



Prevalence of active HCV Infection and identification of its common Genotypes in District Mardan, Pakistan

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Abstract

The present study demonstrate the prevalence of active Hepatitis C viral infection and identification of its common genotypes circulating in district Mardan. A total of 582 patients were enrolled in this study in which 267 were male and 315 were female. Out of 582 samples, 329 were found HCV RNA positive through real-time PCR. The rates of active HCV infection were 59.9% in male while 53.6% in female. Age group above 60 were found with high rate of HCV infection, that is 80%. Out of 329 samples. Further a sum of 220 PCR based HCV RNA positive samples were screened for identification of HCV genotypes. Out of 220 positive samples Genotype 3a was found in 68.1%, 1a in 11.3%, 1b in 4.5%, Mixed-genotype in 4% and 3b in 2.7% while Untypable genotype in 9.5%. It was concluded from the present study that occurrence of active HCV infection rate were high in District Mardan and 3a was the most common genotype present in District Mardan.

Key words: GCMBDR, Mardan, Khyber Pakhtunkhwa.

Introduction

Hepatitis C infection is one of the major causes of liver diseases all over the world. HCV is the main cause of Hepatitis C infection and the majority of patients with chronic hepatitis is live untreated, which may cause hepatocellular carcinoma (HCC) and finally liver cirrhosis¹. Hepatitis C virus belongs to the family Flaviviridae and is an RNA virus^{2,3}. Hepatitis C virus is the principal source of chronic liver disease⁴ with estimated 170-200 million infected persons globally⁵ counting about 17 million in Pakistan⁶.

The main cause of HCV transmission in Pakistan is multiple use of syringes, minor and major surgery, dental procedures, blood and blood product transfusion, using of razors for multiple shaving or shaving in the shops of barbers, sharp instruments, nail cutters, tooth brushes and sexual transmission^{7,8}. Hepatitis C virus has major six genotypes and each genotype has several subtypes^{9,10}. The distributions of these genotypes are different globally and this difference is about one third of the variation in the genetic material from country to country. Genotypes 1, 2 and 3 are circulated worldwide. In Europe, Japan and USA, 1a and 1b are the most prevalent genotypes¹¹⁻¹³. HCV subtype 3a is the major genotype present in India¹⁴, Nepal¹⁵ and Pakistan⁸. The genotype 4 of Hepatitis C Virus is more prevalent in North

Africa and Middle East¹⁶, genotype 5 in South Africa and the genotype 6 appears to be most common in Hong Kong¹⁷. In Pakistan, the most prevalent HCV genotype is 3a, followed by 3b and 1a, with a strong association linking to chronic Hepatitis C viral infection and Hepato cellular carcinoma in Pakistan associated with genotype 3a¹⁸. The present study evaluated prevalence rate of active HCV infection and identification of its common genotypes present in general population of District Mardan.

Material and Methods

Study Design: This was lab based observational and descriptive cross-sectional study carried out from January 2011 to March 2011 at District Mardan.

Study Area and Studied population: The present study was carried out in district Mardan, Khyber Paktunkhwa. For the current study general population of district Mardan were selected. Patients from various areas of district Mardan were attended D.H.Q hospital and Mardan Medical Complex and was screened for HCV RNA through Enzyme linked immune Sorbent assay (ELISA) and ICT strip methods. A sum of 582 patients were found positive by ELISA and ICT strip method. Out of these 582 ELISA and ICT strip positive patients, 315

were female and 276 were male. Serological and biochemical data of these patients were recorded. For further confirmation of HCV RNA blood samples were taken from these patient, sera was separated and send to Genome Center for Molecular Based Diagnosis and Research Lahore (GCMBDR). GCMBDR is a state of the art molecular based diagnostic laboratory with responsive, precise and consistent detection tests on cost-to-cost basis utilizing PCR and real-time PCR procedures.

Data collection: Data was collected from Genome Center for Molecular Based Diagnostics and Research.

Statistical Analysis: All results were presented in percentage form and were analyzed through Statistix 9 version software. Chi square test were used for significance relationship amongst the categorical parameters. P-value <0.005 are considered as significant.

