Prevalence and Risk Factors Associated with Hepatitis B and Hepatitis C and their Correlation with Inflammatory Markers among Southern Region of Punjab

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ABSTARCT: Hepatitis is becoming a major health problem worldwide. HBV is majorly transmitted by blood transfusion or by various body fluids like vaginal and semen fluids. HCV is also one of the major diseases that cause many prolonged effects like hepatocellular carcinoma (HCC) and liver cirrhosis. This observational study was conducted in Department of Pathology, Nishtar Medical College and Hospital, Multan as a part of internship. In this study many tests related to liver function, hematological parameters such as neutrophils, lymphocytes and Hepatitis B and Hepatitis C virus tests are included. Statistical analysis system was used to perform data analysis. The results were considered to be statistically significant when *p* values were < 0.05. In patients of HCB and HCV the severity of infection can be determined by inflammatory markers of liver such as ALT, AST in addition to other biomarkers. According to data analysis it was suggested that there is low prevalence of HBV and high prevalence of HCV in southern areas of Punjab. Therefore, more funding is required to design various projects for the treatment of chronic diseases like HBV and HCV with proper medication. These diseases can be prevented by avoiding the reuse of same razors as well as unprotected drug injections by cooperation of public healthcare workers and other respective teams that are linked with the control of infectious diseases.

Keywords: HCV, HBV, ALT, AST, Inflammatory Markers, Liver Cirrhosis.

INTRODUCTION

Hepatitis is one of the major global health problems. Every year it causes the death of about 1.5 million people. In both under developed and developed countries it has a negative impact on health by causing inflammation of liver Habibzadeh (2014). By origin hepatitis is of two types non-infectious and infectious but the most common among these two is the infectious form of the viral disease. Five different types of viruses A, B, C, D and E are the cause of viral hepatitis but among these five the chronic or acute form of disease is caused by the B and C virus. About 3% of overall world population is affected by hepatitis C but its prevalence in South America Asia and under developed nations of Africa is quite high and affects 10% of the population of these countries. Because of iatrogenic contamination 20% of the Egypt population is affected by hepatitis

C virus Rutherford *et al.*, (2016). The main reason of the mortality and morbidity all over the world is hepatitis B virus (HBV) infection. It was estimated by the world Health Organization (WHO) that HBV infect about 2000 million people worldwide and 350 million of these people are chronically affected Schilsky (2013). Cancer also spread by uncontrolled division of cells which ultimately affect the liver and causes liver cancer Naeem *et al.*, (2019).

Those people that are chronically infected, 65 million of them will die because of liver disease caused by their HBV infection Kane (1995). The major objective of this article is to provide an overview about the epidemiology and natural history of HBV infection with its prevention, diagnosis, care and treatment. When hepatitis enters into liver, the main target is liver cells but viruses also detected by PCR amplification method Dao *et al.*, (2019).

Blood transfusion is the most predominant cause of transmission of Hepatitis C virus. Worldwide HCV is one of those diseases that cause the long-term effects on health in proportion of cases; these health effects include HCV- related cirrhosis and hepatocellular carcinoma (HCC). Several non-invasive parameters such as aspartate aminotransferase (AST)/alanine aminotransferase (ALT) ratio (AAR), aspartate aminotransferase. We investigate whether these parameters are useful variables for determining HCV severity Grebely and Dore (2011).

Pakistan is a developing country that accommodates about 180 million people surviving with low health and educational standards. Human Development Index of the united state ranked Pakistan on 146th position out of 187 countries that carries one of the highest burdens of chronic hepatitis C virus and also have highest mortality rate because of the liver cirrhosis that leads to the cellular carcinoma. In Pakistan studies were conducted on different segments of population and their results show different level of prevalence in different risk groups. It was estimated that in Pakistan overall the country about 9 million people are the carrier of hepatitis B and over 14 million are the carriers of Hepatitis C virus. The prevalence of Hepatitis B Surface Antigen (HBs Ag) and antibodies to hepatitis C virus (anti-HCV) in young healthy Pakistani adults in recent studies carried out in different cross sections of population has ranged from 2.56–3.53% and 2.3–5.3% respectively 3, 4, 5 with segments of much higher prevalence. Although many of these studies were conducted on the healthy young people which were the participants of blood donation or pre-employment screening and this may not be a cross section of whole community Ali *et al*., (2009). When hepatitis enters into the blood also damaging the gastrointestinal tract and stomach upset occurs due to more accumulation of virus Bhardwaj *et al*., (2019)

