for any road accident, usage of drugs prescribed, type of drugs oral or injectable, alcohol intake, history of relatives having any disease and use of razor from barber/beauty

salon were insignificantly associated with anti-HCV status.

Table V shows the effect of sharing utensils and other items by blood donors on their anti-HCV status. Glass sharing was significantly associated (p=0.017, OR=2.327, 95% CI=1.145-4.731) with anti-HCV status for blood donors. Odds ratio shows that the blood donors who shared glass with others were 2.327 times more prone to HCV infection. Other utensils' sharing was not significantly associated with anti-HCV status i.e. Comb sharing (p>0.05, OR=0.550), spoon sharing (p>0.05, OR=0.572), towel sharing (p>0.05, OR=0.818), straw sharing (p>0.05, OR=1.417), razor sharing (p>0.05, OR=1.898), nail cutter sharing (p>0.05, OR=0.976), tooth brush sharing (p>0.05, OR=1.898). Straw, razor and tooth brush sharing were found to be weakly associated risk factors for HCV infection as the odds ratio was slightly high.

#### DISCUSSION

The modern world has developed certain measures which results in the reduction of HCV prevalence. These measures include screening of

Indicators	Dognonge	Anti-H	Anti-HCV Status		Odda matta	Confidence
indicators	Response	Reactive	Non-reactive	p-value	Odds radio	interval
Have you ever visited abroad	Yes	02	10	0.877	0.884	0.186-4.207
	No	35	198		Odds ratio 0.884 0.495 - 6.364 0.782 3.000 0.655 2.893 1.019 0.526 - 2.45 1.180 0.6203 - 0.4267	
Undergone any surgery	Yes	08	25	0.115	0.495	0.204-1.203
	No	29	183			
From where you got surgical treatment	Private Sector	02	13	0.035*	-	-
, , , , , , , , , , , , , , , , , , ,	Public Sector	06	10			
	Not Applicable	29	185			
Did you receive blood transfusion	Yes	09	10	0.000*	6.364	2.38-17.02
5	No	28	198			
Undergone any dental procedure	Yes	04	18	0.672	0.782	0.249-2.455
	No	33	190			
Have you any tattoo on body	Yes	01	16	0.271	3.000	0.386-23.336
5 5 5	No	36	192			
Received injections during treatment	Yes	25	120	0.260	0.655	0.312-1.374
, <u> </u>	No	12	88			
Have you been pricked by sharp	Pricked	33	154	0.045*	2.893	0.979-8.543
5 1 5 1	Never Pricked	04	54			
Have you got any road accident	Yes	10	57	0.962	1.019	0.464-2.239
	No	27	151			
Do you use drugs prescribed by	Doctor	36	205	0.577	0.526	0.0530-5.205
	Non-Doctor	01	03			
If yes what type of drugs	Oral	04	15	0.634	-	-
	Inject able	01	03			
	Both	32	190			
Are you an injecting drug user	Yes	08	21	0.04*	2.45	0.995-6.062
	No	29	187			
Do you use Alcohol	Yes	00	03	0.462	1.180	1.119-1.245
-	No	37	205			
Do you have any relative having	Hepatitis	04	34	0.391	0.6203	0.2063-1.865
	No Disease	33	174			
If yes what kind of relationship	Sexual	01	03	0.480	-	-
· •	Non- Sexual	03	31			
	Not applicable	33	174			
Visited barber/beauty saloon	Razor used	32	195	0.118	0.4267	0.1425-1.278
-	Razor not used	05	13			

Table IV.- Summary of association between hepatitis C and various indicators about blood donors.

Chi-Square test was applied and p<0.05 at 95% confidence interval was considered as significant

\*p=value is significant

blood or blood products along with laboratory tests for liver disease, questioning blood donors about HIV risk factors, anti-HCV testing, and HCV nucleic acid testing (Busch *et al.*, 2003).

This study included apparently healthy blood donors who are considered to be more important in transmission of HCV to others. In our study out of total 245 apparently healthy blood donors, 37 (17.78%) were found reactive for anti-HCV ELISA. Our results are contrary to the findings of Ryas *et al.* (2001), Rehman *et al.* (2002), Asif *et al.* (2004),

Farooqi *et al.* (2007), who reported the prevalence of hepatitis C as 4.7%, 4.1%, 5.14% and 3.21%, respectively. The difference in prevalence percentage might be due to the difference in diagnostic techniques as the above studies use Immuno-chromatographic test (ICT) as compared to the present study which applied third generation ELISA. Furthermore, the above mentioned studies were conducted few years ago and during that time the disease burden might be less in the country and as the time has progressed the prevalence increased