Results and Discussion

Gender wise and Age wise prevalence of HCV infection in District Mardan: In the current study a total of 582 patients were enrolled in which the rate of active HCV infection was 56.5% (n=329). Of these 582 samples 267 were male and 315 were female in which the rate of Hepatitis C virus infection

59.9% and 53.6% respectively which are shown in table 1. Active Hepatitis C viral infections in various age group were shown in figure 1. All the patients were categorized into three groups from 0-30 years, 31-60 years and above 60 years (table 1, figure 1).

Distribution of HCV Genotype in Both Gender of District Mardan: The table 2 show the rate of HCV genotypes and Subtypes in district Mardan. Out of 329 positive samples 220 were further studied for HCV genotypes and subtypes. Out of these 220 positive samples genotype 3a was found in 68.1% (n=149), 1a in 11.3% (n=25), 1b in 4.5% (n=10), Mixed 4% (n=09) and 3b in 2.7% (n=06) patients while 9.5% (n=21) were found with Untypable genotype. The rate of Hepatitis C virus Genotypes and Subtypes between male and female patients were shown in table 3 (table 2, table 3).

Discussion: HCV has come to the top regarding virus-induced liver diseases in several parts of the globe and have acquired endemic proportions in our population, but there is no national data variety system for evaluation of genotypes regarding HCV infection. The results on the geographic supply of genotypes regarding HCV are amongst the largest of their kind from Pakistan¹⁹.

Table-1
Gender wise Distribution of HCV in District Mardan

Gender	Total HCV Elisa and ICT strip Positive samples	HCV RNA Detected	Percentage	P Value
Male	267	160	59.9%	0.0006 significant
Female	315	169	53.6%	
Total	582	329	56.5%	

Table-2
Distribution of various HCV Genotypes in District Mardan

HCV Genotypes	Subtypes	No. of isolates	Percentage	P-Value
1	1a	25	11.3%	0.9998 Non-significant
	1b	10	4.5%	
3	3a	149	68.1%	
	3b	06	2.7%	
Untypable		21	9.5%	
Mixed		09	4%	
Total		220	100%	

Table-3
Gender wise distribution of HCV Genotypes and Subtypes in District Mardan

HCV Genotypes	Subtypes	Male	Female	Total	P-Value
1	1a	15(6.8%)	10(4.5%)	25(11.3%)	0.8722 Non-significant
	1b	8(3.6%)	2(0.9%)	10(4.5%)	
3	3a	63(28.6%)	86(39.5%)	149(68.1%)	
	3b	01(0.4%)	05(2.2%)	06(2.7%)	
Untypable		10(4.5%)	11(5%)	21(9.5%)	
Mixed		05(2.2%)	04(1.8%)	09(4%)	
Total		102(46.3%)	118(53.6%)	220	

