



Study of some biochemical factors among patients with Hepatitis C virus in Diyala Governorate

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ABSTRACT

Hepatitis C virus (HCV) infection acute or chronic, with ranging from a mild, self-limiting illness lasting to, lifelong condition. Chronic HCV infection is cause of liver cancer. This study was among renal dialysis patients, thalassemia patients, blood donors, and healthy control individuals in Diyala Province,. 360 participants, with a mean age of 30.0 ± 17.5 years (range: 3–69 years). The majority is (76.9% versus $\pm 23.1\%$). renal dialysis patients ($n = 90$), thalassemia patients ($n = 90$), blood donors ($n = 90$), and healthy control individuals ($n = 90$). &, the renal dialysis group included 60 males (66.7%) and 30 females (33.3%). The thalassemia group comprised 39 males (43.3%) and 51 females (56.7%). In the blood donor group, 89 participants were males (98.9%), while only one female (1.1%) was included. Similarly, the healthy control group consisted of 89 males (98.9%) and one female (1.1%). The presence of anti-HCV antibodies was detected using an enzyme-linked immunosorbent assay (ELISA). &, biochemical markers were measured in the serum samples. The results of this study was that 36 participants (10%) were positive for anti-HCV antibodies. The positivity rates were 32.2% among renal dialysis patients and 7.8% among thalassemia, ($P = 0.0001$, $p < 0.05$). &, all blood donors and healthy individuals tested negative for anti-HCV antibodies. Age-wise analysis demonstrated that the anti-HCV Ab positivity rate was higher in the 50–59-year age group compared with other age, ($P = 0.0001$, $p < 0.05$)., residence, and educational level, the results showed no differences in anti-HCV Ab positivity rates. the difference between males and females (8.7% vs. 14.5%) ($P = 0.123$, $p > 0.05$).&., showed that anti-HCV Ab–positive cases exhibited significantly higher .

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1. INTRODUCTION

Hepatitis is an inflammatory disease of the liver that may resolve spontaneously or progress to serious complications, including fibrosis, cirrhosis, and hepatocellular carcinoma [1]. Viral infections represent the most common cause of hepatitis; however, other factors such as alcohol consumption, certain medications, bacterial infections, and autoimmune disorders may also contribute to disease development. Viral hepatitis constitutes a systemic illness primarily affecting the liver and is caused by seven major viruses: hepatitis A virus (HAV), hepatitis B virus (HBV), hepatitis C virus (HCV), hepatitis D virus (HDV), hepatitis E virus (HEV), hepatitis G virus (HGV), and transfusion-transmitted virus (TTV) [2, 3]. Among these, HAV and HEV infections are typically self-limiting and transmitted via the fecal–oral route, whereas HBV, HCV, HDV, HGV, and TTV are blood-borne and may lead to chronic infections associated with long-term hepatic and extrahepatic complications [2, 3]. HCV, a member of the Flaviviridae family and genus Hepacivirus, is an enveloped, single-stranded RNA virus with a particle diameter of 45–65 nm [4, 5]. It is classified into seven confirmed genotypes and multiple subtypes, which play a crucial role in disease prognosis and the selection of antiviral therapy [6]. The pathogenesis of HCV-induced liver injury is predominantly immune-mediated rather than directly cytopathic, with hepatocytes as the primary target cells; persistent infection can lead to hepatocellular damage, portal and parenchymal inflammation, necrosis, and progressive fibrosis, which are key determinants of disease outcomes, including cirrhosis and hepatocellular carcinoma [7].