Items shared		Anti-H	ICV status	p-value	Odds ratio	Confidence interval
		Reactive	Non-reactive	_		
Comb sharing	Yes	33	195	0.314	0.550	0.169-1.790
U	No	04	13			
Glass sharing	Yes	21	75	0.017*	2.327	1.145-4.731
-	No	16	133			
Spoon	Yes	34	198	0.409	0.572	0.150-2.187
-	No	03	10			
Towel sharing	Yes	34	194	0.761	0.818	0.223-2.998
	No	03	14			
Straw sharing	Yes	01	04	0.757	1.417	0.154-13.04
	No	36	204			
Razor sharing	Yes	01	03	0.577	1.898	0.192-18.75
	No	36	205			
Nail cutter	Yes	33	186	0.966	0.976	0.316-3.014
	No	04	22			
Tooth brush	Yes	01	03	0.577	1.898	0.192-18.75
	No	36	205			

Table V.- The effect of sharing utensils and other items by blood donors from healthy population of Lahore metropolitan.

Chi-Square test was applied and p<0.05 at 95% confidence interval was considered as significant

\*p=value is significant

many folds due to factors like reuse of syringes, not using the disposable kits in blood donation, dialysis and even in the surgical procedures. The increased prevalence in our study can be well explained by relating it to the findings of Idrees *et al.* (2008), who claimed the overall seroprevalence of HCV among blood donors to be 7.2% in 2004 which increased significantly (8.9%) in 2007 (p=0.02).

Blood transfusion is a very important route of transmitting and getting HCV infection (Busch *et al.*, 2003). In our study female to male ratio was 1:16.5 which might be due to the cultural norms of the country.

The mean age of reactive and non-reactive blood donors was 28.64 years and 29.44 years, respectively. Blood transfusion (Gaeta *et al.*, 1999)and intravenous drug abuse have repeatedly been found to be the independent risk factors of HCV infection, although some studies failed to prove this (Win *et al.*, 1994). Other risk factors such as low education level (Goodrick *et al.*, 1994), male gender (Crawford *et al.*, 1994), tattooing (Conry *et al.*, 1996) and surgical procedures are also identified as risk factors in some studies. Results from above mentioned studies are compatible with present study. Among reactive, most of the donors were illiterate and belonged to poor socioeconomic status.

Results of another study carried out in Thailand reported that history of surgery, blood transfusion, and ear or body piercing were associated with HCV infection (Lakkana et al., 2004). According to the results of present study history of surgery and blood transfusion were significantly associated with anti-HCV status among blood donors whereas tattooing on the body was found to be the risk factor as the odds ratio was 3.0. Injecting drug abuse was the strongest risk factor for HCV prevalence among blood donors in northern Thailand. This finding is similar to other reported studies of risk behaviors associated with HCV infection among blood donors in developed countries (Conry et al., 1996; Delage et al., 1999; Conte et al., 2000; Terrault, 2002). Similar to a study conducted in United States of America, about 40 percent of the HCV-positive donors had a history of injecting drug use (IDU) (Conry et al., 1996). According to the results of current study, out of total blood donors who were using injectable drugs, 42.1% were positive for anti-HCV status. The IDU was found to be significantly associated (p<0.05) with HCV infection and a contributing risk factor (OR=2.45, 95% CI=0.995-6.062).

The blood donors who shared glass for drinks were having more chances of getting HCV

infection. The reason could be the cracks/wounds present on lips or inside the mouth through which they might get infection from the infected glass.

## **Recommendations**

There should be highly sensitive screening test before the blood transfusion. Measures like surveillance for hepatitis, public awareness and health education of blood donors should be adopted by the government. Government should make policies to discourage professional blood donors from donating blood. Blood donors who are injectable drug users should not donate blood. Family persons should be preferred for blood donation.

#### *Conflict of interests*

The authors declare that they have no conflict of interest.

#### *Limitations of the study*

During questionnaire filling limitations of recall and recall bias cannot be ruled out.

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#### REFERENCES

- AJACIO, B.M. AND SANDRA, C.F., 2002. Risk factors for hepatitis C virus infection among blood donors in southern Brazil: a case-control study. BMC Gastroenterol., 2: 18.
- ASIF, N., KOKHAR, N. AND ILAHI, F., 2004. Seroprevalence of HBV, HCV and HIV infection among voluntary non remunerated and replacement donors in Northern Pakistan. *Pak. J. med. Sci.* 20: 24-8.
- BUSCH, M.P., GLYNN, S.A., STRAMER, S.L., STRONG, D.M., CAGLIOTI, S., WRIGHT, D.J., PAPPALARDO, B. AND KLEINMAN, S.H., 2005. A new strategy for estimating risks of transfusion-transmitted viral infections based on rates of detection of recently infected donors. *Transfusion*, **45**: 254-264.
- BUSCH, M.P., KLEINMAN, S.H. AND NEMO, G.J., 2003.

