"Prevalence of HCV in apparently healthy voluntary blood donors"

Ahmad Naeem Sajed¹, Dr. Shaghufta Iram², Dr. Sajjad Haidar², Abid Sarwar³, Dr. Imran Ahmad⁴, Dr. Sadaf Imran⁴, Dr. Qumar Naiz⁴, Dr. Hamid Akbar⁴, Dr. Uzma Farid Durrani⁴, Dr. Muhammad Atif Imran⁵ and Mubashir Akbar¹

¹Emergency Services Academy Rescue 1122 Lahore-Pakistan ²Pathology Department Allama Iqbal Medical College Lahore-Pakistan ³Institute of Molecular Biology and Biotechnology the University of Lahore-Pakistan ⁴University of Veterinary & Animal Sciences Lahore-Pakistan ⁵Armed Forces Post-Graduate Medical Institute Rawalpindi-Pakistan

Absatrct: Hepatitis C is a silient murderer that may create no symptoms for decades. The hepatitis C virus (HCV) is a chief source of both acute and chronic hepatitis. The transmission of HCV is primarly all the way through exposure to contaminated blood. In this research aim was to find out the prevalence of HCV in apparently healthy voluntary blood donors by using immunochromatographic test and ELISA kits. One hundersed blood samples were screened at "The Medical Laboratories Lahore-Pakistan". Immunochromatographic test kits were used for qualitative detection of anti-HCV and ELISA for quantitative detection of anti-HCV. ELISA system was found to be quite sensitive in detecting anti-HCV as compared to ICT. On the basis of gender category the prevalence of HCV in males was 10% when screened through ICT while 13.7% were positive via ELISA. In females prevalence of HCV was 5% when samples performed through ICT when these samples were performed through ELISA they were 10% positive. The prevalence of HCV differs by gender which is higher among males than females. Notable increase in prevalence of HCV among donors of age (36-45) years was recorded. Risks for transmission include blood donation. So blood donors should be tested from athentic laboratory before donating the blood.

Keywords: Prevalence, Hepatitis C, Healthy blood donors

I. Introduction

The term "hepatitis" is used to describe a common form of liver injury. Hepatitis simply means inflammation of the liver¹. It may be Hepatitis A (infection hepatitis), Hepatitis B (serum hepatitis), Hepatitis C (non-A, non-B hepatitis), Hepatitis D (delta hepatitis), Hepatitis E, Hepatitis F and Hepatitis G.

Hepatitis C is a silent killer that may produce no symptoms for long time. Often the first sign of illness occurs when a person's liver stops working or they develop liver cancer. Some physicians have compared the experience to suddenly falling off a cliff². Hepatitis C is caused by the hepatitis C virus (HCV) which was identified in 1989³. Hepatitis C virus (HCV) is a major cause of both acute and chronic hepatitis⁴. Most patients infected with HCV have chronic liver disease which can progress to cirrhosis and hepatocellular carcinoma (HCC). Chronic infection with HCV is one of the most important causes of chronic liver disease. Chronic hepatitis C infection develops in approximately 75% of patients acutely infected⁵. The infection is often asymptomatic but chronic infection can lead to scarring of the liver and ultimately to cirrhosis which is generally apparent after many years³. Approximately 20% of chronic hepatitis C patients can be expected to develop cirrhosis, out of these 6 % will decompensate to end-stage liver disease (ESLD) and an additional 4% will develop hepatocellular carcinoma (HCC)⁶. Those who develop cirrhosis or liver cancer may require a liver transplant. Hepatitis C is the leading cause of liver transplantation though the virus usually recurs after transplantation⁷. Co-factors such as alcohol intake, obesity and underlying liver-related diseases (e.g. haemochromatosis) play a major role in the progression of the liver disease⁸.

In population with chronic hepatitis C, the immune system attacks infected liver cells and causes inflammation. Over the course of many years this inflammation damages the liver causing healthy tissue to be replaced with scar tissue⁹. Because liver damage gets worse over time and many people with chronic hepatitis C have been infected for decades, the number of people with liver failure is expected to more than double over the period from 2000 to 2030. The number of people who die from hepatitis C- related liver problems is expected to increase by 207 percent in this period of time. The rates of chronic HCV infection developing in patients with human immunodeficiency virus (HIV) infection and CD4 < 200 have been higher than in patients without HIV infection². Similar to acute hepatitis C infection, chronic hepatitis C may produce recognizable symptoms. A person could be infected for more than 20 or 30 years before signs of severe liver damage appear¹⁰.