Chronic HCV infection may also result in extrahepatic manifestations, such as renal disorders, lymphoma, diabetes mellitus, and cryoglobulinemia[8]. Clinically, chronic hepatitis C is frequently asymptomatic for long periods, often delaying diagnosis until advanced liver damage occurs, whereas symptomatic cases may present with fatigue, anorexia, jaundice, dark urine, pruritus, ascites, peripheral edema, weight loss, or neurological manifestations related to hepatic encephalopathy [9, 10]. Acute HCV infection, representing the initial phase, is commonly asymptomatic, though symptomatic cases may appear within 1–3 months after exposure, presenting with jaundice, fatigue, nausea, fever, and myalgia [11]. Based on this background, the current study aims to determine the prevalence of anti-HCV antibodies among renal dialysis patients, thalassemia patients, blood donors, and healthy control individuals in Diyala Province, Iraq, to compare their distribution across these groups, assess potential associations with demographic and clinical factors, and provide baseline data to guide preventive measures and improve screening strategies in private laboratories and healthcare settings.

2. METHOD

2.1 Study groups

This study was conducted in Diyala Province, private laboratory from January to September 2025 and included a total of 360 participants. The mean age of the study population was 30.0 ± 17.5 years, with ages ranging from 3 to 69 years. Males constituted the majority of participants ($n = 277$, 76.9%), while females accounted for 83 participants (23.1%). The study population was divided into four equal groups. The renal dialysis group comprised 90 patients, including 60 males (66.7%) and 30 females (33.3%), with the majority aged 60 years and older. The thalassemia group included 90 patients, consisting of 39 males (43.3%) and 51 females (56.7%), most of whom were children and young adults, representing 89.9% of this group. The blood donor group consisted of 90 individuals, predominantly males ($n = 89$, 98.9%), with only one female participant (1.1%). Similarly, the healthy control group included 90 individuals, of whom 89 were males (98.9%) and one was female (1.1%), with approximately half of the participants aged between 30 and 39 years. To collect demographic and background information, a structured questionnaire was developed to obtain data on age, sex, residence, and educational level. Data were gathered through brief face-to-face interviews with all participants.

2.2 Blood samples collection

We collected 5 milliliters of venous blood from each participant using sterile plastic syringes after cleaning the area with 70% ethyl alcohol. The blood samples were quickly transferred to 10 milliliters plastic tubes right after collection. We let the tubes sit upright in a rack for about 30 minutes at room temperature to allow them to clot. After that, we moved the tubes to the lab in a cool box. Once there, we centrifuged the blood samples at 3000-4000 RPM for 5 minutes with a bench centrifuge to separate the serum. Using a 250 microliter automatic pipette with disposable tips, we aspirated the serum into new plastic tubes. The serum was then divided into 250 microliter aliquots in Eppendorf tubes, which were stored upright in a rack at -20°C until they were needed.

2.3 Detection of Ani-HCV Ab serological marker in serum by ELISA test

This test was performed using a commercially available kit (Dia.PRO, Italy HBs Ag ,ELISA). Reactive results were indicated by an absorbance reading of 1.1 and above, while non-reactive results were indicated by an absorbance reading less than 0.9. These values represent the kit-specific cut-off, as defined by the manufacturer, and are not universal standards.

2.4 Determination of biochemical markers in this study

The biochemical markers (Glucose test (75-115 mg/dL), Blood urea test (10-50 mg/dL), Creatinine test ($M=53-97$; $F=44-80$ Mmol/L), Total bilirubin test (<21 Mmol/L), Albumin test (38-51 gm/L), Total protein test (66-87 gm/L), ALT test (<40 IU/L) and AST test (<40 IU/L)) were determined regarding manufacturer's guidelines.

2.5 Statistical Analysis

The Statistical Packages of Social Sciences-SPSS (2019) program was used to detect the effect of difference groups in study parameters. T-test was used to significant compare between means. Chi-Square test was used to significant compare between percentages in this study [12].

3. RESULTS AND DISCUSSION

3.1 Study population

A total of 360 participants were enrolled in this study. The Mean \pm SD of age was 30.0 ± 17.5 years with a range (3-69) years. The majority are men, (76.9% versus $\pm 23.1\%$). Four groups were included in this study: 90 patients under renal dialysis were enrolled. The majority of them were male with 60 years and older. Again ninety patients with thalassemia were included in this study. Mostly are children and young adults (89.9%). Mostly were females (56.7%). The blood donors group consist of 90 individuals, most of them were males 98.9% and another 90 individuals were included as healthy control. Half of them were 30-39 years old. Mostly males (98.9%), table (1).