Current and emerging infectious risks of blood transfusions. J. Am. med. Assoc., **289**: 959–63.

- CONRY, C.C., VANRADEN, M. AND GIBBLE, J., 1996. Routes of infection, viremia, and liver disease in blood donors found to have hepatitis C virus infection. *N. Engl. J. Med.*, **334**: 1691–1696.
- CONTE, D., FRAQUELLI, M. AND PRATI, D., 2000. Prevalence and clinical course of chronic hepatitis C virus (HCV) infection and rate of HCV vertical transmission in a cohort of 15,250 pregnant women. *Hepatology*, **31**: 751-5.
- CRAWFORD, R.J., GILLON, J. AND YAP, P.L., 1994. Prevalence and epidemiological characteristics of hepatitis C in Scottish blood donors. *Transfus. Med.*, 4: 121–124.
- DELAGE, G., INFANTE, R.C. AND CHIAVETTA, J.A., 1999. Risk factors for acquisition of hepatitis C virus infection in blood donors. Results of a case-control study. *Gastroenterology*, **116**: 893-899.
- FAROOQI, J.I., FAROOQI, R.J. AND KHAN, N.M., 2007. Frequency of hepatitis B and C in selected groups of populations in NWFP, Pakistan. J. Postgrad. med. Ins., 21: 165-168.
- GAETA, G.B., STROFFOLINI, T., TALIANI, G., IPPOLITO, F.M., GIUSTI, G. AND DE-BAC, C., 1999. Surgical procedures as a major risk factor for chronic hepatitis C virus infection in Italy: evidence from a case- control study. *Int. J. Infect. Dis.*, **3**: 207–210.
- GOODRICK, M.J., GRAY, S.F., ROUSE, A.M., WATERS, A.J. AND ANDERSON, N.A., 1994. Hepatitis C (HCV)-positive blood donors in south west England: a case control study. *Transfus. Med.*, 4: 113–19.
- HLADIK, W., KATAAHA, P., MERMIN, J., PURDY, M., OTEKAT, G., LACKRITZ, E., ALTER, M.J. AND DOWNING, R., 2006. Prevalence and screening costs of hepatitis C virus among Ugandan blood donors. *Trop. Med. Int. Hlth.*, **11**: 951-954.
- IDREES, M., LAL, A., NASEEM, M. AND KHALID, M., 2008. High prevalence of hepatitis C virus infection in the largest province of Pakistan. J. Dig. Dis., 9: 96–104. doi: 10.1111/j.1751-2980.2008.00329.x.
- KATO, N., 2000. Genome of human hepatitis C virus (HCV), gene organization, sequence diversity, and variation. *Microb. Comp. Genomics*, 5: 129-51.
- LAKKANA, T., SATAWAT, T., NIWAT, M., DALE, N., DAVID, L.T. AND KENRAD, E.N., 2004. Risk factors for hepatitis C virus infection among blood donorsin northern Thailand. *Transfusion*, 44: 1433-1440.
- REBUZZINI, D.G., 2008. Study of the hepatitis C virus NS3 helicase domain for application in a chemiluminescent immunoassay. Ph.D. thesis, Department of Biotechnology and Bioscience, University of Milan Biococca, Italy. pp. 30.
- REHMAN, M.U., AKHTAR, G.N. AND LODHI, Y., 2002. Seroprevalence of hepatitis C antibodies in blood

donors. Pak. J. med. Sci., 18: 193-196.

- RYAN, K.J. AND RAY, C.G., 2004. Sherris Medical microbiology. McGraw Hill, New York, pp. 551-2.
- RYAS, M., HUSSAIN, T., BHATTI, F.A., AHMAD, F., TARIQ, W.U. AND KHATTACK, M.F., 2001. Epidemiology of hepatitis C virus infection in blood donors in Northern Pakistan. J. Rawalpindi med. Coll., 5: 56-59.
- TERRAULT, N., 2002. Sexual activity as a risk factor for hepatitis C. *Hepatology*, **36**: 99-105.
- WHO: Diseases; Hepatitis C. [http://www.who.int/csr/disease/ hepatitis/whocdscsrlyo2003/en/index3.html], [Accessed in 2011].
- WIN, N., FRAME, D., WATKINS, R. AND MITCHELL, R., 1994. The low risk of hepatitis C virus transmission among sexual partners of confirmed HCV-positive blood donors. *Transfus. Med.*, **4**: 243–244.

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