Hepatitis B virus which is also known as "serum hepatitis" cause the infectious disease of liver called Hepatitis B and this disease is endemic only in China and also cause the epidemics in different parts of Asia and Africa Barker *et al*., (1996). There are about 350 million people that are chronic carriers of hepatitis and it was said that about one third of human population has been infected once in their life time with this virus Williams (2006). The major transmitting factors of this virus are contact with infected blood or different body fluids like semen and vaginal fluids. Viral DNA has been found in the urine, saliva and tears of the individuals that are chronic carrier of hepatitis virus. In under developed countries perinatal infection is the major route of hepatitis infection in humans Coopstead (2010). While working in a healthcare setting with low safety level, different fluids transfusions, tattooing, sharing tooth brushes or razors with infected person, dialysis, residence in an institution with poor hygiene conditions and travel in countries where hepatis is endemic are the major risk factors that cause the transmission and incidence of Hepatitis B infection

Sleisenger *et al*., (2006). Although HBV cannot be transmitted by sharing eating utensils, kissing, holding hands, coughing, breastfeeding or sneezing.

B virus infection continues to be a major public health problem worldwide and only in united annual infections occurring over the past 2 decades. However, because the majority of children and adults infected with hepatitis B virus do not develop clinical disease Bhattacharya *et al*., (2007).

The more detailed picture that describes the distribution of this infection is provided by the seroepidemiologic studies which are more accurate than the results of acute disease surveillance Behal *et al.,* (2008). For the evolution of highly effective vaccine of infants and older age adults that are at the high risk of disease different trends in hepatitis B infection plays a vital role.

Pakistan is one of those countries which have the highest fertility rate with more than four children per woman. It is slightly less than twice the size of state of California present in USA and by area it is about 800000 sq-km. Pakistan is larger in size as compared to Turkey or Chile. Pakistan is consisting of four provinces including Punjab, Baluchistan, Sindh and Northwest Frontier Province (NWFP). In addition of Federally Administered Tribal areas (FATA) and western third of Jammu and Kashmir, makes Pakistani population really large in size that's why here is more information about the risk factors and prevalence of hepatitis virus. Over the period of 13 years the medical and public health literature was studies on the prevalence of HBV and HCV in Pakistan that includes the analysis of data separately for the high risk population and general population in each of the four provinces. Also the published literature was reviewed that was related to the risk factor and major mode of transmission of HBV and HCV in Pakistan Perz *et al*., (2006).

In the current study, we have measured the seroprevalence of anti-HCV and anti-HBV antibodies to perform retrospective analysis of HCV infections in general population of major cities of the Punjab, Pakistan. Aim of this analysis was to identify socio-demographic groups with higher HCV prevalence and also HBV so that these groups could be further investigated for factors contributing to higher HCV infections and also HBV. Findings from the study will help in better management of hepatitis C majorly and B prevention and treatment strategies in the country.

MATERIALS AND METHODS

This observational study was conducted in the Department of Pathology, Nishtar Medical College and Hospital, Multan as a part of internship. A total of 200 patients included in this study in which males were 110 and 90 belonging to Southern Region of Punjab, Multan. This study consisted of age, address, occupation, education, marital and socioeconomic status.

Liver functions tests, Hepatitis B and Hepatitis C virus and other hematological parameters such as lymphocytes, neutrophils included in this study. Detection of hepatitis B surface antigen (HBs Ag) was considered the marker of chronic HBV and detection of hepatitis C virus antibody (HCVAb) was considered the marker of chronic HCV

The patients with age more than 10 years included in this study and relevant clinical and demographic data taken from this group in appropriate manner. The patients with age less than 10 years excluded from the study and relevant taken from these group.5ml sample of blood was taken from patients by using surgical syringe and mixed with EDTA as anticoagulant agent for clinical chemistry and serology testing and attending the Department of Pathology, Nishtar Medical College and Hospital, Multan. Blood was centrifuge by passing through centrifuge machine. We identified additional articles through searches of specific authors working in this field and through the cited references of relevant articles.

