






Original Article

## Episode of Hepatitis C viral infection in the people of Swat, Pakistan

Episódio de infecção viral por Hepatite C no povo de Swat, Paquistão

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### ABSTRACT

Infectious agents cause serious diseases in humans worldwide and are responsible for the high rate of morbidity and mortality. The prevalence and epidemiology of infectious disease (HCV) in the hospital visited patients referred by the physicians through the initial findings and their associated risk factors were studied in Swat. The data of 174 infected patients were collected during the period of 2015 to 2017 from two clinical laboratories of Tehsil Matta Swat. Informed consent form was taken before blood collection. After taking informed consent blood samples were collected and ICT test was performed and then ICT positive cases were confirmed through PCR. A total of 174 ICT positive samples [106 male and 68 females] were included in this study. Age was considered from 10 to 72 years. Of the 174 ICT strip positive, 99 [63 males, 36 females] were confirmed through PCR. The prevalence rate was recorded 56.89%. I.V/I.M injection was recorded in 100% of the individuals. Visits to the barber shop was reported in (58%) of the individuals, married individuals were (81.0), surgical operation was reported in (44.8%), sharing toothbrush was observed in (29.9%), piercing was reported in (39.7%), family history was reported in (26.4%), dental treatment was observed in (21.8%), jaundice were (13.2%) and tattooing was (1.7%). Blood transfusion, surgical operations, Jaundice, family history and dental treatment were found significant risk factors for acquiring HCV infection. It was concluded that proper implementation of precautionary measures should be needed to control the spread of HCV in far near future.

**Keywords:** viral hepatitis C, Anti HCV Antibodies, risk factors, disease frequency, swat.

### RESUMO

Agentes infecciosos causam doenças graves em humanos em todo o mundo, e são responsáveis pelo alto índice de morbimortalidade. A prevalência e a epidemiologia das doenças infecciosas no hospital que atendeu pacientes encaminhados pelos médicos por meio dos achados iniciais e seus fatores de risco associados foram estudadas em Peshawar. Os dados de 174 pacientes infectados foram coletados durante o período de 2015 a 2017 oriundos de dois laboratórios clínicos de Tehsil Matta Swat. O formulário de consentimento informado foi obtido antes da coleta de sangue. Após a obtenção do consentimento informado, foram coletadas amostras de sangue e foi realizado o teste ICT e, em seguida, os casos ICT positivos foram confirmados por PCR. Um total de 174 amostras ICT positivas [106 homens e 68 mulheres] foi incluído neste estudo. A idade considerada foi de 10 a 72 anos. Das 174 tiras de ICT positivas, 99 casos [63 homens, 36 mulheres] foram confirmados por PCR. A taxa de prevalência foi de 56,89%. A injeção IV / IM foi registrada em 100% dos indivíduos. A visita à barbearia foi relatada em (58%) dos indivíduos, os números de casados foram (81,0%), e a operação cirúrgica foi relatada em (44,8%), o compartilhamento de escova de dente foi observado em (29,9%), o piercing foi relatado em (39,7%), antecedentes familiares foram relatados em (26,4%), tratamento odontológico em (21,8%), icterícia (13,2%) e tatuagem em (1,7%). Transfusão de sangue, operações cirúrgicas, icterícia, histórico familiar e tratamento odontológico foram fatores de risco significativos para adquirir infecção por Vírus da Hepatite C (VHC). Concluiu-se que a implementação adequada de medidas de precaução deve ser necessária para controlar a propagação do VHC em um futuro próximo.

**Palavras-chave:** hepatite C viral, anticorpos anti-VHC, fatores de risco, frequência da doença, swat.

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## 1. Introduction

Disease with hepatitis C virus (HCV) is a noteworthy worldwide wellbeing concern. It is expected that about 170 million individuals are contaminated with HCV around the world; this ailment is turned out to be a heightening financial, social and wellbeing trouble (Lavanchy, 2011). The weight of this sickness in a significant number of the developed and developing nations is consistently on the ascent (Hajarizadeh et al., 2013). Even though pervasiveness of HCV infection appears to have declined in the previous two decades in the vast majority of the developed nations of the world like, USA, Europe (Western and Northern), Australia and Japan (Razali et al., 2007). The occurrence of HCV differs by area. HCV occurrence less than 2% was noted in areas as America, Europe (Western) and Australia, while zones with the most astounding HCV infection are Africa and the Mediterranean (eastern) (Lavanchy, 2011). Hepatitis C Virus belongs to the family Flaviviridae with an around 9.6 kb, single-stranded RNA genome. Due to the poor constancy of HCV RNA dependent NS5B protein, the virus displays an abnormal state of arrangement heterogeneity (Moradpour et al., 2007). Considering grouping homology six noteworthy HCV genotypes (Duberg et al., 2008) and various distinctive subtypes e.g. 1b, 3a and so forth, have so far been recognized (WHO, 2003). Hepatitis C is the main source of end stage liver disease, including HCC (Hepatocellular carcinoma) and requirement for liver transplantation; recurrence of these was anticipated to build up 2-3 folds by 2030 (Dufour, 2004). Increased hazard of injection practices is the main source of HCV disease among drug abusers. There were almost 13.2 million drug abusers around the world (Aceijas et al., 2004) and half of them appear to have been infected with HCV (Aceijas and Rhodes, 2007).