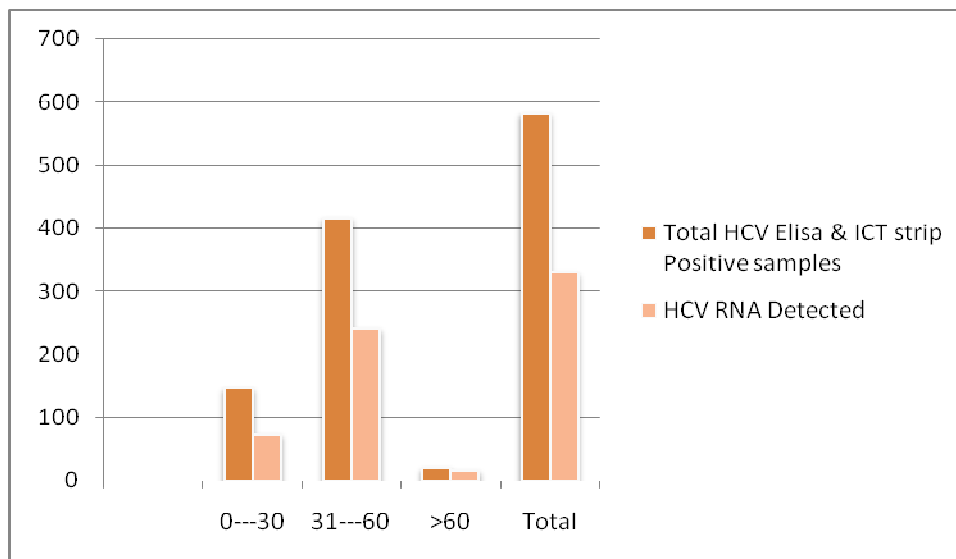


Figure-1
Age wise distribution of HCV in District Mardan

According to Attaullah *et al.*, and Sher Zaman Safi *et al.*, that genotype 3a, 3b, 1a, 1b and 2a were the common genotypes in Khyber Pakhtunkhwa while Genotype 4, 5, and 6 were absent²⁰ and were rare in Pakistan¹⁹.

The first finding of our research is to find out the rate of active Hepatitis C viral infection in male and female patients of District Mardan. Out of 582 samples 329 (56.45%) were found HCV RNA positive, in which the rate of HCV infection in male was 59.9% (n=160) and in female were 53.6% (n=169). There is no large difference of the rate of HCV infection between both genders. The HCV infection is more in male as compared to female. The increases of HCV in male were due to more interaction with population and surrounding environment. Our research with the agreement of the studies of Masroor *et al.*,²¹ and Ali *et al.*,²² in which the rate of HCV in male was 58.2% and female was 42.8%. While in a study conducted in Provence Punjab every one of the genotypes were distributed in the same frequency in male and females²³ which may be the geographical differences or differences in the mode of living in both of the provinces.

The second objective of research to finds out HCV in different age groups. Out 582 samples the rate of Hepatitis C viral infection in age range 0-30 years were 50% (n=146), 57.69% (n=416) were from age range 31-60 years and 80% (n=20) were from above 60 years. The results show that the HCV infections were high in all age groups but infection rate is significantly higher in the age group above 60 years. This may be due to the low immunity to resist to Hepatitis C viral infection. According to the study of Inamullah *et al.*,²⁴ highest prevalence of HCV was recorded in age group 20-30 and this may due to the geographical difference. While by another study, the HCV occurrence was higher in young individuals compared to older persons and seemed to be indicative connected with reduced

success rate beyond 40 years²⁵. Furthermore, the age group between 31–40 years showed that males of the group are at danger of having an HCV disease while females’ old group between 41–50 years displayed high HCV disease rate than males with not clear of the factors involving in this gender²⁵. In Provence, Punjab the prevalence in different age groups was different which was shown by as a study conducted in Provence Punjab in which essentially the most impacted age range regarding affected individuals has been 21-40 decades when compared having adolescent in addition to more mature age groups.

The third finding of our research to demonstrate the distribution of HCV genotypes in subtypes in the population of District Mardan. Out 220 positive samples the rate of 3a was 68.1% (n=149), 1a were 11.3% (n=25), Untypable were 9.5% (21), 1b were 4.5% (n=10), Mixed were 4% (n=09) and 3b were 2.7% (n=06). This study shows that the 3a are the most common in the HCV infected population of Mardan. Other studies also show that the 3a were more common in the population of Pakistan such as Inamullah *et al.*,²⁴ Ali *et al.*,²⁶ and Idrees and Riazuddin⁸. The present study was also correlated to another study which was conducted in district Mardan, according to which the genotype 3a was probably the most predominant genotype involving HCV RNA constructive samples that has a prevalence involving 90.3%²⁵. The current study also compared with other study conducted in Punjab; another Provence of Pakistan, in which the most prevalent genotype was also 3a (71%) was about similar to our study²³.

Conclusion

It is concluded from the present study that active Hepatitis C virus infection are high in District Mardan. Patients with age group above 60 are found with high rate of HCV infection.

Genotype 3a was found most common genotype in district Mardan. In females the Subgenotypes 3a were more common as compared to male. It is recommended to take further study, need of awareness and interferon therapy programs and HCV genotyping.

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