Acute HCV infection produces a wide range of clinical presentations from asymptomatic to icteric illnesses similar to other forms of acute viral hepatitis. Flu like symptoms fever, jaundice, dark urine, fatigue, nausea, vomiting, loss of appetite and abdominal pain are commonly reported in symptomatic patients with acute HCV^{6} .

The transmission of HCV is primarily through exposure to infected blood. Risks for transmission include blood transfusion, intravenous drug use, high-risk sexual activity, solid organ transplantation from an infected donor, occupational exposure, hemodialysis, household exposure, birth to an infected mother and intranasal cocaine use¹¹. Since hepatitis C virus (HCV) was first identified, one of the best known and most extensively studied routes of HCV transmission has been blood or blood derivative transfusion. In the second half of the 20th century, HCV was transmitted widely through the use of parenteral injections, invasive medical and surgical procedures and transfusion of blood products¹².

Supplemental tests include a serologic anti-HCV assay and NATs for HCV RNA. In the United States, the only FDA-licensed supplemental anti-HCV test is the strip immunoblot assay. FDA-approved diagnostic NATs for qualitative detection of HCV RNA using reverse transcriptase polymerase chain reaction¹³. The advantage of immunochromatographic method is that it can be completed in 10-20 minutes and performed by nurses or technicians with a minimum of training¹⁴. Rapid tests are intended for qualitative detection wherever EIA methods are impractical. Enzyme-linked immunosorbent assay (ELISA) has become household names for medical laboratories, manufacturers of in vitro diagnostic products, regulatory bodies, and external quality assessment and proficiency-testing organizations. This brief historical note spotlights the development of enzyme labels in immunoassay from the invention of this method in the 1960s through its development and early use during the 1970s and 1980s. The first published EIA and ELISA systems differed in assay design but both techniques are based on the principle of immunoassay with an enzyme rather than radioactivity as the reporter label. Two scientific research groups independently and simultaneously developed this idea and executed the necessary experiments to demonstrate its feasibility. The ELISA technique was conceptualized and developed by Peter Perlmann¹⁶. ICT is a qualitative membrane based immunoassay for the detection of antibody to HCV in serum or plasma. The membrane is coated with recombinant HCV antigen on the test line region of the card. During testing the serum or plasma specimen reacts with the recombinant HCV antigen coated colloidal gold. The mixture migrates upward on the membrane chromatographically by capillary action to react with another recombinant HCV antigen on the membrane and generate a colored line. Presence of this colored line indicates a positive result while its absence indicates a negative result. To serve as a procedural control, a colored line will always appear at the control line region indicating that proper volume of specimen has been added and membrane wicking has occurred.

II. Material And Methods

This study was carried out in "The Medical Laboratories Lahore-Pakistan". Over the course of study, one hundred blood samples were collected from different government and private organizations of blood donation according to standard methodology. Donors with history of HCV infection, below 18 years and more than 45 years of age were excluded. Present study was conducted to evaluate the prevalence of HCV in apparently healthy voluntary blood donors.

Immunochromatographic test (ICT) was performed by kit method (Global, Cat. No. RT03231) and Enzyme-linked immunosorbent assay (ELISA) was performed by kit method (Biotech, Cat. No. E0510). Procedure performed was according to manufacture instructions.

Finally data was tabulated and analyzed using the Statistical Package for Social Sciences SPSS 17 software. Data were presented as frequencies. Evaluations were carried out at 95% confidence level and P < 0.05 was considered statistically significant.

III. Results And Discussion

It was found that out of 100 subjects screened including males and females, hepatitis C virus specific antibodies (Anti-HCV) were detected in 9 (9%) subjects using ICT kits and 100 subjects were screened by ELISA and Anti-HCV antibodies were detected in 13 (13%) subjects.