Table1. Distribution of study groups according to social factors.

Age (Ys)	Renal Dialysis		Thalassemia		Blood donors		Healthy control		P value
	No.	%	No.	%	No.	%	No.	%	
<10 years		-	65	72.2	-	-	-	-	0.0001*
10---19	5	5.6	15	16.7	4	4.4	4	4.4	
20---29	8	8.9	9	10.0	34	37.8	34	37.8	
30---39	9	10.0	-	-	45	50.0	45	50.0	
40---49	13	14.4	-	-	7	7.8	7	7.8	
50---59	18	20.0	-	-	-	-	-	-	
≥ 60 years	37	41.1	1	1.1	-	-	-	-	
Gender	Renal Dialysis		Thalassemia		Blood donors		Healthy control		P value
	No.	%	No.	%	No.	%	No.	%	
Male	60	66.7	39	43.3	89	98.9	89	98.9	0.0001*
Female	30	33.3	51	56.7	1	1.1	1	1.1	
Residence	Renal Dialysis		Thalassemia		Blood donors		Healthy control		P value
	No.	%	No.	%	No.	%	No.	%	
Urban	13	14.4	37	41.1	38	42.2	38	42.2	0.0001*
Rural	77	85.6	53	58.9	52	57.8	52	57.8	
Education	Renal Dialysis		Thalassemia		Blood donors		Healthy control		P value
	No.	%	No.	%	No.	%	No.	%	
Illiterate	20	22.2	46	51.1	-	-	-	-	0.0001*
Primary	33	36.7	34	37.8	13	14.4	13	14.4	
Intermediate	31	34.4	9	10.0	35	38.9	35	38.9	
Secondary	6	6.7	1	1.1	42	46.7	42	46.7	

*Significant difference between proportions using Pearson Chi-square test at 0.05 level.

3. 2. Serological marker:

The positivity rate of serological marker of HCV among study population was shown in table (2). 36 (10%) were positive for anti-HCV Abs.

Table 2. Positivity rate of HCV serological marker.

Marker	Status	No.	%
Anti-HCV Ab	Positive	36	10.0
	Negative	324	90.0

3.3. Biochemical markers:

The mean \pm SD and the range of biochemical markers among the participants were revealed in table (3). The mean \pm SD serum glucose level (mg/dl) was 102.336 ± 35.942 mg/dl with a range of (57-360) mg/dl. The mean \pm SD of blood urea levels 60.406 ± 53.411 mg/dl with a range of (15-318) mg/dl. The mean \pm SD of serum creatinine was 46.707 ± 31.293 mmol/L while the range was (1-102.45) mmol/L. The mean \pm SD serum total bilirubin was 22.116 ± 9.540 mmol/L with a range of (10.49-84.98) mmol/L. The mean \pm SD of total serum protein was 74.225 ± 8.697 gm/L with a range of (45-110) gm/L. Regarding the liver enzymes, ALT and AST levels, the mean \pm SD were 30.703 ± 17.596 IU/L and 31.071 ± 15.301 IU/L respectively, while the ranges were (3-121) IU/L and (9-143) IU/L.

Table 3. Mean \pm SD of biochemical markers among participants.

Biochemical Marker	Mean \pm SD	Range
Glucose (mg/dL)	102.336 ± 35.942	(57-360)
Blood urea (mg/dL)	60.406 ± 53.411	(15-318)
Creatinine (Mmol/L)	46.707 ± 31.293	(1-102.45)
Total bilirubin (Mmol/L)	22.116 ± 9.540	(10.49-84.98)
Albumin (g/L)	41.522 ± 5.866	(24-60)
Total protein (g/L)	74.225 ± 8.697	(45-110)
ALT (IU/L)	30.703 ± 17.596	(3-121)
AST (IU/L)	31.071 ± 15.301	(9-143)

3.4. Distribution of serological marker according to study groups:

Table (4) revealed that the anti-HCV Ab positivity rate among the renal dialysis and thalassemia groups were 32.2% and 7.8% respectively with a statistically high difference ($P= 0.0001$). However, all participants in the blood donors and healthy individuals were negative for anti-HCV Ab, so no statistical analysis can be applied.