HCV is commonly spread in Pakistan and its load is predicted to extend in upcoming decades due to extensive use of insecure medical practices. The occurrence of Hepatitis C virus infection in Pakistan had been evaluated on past and about 86 such studies are reported in the literature. Therefore, an improved efficient study was required to integrate the new information. A precise audit of information performed between 2010 and 2015 demonstrated that HCV sero-prevalence amongst the common adult Pakistani populace was 6.8%, while recent data on HCV established was around 6.0% of the population. In most surveys, it was indicated that HCV infection was high in rural and peri urban areas (up to 25%) (Umer and Iqbal, 2016). In Pakistan, there are different risk factors as compared to rest of the globe. HCV infection transmission is usually related to infected blood (blood products), drug addicts (intravenous), and needle pricking mostly accidentally or perinatal. Vertical and sexual routes are also reported as less frequent causes of HCV transmission (Dufour, 2004). Current research was aimed to identify different hazard factors among patients with HCV infection by analyzing their impact in our local community setup.

## 2. Materials and Methods

### 2.1. Data collection

Prevalence and clinical epidemiology of infectious agents related data were collected from hospitalized infected patients referred by the physicians through the initial findings, who have been visited to either of the two main clinical laboratories, Ahmad diagnostic center and Eilahi clinical laboratory opposite THQ hospital Mirkhel medical plaza Matta swat. The data were collected with the proper approval of the authorities from infected patients in the period of 2015 to 2017. The data of 174 infected patients were traced and recorded. The data according to the prevalence and epidemiology related information was recorded. Finally, calculate the infectious agents/ diseases frequency and percentage.

### 2.2. Sample screening

Individual's ages from 10 to 72 years was considered in the present study. 5 ml blood was collected with the help of sterile syringe. The isolated samples were transferred to the laboratory and processed to collect the serum and further to screen out the hepatitis C virus by screening kits (ICT: ACON, ACON Laboratories Inc., San Diego, CA 92121, USA) (Kalim et al., 2017; Rahat et al., 2018).

### 2.3. PCR confirmation

Total viral RNA was extracted using TRIzol reagent kit (Invitrogen Inc.) following the manufacturer's protocol. cDNA was obtained by reverse polymerases (MMLV-Moloney Murine Leukemia Virus) and the final product was used for PCR amplification. Two round PCR and two sets of primer (Table 1). Infection of HCV was detected on 1% agarose gel and the band were observed an under UV light (Figure 1).

### 2.4. Statistical Analysis

Statistical analysis was done using SPSSv23 (IBM SPSS Statistics Inc) to find out the percentage, frequency and correlation among the hepatitis C with risk factor.

## 3. Result

### 3.1. Prevalence of HCV

Males were (60.91%) and females were (39.1%) in the study population. ICT result shows 100% individuals are positive, but after by PCR conformation the result shows (59.43%) males and (52.94%) females were infected with HCV. Overall prevalence was recorded 56.89% (Table 2).

### 3.2. Age wise prevalence of HCV

10-72 year of individuals were considered and further divided into 6 age groups. A maximum number of the affected was in the age group of 41-50 consisted (28.16%) individuals followed by 31 to 41 age group, 25.28%, 51-60 (16.66%), 21 to 30 (14.94%), 61 and above (6.89%) and 10 to 20(4.59%) (Table 3).