In our study majority of our blood donor population was constituted males 80% and females were only 20%. On the basis of gender, it was found that prevalence of HCV in males was 10%, i.e 08 were positive out of 80 via ICT while 13.7% i.e 11 cases were positive via ELISA. In females prevalence of HCV was 5% i.e 1 were positive out of 20 samples performed through ICT when these samples were performed through ELISA they were 10% i.e 2 were positive out of 20. According to these results it is clear that Enzyme-linked immunosorbent assay (ELISA) is more sensitive test than Immunochromatographic test (ICT). The prevalence of HCV is high in male population than female population.

Subjects			Anti-HCV Reactive cases		Anti-HCV Reactive cases	
Gender	Total No.		(ICT +)		(ELISA+)	
	No.	(%)	No.	(%)	No.	(%)
Males	80	(80)	08	(10)	11	(13.7)
Females	20	(20)	01	(05)	02	(10)
Grand Total	100	(100)	09	(09)	13	(13)

 Table 1: Gender wise distribution of HCV reactive subjects:

The prevalence of HCV differs by gender which is higher among males than females. It also increased significantly with age. On the basis of age, it was found that donors in group (18-25 years) 1 (5%) males were positive when diagnosed through "ICT" and but 2(10%) males were positive when samples proceeded through ELISA. The second group (26-35 years) 3 (6.5%) males and 1(11.1%) female were positive via ICT but 4 (8.69%) males and 1(11.1%) females cases were positive via ELISA. In third group (36-45 years), 4 (28.5%) were positive cases among males through ICT. When these samples were proceeded through "ELISA" the positive results were 5(35.7%) and 1(16.7) among males and females. These results have shown that the prevalence of HCV is higher in males than female healthy donors and it has increased with increase in age. There is significantly difference between ICT and ELISA. ELISA is more sensitive and reliable as diagnostic tool.

Age Group (Years)	Subjects Screened		Anti-HCV Reactive cases Positive (ICT+)		Anti-HCV Reactive cases Positive (ELISA+)	
	Males	Females	Males	Females	Males	Females
18-25	20	5	1 (5%)	0(00%)	2 (10%)	0(00%)
26-35	46	9	3 (6.5%)	1(11.1%)	4 (8.69%)	1(11.1%)
36-45	14	6	4 (28.5%)	0(00%)	5(35.7%)	1(16.7)
Total	80	20	8 (10%)	1(05%)	11 (13.75%)	2(10%)

Table 2: Distribution of Hepatitis C reactive cases in different age groups:

Hepatitis C viral infection is endemic not only in Pakistan but also all over the world. HCV infection has shown distribution, occurring among patients of all ages, gender, race, regions and affecting viral immune response. Hepatitis C virus (HCV) is a blood-borne virus that causes chronic hepatitis, cirrhosis, and hepatocellular carcinoma. The most common routes of transmission of HCV in developed countries include intravenous drug use, blood transfusions, haemodialysis, needle-stick injuries, tattooing, sexual intercourse and peri-natal infections. In hepatitis C six serotypes have been identified. Global studies have shown that serotype 3 is most easy to treat with a cure rate of around 80 percent. In Pakistan serotype 3 is most common

The prevalence of HCV was (13%) in apparently healthy voluntary blood donors and is comparable with other published research papers. Same kind of work was performed by Rehman *et al*¹⁷ in 2002 but individual percentages vary considerably. In this study 11% were confirmed to be anti- HCV positive among voluntary blood donors in Lahore. The same kind of study was also conducted by Sulma *et al*¹⁸ in 2009 who reported 14.5% positive for HCV.

High prevalence of HCV infection in male population was recorded i.e 13.7% in males and 10% in females. Because HCV was transmitted widely through transfusion of blood and percentage of male blood donors is greater than females. Secondly HCV is mainly associated with injecting drug use. Injecting drug user are commonly males.

Due to the high cost of treatment of hepatitis C virus infection and the unavailability of a vaccine against HCV, the main focus should be on preventive aspects. In Pakistan there is an urgent need to raise public awareness about hepatitis C which can be accomplished through programs in schools, colleges and universities and through information media. Availability of safe blood for transfusion is a must for the recipients and the community as well and can be achieved by vigorous screening of donors and donated bloods.