In clinical chemistry section majorly serology, samples were screened serologically for diagnosis of Hepatitis B and Hepatitis C virus by rapid kit under the examination of senior medical technician. A sample of 2-3 drops of serum putted on separate Hepatitis B and Hepatitis C devices. Clear dark line on the respective device indicated that positive Hepatitis B and Hepatitis C. HBV and HCV further testified by electrochemiluminescence method (ECLIA). Light was passed through specimen and color reaction occurred in ECLIA. Liver functions tests including aspartate aminotransferase, alanine aminotransferase, alkaline phosphatase, total bilirubinwere analyzed by Cobas analyzer-702 for the checking of working of enzymes in liver. Several other parameters hemoglobin, lymphocytes, nuetrophils were tested by advanced analyzer instrumentation Abdullah *et al.,* (2013). Data analysis was performed using the statistical analysis system. The results were considered to be statistically significant when *p* values were < 0.05.

RESULTS AND DISCUSSIONS

Various demographic characteristics, physical parameters, laboratory findings of Hepatitis C and were shown in the Table 1. The data included in this study was on the basis of gender, employment status, educational status, marital status, residency of selected individuals under observation. A total of 200 patients were included in this study in which male were 110 and female were 90. Out of 110 male patients, 70 patients were infected with hepatitis C and 40 were non-infected individuals. Out of 90 female patients, 40 patients were infected with hepatitis C and 50 were non-infected individuals. Included in this study in which male were 110 and female were 90. Out of these 200 patients taken 55 were unemployed out of which 35 were infected with hepatitis C virus and 20 were negative for hepatitis C virus, 105 were employed of which 45 were infected with hepatitis C virus and 60 were negative for hepatitis C virus rest of 40 patients were self-employed out of which 30 were positive for hepatitis C and 10 showed resistance to hepatitis C means their results were negative for HCV.

In our study of 200 individuals 100 were illiterate of which 90 were infected with hepatitis C virus and 20 were negative for hepatitis C virus which shows illiterate people are mostly susceptible to Hepatitis C, 70 individuals were undergoing high school education of which 5 were infected with hepatitis C virus and 65 were negative for hepatitis C virus whereas 20 individuals out of 200 were undergoing college level study out of which 10 were infected with hepatitis C virus and 10 were negative for hepatitis C virus and rest of 10 individuals were studying at university level and their results showed equal ratio of infected and uninfected 5.

Table 1: Demographic Characteristic of Hepatitis C among Population of Southern Punjab.

Variable value	No. of individuals Total taken	No. of HCV +ve individuals	No. of HCV –ve individuals
Gender			
Male	110	70	40
Female	90	40	50
Employment status			
Unemployed	55	35	20
Employed	105	45	60
Self employed	40	30	10
Education status			
Illiterate	100	90	10
High school	70	5	65
College study	20	10	10
University	10	5	5
Residency			
Rural	140	90	50
Urban	60	20	40
Marital status			
Single	40	10	30
Married	120	70	50
Other	40	30	10

After that we elaborated our study to rural and urban on basis of their residency. Out of 200 total individuals taken 160 were belonging to rural areas of which 90 were infected with hepatitis C virus and 50 were negative for HCV whereas out of 60 urban individuals 20 were positive for HCV and 40 were negative for HCV which showed that people belonging to rural areas were mostly positive for hepatitis C virus.

On further expanding over study to marital status we came to know that out of 200 individuals 40 were single of which 10 were positive for HCV and 30 were negative for hepatitis C virus whereas 120 were married out of which 70 were infected with hepatitis C virus and 50 were negative for HCV. Rest of 40 individuals was widow of which 30 were positive for HCV and 10 were not infected by hepatitis C virus. Overall on basis of marital status we come to know that married people are more susgceptible to hepatitis C virus as compared to single and widow people.