**Table 1.** First & Second round primer used in the study.

Round	Primer	Forward/ Reverse
First Round	CCCTGTGNNNNNCTGTCTTCACGC	Forward direction
	ACTCGCAAGCNNNNNAGGCAGTAC	Reverse direction
Second Round	GAAAGCGTNNNNNTGGCG	Forward direction
	CACAAGGNNNNCGACC	Reverse direction

**Table 2.** Hepatitis positive patient among total screened samples.

Gender	Frequency	% of population	ICT + samples	% of ICT	PCR + samples	PCR %
Male	106	60.91	106	60.91	63	63.63
Female	68	39.1	68	68	36	36.36
Total	174	100.0	174	174	99	100

**Table 3.** Age wise prevalence of HCV.

Gender	10-20	21-30	31-40	41-50	51-60	61-above	Total
Male	5	19	28	23	19	12	106
Female	3	7	16	26	10	6	68
Percentage	4.59	14.94	25.28	28.16	16.66	10.34	100
Total	8	26	44	49	29	18	174

**Table 4.** Statistics of HCV risk factors and co-morbidities. n = 174.

Selected Perimeter of the study	Number of male & Female in the study population		Percentage of Male & Female in the study		Total individual	Total Percentage
	Male	Female	Male	Female		
Married	78	63	55.31	44.68	141	81.0
Family history	27	19	58.69	41.30	46	26.4
Visited to barbershop	101	1	99.1	0.98	102	58.6
Sharing tooth brush	28	24	53.84	46.15	52	29.9
Tattooing	3	0	100	0	3	1.7
I.V/I.M Inj	106	68	60.91	39.08	174	100.0
Blood transfusion	28	12	70	30	40	23.0
Dental Treatment	20	18	52.63	47.36	38	21.8
<b>Jaundice</b>	12	11	52.17	47.82	23	13.2
<b>Piercing</b>	2	67	2.89	97.10	69	39.7
<b>Surgical operation</b>	41	37	52.56	47.43	78	44.8

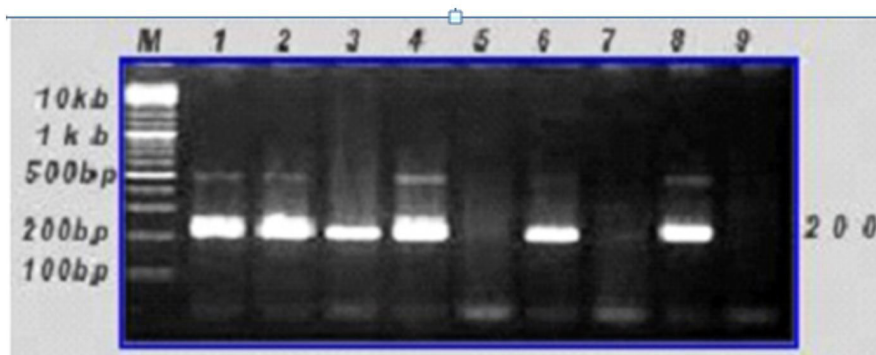
### 3.3. Demographic Variables

In the current study (81.0%) individuals were married. HCV positive family history was founded in (26.4%) followed by individual who visited to the barber shop were (58.6%), those who sharing tooth brush (29.9%), tattooing (1.7%), and I.V/I.M injection was (100%), Blood transfusion was recorded in (23.0%), dental treatment in (21.8%), jaundice

in (13.2%), piercing (39.7%) and surgical operation were reported in (44.8%) (Table 4).

### 3.4. Correlation between different risk factors in subjects with HCV

The typical thing in our research was that in the majority of patients there was more than one risk factor. Tattoo



**Figure 1.** Gel photograph of PCR product. 1% agarose gel was used. Lane 1 indicate 100bp ladder. Lane 5, 7 negative results while the remaining lanes indicate a positive result.

marks were less common in our locality, surgical and dental procedures, jaundice, family history were a special matter of concern to control. Pearson Correlation Sig. (2-tailed) of HCV was related to various risk factors which were statistically at significant levels. The correlations of various risk factors were shown in (Table 5).

#### 4. Discussion

Infections caused by the pathogenic agents cause serious diseases in humans and are responsible for the higher rate of morbidity/ mortality in human population worldwide. In the present study it is reported that 56.89% of individuals are reported positive for HCV infection. But previous studies show different results (Kalim et al., 2017) conducted a study on orphan individuals and reported 34.85% another study by (Ahmad et al., 2009) reported 13.8%. In the present study, none of the co- infection were reported both of hepatitis B and C virus. But previous studies show different results to our study (Baig et al., 2009) reported a higher frequency of co- infection working on cirrhosis patients in Pakistan. Sarwar and co-workers from different area of Abbottabad region reported 40% of co-infection in the study population (Sarwar et al., 2008). Higher Prevalence in our study may be due to the infected individuals who visited to doctor clinic. But previous data show general population. In the present study, we also found that the prevalence of HCV is male individuals more compared to female individuals. A similar study was reported by (Muhammad and Jan, 2005).