IV. Conclusion

Prevalence of HCV is high in healthy voluntary blood donor males and females whose ages are above 40 years of age. Blood donors must screen the blood from authentic pathology laboratory before donating the blood. ELISA is more sensitive and reliable as diagnostic tool than ICT.

Acknowledgements

I am very thankful to librarian Emergency Services Academy Rescue 1122 Lahore Pakistan.

References

- [1]. Ghany, M. G., Strader, D. B. and Thomas, D. L. Diagnosis, management and treatment of hepatitis C: an update. Hepatol, 49., 2009, 1335.
- [2]. Thomas, D. L. The natural history of hepatitis C virus infection: host, viral and environmental factors. J. Am. Med. Assoc., 284 (4), 2000, 450–456.
- [3]. Ryan, K. J. and Ray, C. G. Sherris Medical Microbiology. Mcgraw. Hill. Med., 4, 2004, 551–552.
- [4]. Frank. C., Mohamed, M. K., Strickland, G. T., Lavanchy, D., Arthur, R. R. and Magder, L. S. The role of parenteral antischistosomal therapy in the spread of hepatitis C virus in Egypt. Lancet., 355 (9207), 2000, 887-891.
- [5]. Holmberg, S. D. The increasing burden of mortality from viral hepatitis in the United States between 1999 and 2007. Ann. Intern. Med., 156 (4), 2012, 271-278.
- [6]. Thomas, D. L. and Seeff, L. B. Natural history of hepatitis C. Clin. Liver. Dis, 9, 2005, 383–398.
- [7]. Rosen, H. R. Clinical practice. Chronic hepatitis C infection. New. Eng. J. Med., 364 (25), 2011, 2429–2438.
- [8]. Negro, F. and Clement, S. Impact of obesity, steatosis and insulin resistance on progression and response to therapy of hepatitis C. J. Viral. Hepatol., 16 (10), 2009, 681-688.
- [9]. Elserag, H. B. and Rudolph, K. L. Hepatocellular carcinoma: epidemiology and molecular carcinogenesis. Gastroenterology, 132, 2007, 2557-2576.
- [10]. Davis, G. L., Albright, J. E., Cook, S. F. and Rosenberg, D. M. Projecting future complications of chronic hepatitis C in the United States. Liver. Transpl., 9 (4), 2003, 331-338.
- [11]. Alter, M. J. The epidemiology of acute and chronic hepatitis C. Clin. Liver. Dis., 1 (3), 1997, 559-568.
- [12]. Schreiber, G. B., Busch, M. P., Kleinman, S. H. and Korelitz, J. J. The risk of transfusion-transmitted viral infections. New. Engl. J. Med., 334, 1996, 1685-1690.
- [13]. Pawlotsky, J. M. Use and interpretation of virological tests for hepatitis C. Hepatol., 36, 2002, S65-S73.
- [14]. Sato, K., Ichiyama, S., Iinuma, Y., Nada, T., Shimokata, K. and Nakashima, N. Evaluation of immunochromatographic assay systems for rapid detection of hepatitis B surface antigen and antibody, Dainascreen HBsAg and Dainascreen Ausab. J. Clin. Microbial, 34, 1996, 1420-1422.
- [15]. Torlesse, H., Wurie, I. M. and M. H. The use of immunochromatography test cards in the diagnosis of hepatitis B surface antigen among pregnant women in West Africa. Br. J. Biomed. Sci., 54, 1997, 256-259.
- [16]. Rudolf, M. L. Enzyme Immunoassay (EIA) Enzyme- Linked Immunosorbent Assay (ELISA). Clin. Chem., 12, 2005, 2415-2418.
- [17]. Rehman, M. U., Akhtar, G. N. and Lodhi, y. Seroprevelance of hepatitis C antibodies in blood donors. Pak. J. Med. Sci., 18 (1), 2002, 18-25.
- [18]. Salma, G. N., Ghazal, Z., Zaheer, A. and Shamim, M. Frequency of Hepatitis C Virus Infection and Estimation of Serum Alanine Aminotransferase in HCV Positive Patients. J. I. M. D. C., 1211 (1), 2009, 1.