Several risk factors associated with spreading of hepatitis C included Injected drug use, Syringes, Unprotected sexual contact, Blood transfusion, Tatoo, Dialysis, Medication were shown in Table 2. Number of individuals infected with hepatitis C virus due to Injection of drug along with their respective percentage in this study were 60 (36.6%), Similarly other risk factors along with their respective infected individual percentages are as follows due to contaminated Syringes 10 (9.09%), Unprotected sexual contact 12 (10.9%), Blood transfusion 10 (9.09%), Tatoo 2 (1.8%), Dialysis 3 (2.7%), Medication 5 (4.5%).
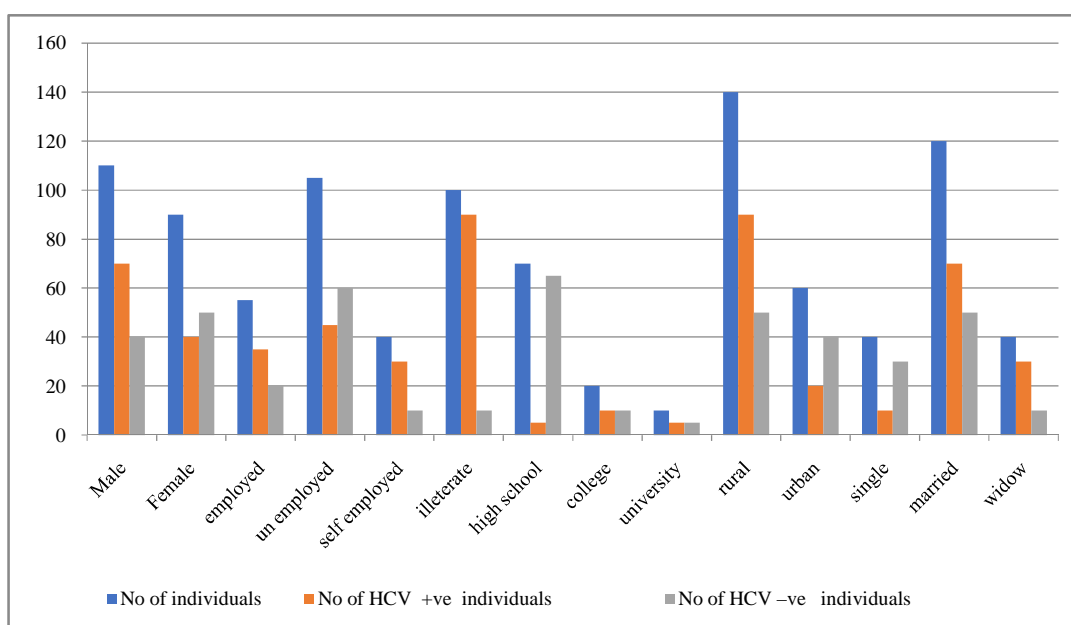

Fig. 1. Graph shows Sociodemographic Characteristic of Hepatitis C among Population of Southern Punjab.

Table 2: Risk Factors Associated with Hepatitis C Infected individuals among Population of Southern Punjab with respective to percentage.

Various risk factors	No. of individual infected	HCV infected percentage%
Injected drug use	60	36.6
Syringes	10	9.09
Razors	8	7.27
Unprotected sexual contact	12	10.9
Blood transfusion	10	9.09
Tattoo	2	1.8
Dialysis	3	2.7
Medication	5	4.5

Biochemical parameters such as aspartate aminotransferase, alanine aminotransferase (ALT), Alkaline Phosphatse, Total Bilirubin, WBC count, Hb (%), Neutrophils, Lymphocyte's, Platelets with statistical p-value shown in Table 3. p-value of AST, ALT, Total bilirubin, nuetrophils and platelets are significant while p-values ALP,WBC count, Hb and lymphocytes are non-significant.

Table 3: Clinical Parameters and their p-value among HCV infected Individuals of Southern Punjab.