In the current study higher prevalence rate was found in the age group of 41-50. Similar report to our study shown by (Al-Hegami et al., 2015 Almezgagi et al., 2020). But the study of (Baghza, 2014) observed the high prevalence of HCV infection was (13%) among the age group more than 65 years.

In the present study (39.7%) subjects with a history of needle pricking by non-medical and medical professionals, accidentally or nose and ear piercing. Needle pricking ratio was very high in local setup. Previous studies conducted in Iran analyzed strong association of HCV seropositive with injections, tattooing, sharing equipment and history of imprisonment (Mir-Nasser et al., 2011; Nokhodian et al., 2012; Sharhani et al., 2017). Previous studies on needle pricking,

recapping needle pricking syringes were responsible for 2 to 10% transmission of hepatitis C in health care worker (Hamid et al., 1999; Mujeeb et al., 1998; Aziz et al., 2002) another study in Mirpur Khas, Hyderabad and Sukkur the incident of hepatitis C virus among the population sharing reused of injection equipment were reported 68%, 8.5% and 33.6% respectively (Altaf et al., 2009; Janjua et al., 2005). Incidence of hepatitis C virus in drug abusers was not reported in our study as no one was positive for HCV, but a previous study showed the incidence was 17.7% to 57% (Kuo et al., 2006; Achakzai et al., 2007; Platt et al., 2009).

In the current study (23%) subject was identified that had a history of blood transfusion. Compared to previous studies, its show different results,  $48.67\% \pm 1.75\%$  hepatitis C were reported in blood disorders, population (Hussain et al., 2008). A similar study was conducted by Luby et al. (2000) reported that 25% of blood blanks do not follow WHO recommended criteria during blood collection, but only 23% fulfill the recommend criteria. About 66% of the Pakistani population belongs to the rural community where blood transfusion facilities are insufficient. At the Pakistan round about 1.2 - 1.5 million transfusions are given each year as reported by W.H.O (Asif et al., 2004). Professional donors should be discouraged to promote the hazard-free blood supply (Luby et al., 2000).

In the current study (58.6%) male and female subjects had a history of barber history. Barbers in Pakistan and other developing nations usually do not have knowledge of health-related infections which they spread through reuse of unsterilized razors and scissors (Khaliq and Smego, 2008). Approximately 13% of barbers were aware that hepatitis could be transmitted by contaminated razors/scissors, etc. (Janjua and Nizamy, 2004). Studies show that most of the barbers are performing shavings in contaminated conditions and do not know about HCV infection (Wazir et al., 2008). Viral hepatitis C was present in 70.0% of facial and 48.0% of axillary shavings by barbers (Bari et al., 2001).

Major and minor surgical procedures were also analyzed in the current study. History of surgery subjects (44.8%), dental treatment were reported in 21.8%.

The prevalence of hepatitis c was significantly higher among patients with a history of blood transfusion. A significant relationship was found between the hepatitis

**Table 5.** Correlation between different risk factors in subjects with HCV. n = 174.

	Surgical Operation	Jaundice	Dental treatment	Blood transfusion	IV, IM Injection	drug Abuse	Tattooing	Sharing brush	visit to barber shop	Piercing Nose/ Ear	Family history	Married	HCV Patient
Surgical Operation	1	.228**	.391**	.222**	. <sup>a</sup>	. <sup>a</sup>	.058	.219**	-.158*	.191*	.272**	.289**	.271**
	Pearson Correlation												
	Sig. (2-tailed)	.002	.000	.003	.	.	.446	.004	.038	.012	.000	.000	.000
	N	174	174	174	174	174	174	174	174	174	174	174	174
Jaundice	.228**	1	.245**	.271**	. <sup>a</sup>	. <sup>a</sup>	.079	.042	-.051	.065	.074	.102	.305**
	Pearson Correlation												
	Sig. (2-tailed)	.002	.001	.000	.	.	.302	.584	.503	.393	.333	.179	.000
	N	174	174	174	174	174	174	174	174	174	174	174	174
Dental treatment	.391**	.245**	1	.240**	. <sup>a</sup>	. <sup>a</sup>	.037	.323**	-.121	.140	.156*	.256**	.235**
	Pearson Correlation												
	Sig. (2-tailed)	.000	.001	.001	.	.	.629	.000	.112	.065	.039	.001	.002
	N	174	174	174	174	174	174	174	174	174	174	174	174
Blood Transfusion	.222**	.271**	.240**	1	. <sup>a</sup>	. <sup>a</sup>	.033	.240**	.099	-.080	.044	.055	.200**
	Pearson Correlation												
	Sig. (2-tailed)	.003	.000	.001	.	.	.670	.001	.196	.295	.563	.469	.008
	N	174	174	174	174	174	174	174	174	174	174	174	174
IV/ IM Injection	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>
	Pearson Correlation												
	Sig. (2-tailed)	.	.	.	.	.	.	.	.	.	.	.	.
	N	174	174	174	174	174	174	174	174	174	174	174	174
drug Abuse	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>
	Pearson Correlation												
	Sig. (2-tailed)	.	.	.	.	.	.	.	.	.	.	.	.
	N	174	174	174	174	174	174	174	174	174	174	174	174
Tattooing	.058	.079	.037	.033	. <sup>a</sup>	. <sup>a</sup>	1	.010	.111	-.107	.021	-4.854E-2	.026
	Pearson Correlation												
	Sig. (2-tailed)	.446	.629	.670	.	.	.	.896	.144	.158	.786	.525	.732
	N	174	174	174	174	174	174	174	174	174	174	174	174