Parameters	HCV Infected	p-value
ALT(SGPT)(U/L)	35.7 ± 3.4	0.01*
AST(SGOT)(U/L)	34.35 ± 5.6	0.01*
Alkaline Phosphatse	96.10 ± 55.5	0.07
Total Bilirubin	12.6 ± 5.5	0.01*
WBC count	3.85 ± 1.02	0.06
Hb (%)	10.2 ± 1.5	0.08
Neutrophils	32.5 ± 6.6	0.01*
Lymphocytes	22.6 ± 2.2	0.07
Platelets	130.2 ± 5.5	0.001*

Data are expressed as standard deviavtion mean $\pm$ SD. HCV: hepatitis C virus; AST: aspartate aminotransferase; ALT: alanine aminotransferase.
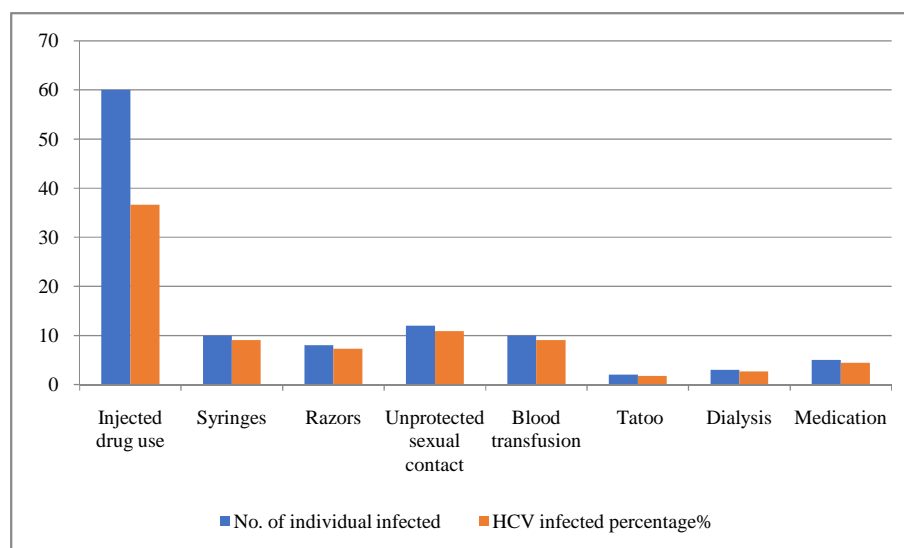

Fig. 2. Risk Factors Associated with Hepatitis C Infected individuals among Population of Southern Punjab with respective to percentage.

Various, physical parameters, demographic characteristics and laboratory findings of Hepatitis B were shown in the Table 4. The data included in this study was based on gender, employment status, educational status, marital status, residency of patients. A total of 200 patients were included in this study in which male were 110 and female were 90. Out of 110 male patients, 42 patients were infected with hepatitis B and 68 were non-infected individuals. Out of 90 female patients, 28 patients were infected with hepatitis B and 62 were non-infected individuals. Out of these 200 individuals taken 55 were unemployed of which 17 were infected with hepatitis B virus and 38 were negative for hepatitis B virus, 105 were employed of which 32 were infected with hepatitis B virus and 73 were negative for hepatitis B virus rest of 40 patients were self employed out of which 21 were positive for hepatitis B and 10 showed resistance to hepatitis B means their results were negative for HBV.

In our study of 200 individuals 100 were illiterate of which 38 were infected with hepatitis B virus and 62 were negative for hepatitis B virus. 70 individuals were undergoing high 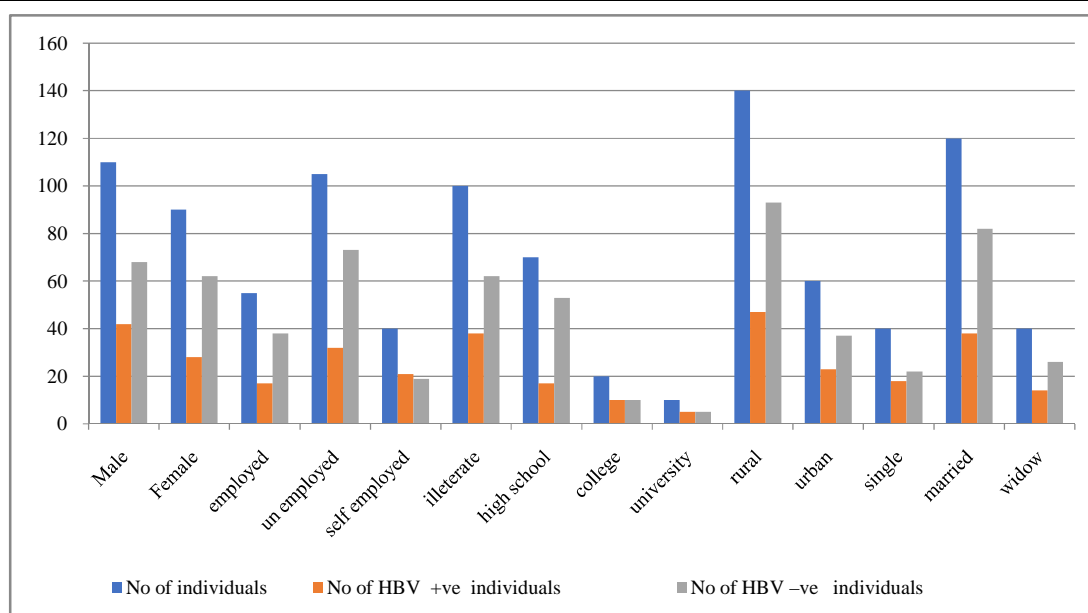school education of which 17 were infected with hepatitis B virus and 53 were negative for hepatitis B virus whereas 20 individuals out of 200 were undergoing college level study of which of which 10 were infected with hepatitis B virus and 10 were negative for hepatitis B virus and rest of 10 individuals were studying at university level and their results showed equal ratio of infected and uninfected 5.

After that we elaborated our study to rural and urban on basis of their residency. Out of 200 total individuals taken 160 were belonging to rural areas of which 47 were infected with hepatitis B virus and 93 were negative for HBV whereas out of 60 urban individuals 23 were positive for HBV and 37 were negative for HBV which showed that people belonging to rural and urban areas were mostly equally affected with HBV.

On further expanding over study to marital status we came to know that out of 200 individuals 40 were single of which 18 were positive for HBV and 22 were negative for hepatitis B virus whereas 120 were married out of which 38 were infected with hepatitis C virus and 82 were negative for HBV. Rest of 40 individuals was widow of which14 were positive for HBV and 26 were not infected by hepatitis B virus.

Table 4: Demographic Characteristic of Hepatitis B among Population of Southern Punjab.

Variable value	No. of individuals Total taken	No. of HBV +ve individuals	No of HBV –ve individuals
Gender			
Male	110	42	68
Female	90	28	62
Employment status			
Unemployed	55	17	38
Employed	105	32	73
Self employed	40	21	19
Education status			
Illitrate	100	38	62
High school	70	17	53
College study	20	10	10
University	10	5	5
Residency			
Rural	140	47	93
Urban	60	23	37
Martial status			
Single	40	18	22
Married	120	38	82
Other	40	14	26

Fig. 3. Graph shows Sociodemographic Characteristic of Hepatitis C among Population of Southern Punjab.

Table 5: Risk Factors Associated with Hepatitis C Infected individuals among Population of Southern Punjab with respective to percentage.

Various risk factors	No. of Individuals	HCV infected percentage%
Injected drug use	28	40
Syringes	11	15.7
Razors	8	11.4
Unprotected sexual contact	9	12.9
Blood transfusion	4	5.7
Tattoo	2	2.8
Dialysis	3	4.2
Medication	5	7.3