\*Correlation is significant at the 0.05 level (2-tailed); \*\*. Correlation is significant at the 0.01 level (2-tailed), a. Cannot be computed because at least one of the variables is constant.

Table 5. Continued...

		Surgical Operation	Jaundice	Dental treatment	Blood transfusion	IV, IM Injection	drug Abuse	Tattooing	Sharing brush	visit to barber shop	Piercing Nose / Ear	Family history	Married	HCV Patient
Sharing brush	Pearson Correlation	.219**	.042	.323**	.240**	. <sup>a</sup>	. <sup>a</sup>	.010	1	-.089	.087	.007	.124	.061
	Sig. (2-tailed)	.004	.584	.000	.001	.	.	.896		.244	.255	.925	.104	.422
visit to barber shop	N	174	174	174	174	174	174	174	174	174	174	174	174	174
	Pearson Correlation	-.158*	-5.109E-2	-1.208E-1	.099	. <sup>a</sup>	. <sup>a</sup>	.111	-8.878E-2	1	-.941**	-1.314E-1	-2.279E-1**	.070
	Sig. (2-tailed)	.038	.503	.112	.196	.	.	.144	.244		.000	.084	.002	.360
Piercing Nose / Ear	N	174	174	174	174	174	174	174	174	174	174	174	174	174
	Pearson Correlation	.191*	.065	.140	-.080	. <sup>a</sup>	. <sup>a</sup>	-.107	.087	-.941**	1	.073	.242**	-.030
	Sig. (2-tailed)	.012	.393	.065	.295	.	.	.158	.255	.000		.335	.001	.696
Family history	N	174	174	174	174	174	174	174	174	174	174	174	174	174
	Pearson Correlation	.272**	.074	.156*	.044	. <sup>a</sup>	. <sup>a</sup>	.021	.007	-.131	.073	1	.091	.338**
	Sig. (2-tailed)	.000	.333	.039	.563	.	.	.786	.925	.084	.335		.235	.000
Married	N	174	174	174	174	174	174	174	174	174	174	174	174	174
	Pearson Correlation	.289**	.102	.256**	.055	. <sup>a</sup>	. <sup>a</sup>	-.049	.124	-.228**	.242**	.091	1	-.066
	Sig. (2-tailed)	.000	.179	.001	.469	.	.	.525	.104	.002	.001	.235		.388
HCV Patient	N	174	174	174	174	174	174	174	174	174	174	174	174	174
	Pearson Correlation	.271**	.305**	.235**	.200**	. <sup>a</sup>	. <sup>a</sup>	.026	.061	.070	-.030	.338**	-6.584E-2	1
	Sig. (2-tailed)	.000	.000	.002	.008	.	.	.732	.422	.360	.696	.000	.388	
	N	174	174	174	174	174	174	174	174	174	174	174	174	174

\*Correlation is significant at the 0.05 level (2-tailed); \*\*. Correlation is significant at the 0.01 level (2-tailed), a. Cannot be computed because at least one of the variables is constant.

c virus with blood transfusion, surgical procedures, dentist visit, family history and jaundice.

## 5. Conclusion

In the present study it is concluded the Needle pricking, Jaundice, family history, blood products and dental procedures are common risk factors in our research.

## 6. Recommendations

It is recommended that there should be a national screening plan for hepatitis C. All subjects who are at risk must be investigated earlier. Screening program at national level should be started in early childhood. Early diagnosis and immediate treatment at standard levels (recommended by WHO/FDA and other international agencies) should be applied in large scale. Medical, surgical and dental instruments should be checked at international standards to avoid contamination. Health education may play vital role in the control of different preventable risk factors.

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