Several risk factors associated with spreading of hepatitis B included Injected drug use, Syringes, unprotected sexual contact, blood transfusion, Tattoo, Dialysis, Medication were shown in Table 5. Out of 70 patients infected with HBV from a population of 200 people 28 were due to Injected drug use and their percentage was 40, likewise effected individuals due to other risk factors with their respective percentage are as follows due to Syringes 11(15.7), Unprotected sexual contact 8(11.4%), Blood transfusion 9(12.9%), Tattoo 2(2.8%), Dialysis 3(4.2%), Medication 5(7.3%).
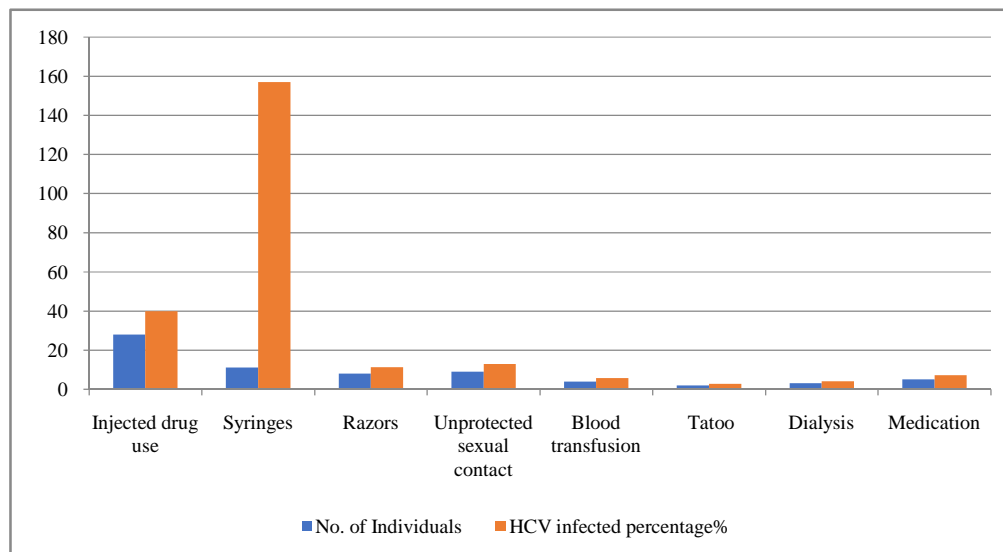
Fig. 4. Risk Factors Associated with Hepatitis C Infected individuals among Population of Southern Punjab with respective to percentage.

We observed different variables seroprevalence for hepatitis C virus and hepatitis B virus from different studies in similar populations within same province of Punjab among southern region. Our studies correlate with risk factors of transmission for HBV and HCV in Pakistan predominantly unsafe drug injections which ultimately leads to damage of liver cells due to more attack of for HBV and HCV Hutin *et al.,* (2003). There are 1.5 million units of blood and related products transfused each year in Pakistan. The blood transfusion is poorly weak network and significant factor in spreading of infectious disease like for HBV and HCV Rahman and Jawaid (2004).

Hepatitis C is progressed through attacking on liver cells and infection have reached the advanced stage. HCV is more likely to develop into liver cirrhosis as compared to hepatitis B virus infection Afdhal *et al.,* (2004). Our study similar to study by Baranova *et al*., (2011) AST, ALT and bilirubin level increased significantly as a liver inflammatory markers than those in chronic hepatitis c virus. These markers used for accessing the infection in liver cells.

HBV and HCV are the major infections that affected the liver and causes liver abnormality spreading throughout the worldwide. As a result of liver damage due to HBV and HCV, liver inflammatory markers such as Aspartate Aminotransferase (AST) and Alanine transaminase (ALT) increases severely in chronic liver disease. The prevalence of hepatitis C is high that correlates with our study indicated that major of the population affected with liver disease. (WHO, 2018).There is needed to design strategies for the prevention of HCV that is chronic form of hepatitis.

CONCLUSION

Certain liver inflammatory markers like AST, ALT along with other biomarkers indicated severity of severity of HCV and HBV infection in patients. Data suggested that there is high prevalence of HCV and low prevalence of HBV in southern region of Punjab. There is more need of funding for projects designing in associate with HBV especially HCV because as a chronic disease needed to treat with proper medications. Risk factors such as using of same razors and unprotected drug injections can be prevented by cooperation of public with health care workers and respective team associated with controlling of infectious diseases.

Conflict of Interest. There is no conflict of interest towards this research. All authors have contributed a significant role for publication of this research in writing of this research article.

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