Table (4): Distribution of study groups according to serological marker.

Education	Renal Dialysis		Thalassemia		Blood donors		Healthy control		P value
	No.	%	No.	%	No.	%	No.		
Anti-HCV Ab									
Positive	29	32.2	7	7.8	-	-	-	-	0.0001
Negative	61	67.8	83	92.2	90	100	90	100	*

*Significant difference between proportions using Pearson Chi-square test at 0.05 level.

3.5 Distribution of anti-HCV antibody positivity rate:

3.5.1. According to social factors:

Table (5) revealed the results of anti-HCV Ab distribution according to social factors. Regarding the age, the results showed that the anti-HCV Ab positivity rate was significantly higher among 50-59 years old compared to other age groups ($P= 0.0001$). The results of gender showed that there was insignificant difference between the anti-HCV Ab positivity rate among male and female (8.7% versus 14.5%) ($P= 0.123$). Similarly, the results found that the anti-HCV Ab positivity rate among urbans and rurals were 9.7% and 11.1%, thus the difference was statistically insignificant ($P= 0.338$). Additionally, the results also showed that the anti-HCV Ab positivity rate was significantly higher (16.1%) among those participants with primary education compared to other education categories ($P= 0.035$).

Table 5. Distribution of Anti-HCV Ab positivity rate according to social factors.

Variables	Anti-HCV antibody positivity rate				P value
	Yes		No		
	No.	%	No.	%	
Age					
<10 years	4	6.2	61	93.8	0.0001*
10---19	3	10.7	25	89.3	
20---29	7	8.2	78	91.8	
30---39	-	-	99	100.0	
40---49	4	14.8	23	85.2	
50---59	6	33.3	12	66.7	
≥ 60 years	12	31.6	26	68.4	
Gender					
Male	24	8.7	253	91.3	0.123
Female	12	14.5	71	85.5	
Residence					
Urban	10	7.9	116	92.1	0.338
Rural	26	11.1	208	88.9	
Level of education					
Illiterate	6	9.1	60	90.9	0.035*
Primary	15	16.1	78	83.9	
Intermediate	12	10.9	98	89.1	
Secondary	3	3.3	88	96.7	

*Significant difference between proportions using Pearson Chi-square test at 0.05 level.

3.5.2. According to study groups:

Results presented in table (6) illustrate the distribution of anti-HCV Ab according to the study groups. In renal dialysis there were 29 (32.2%) who were positive for anti_HCV Ab, while in thalassemia patients there were 7 (7.8%) who were positive for anti-HCV Ab. whereas, none of the r blood donors and healthy control were positive. Thus statistical analysis was inapplicable.

Table 6. Distribution of anti-HCV Ab positivity rate according to study groups.

Study groups	Anti-HCV antibody positivity rate				P value
	No		No		
	No.	%	No.	%	
Renal dialysis	29	32.2	61	67.8	0.0001*
Thalassemia	7	7.8	83	92.2	
Blood donors	-	-	90	100.0	
Controls	-	-	90	100.0	

*Significant difference between proportions using Pearson Chi-square test at 0.05 level.

3.5.3. According Biochemical markers:

Results shown in Table (7) revealed the distribution of anti-HCV Ab positivity according to biochemical markers (glucose, blood urea, creatinine, total bilirubin, albumin, total protein, ALT, and AST) was evaluated using the Pearson Chi-square test. The results were significantly higher among all biochemical markers in anti-HCV Ab-positive cases ($p = 0.0001$, $p < 0.05$)

Table 7. Distribution of anti-HCV Ab positivity rate according to biochemical markers.

Biochemical marker	Anti-HCV antibody positivity rate				P value
	Yes		No		
	No.	%	No.	%	
Total serum bilirubin (<21 Mmol/L)					
Low	-	-	-	-	0.0001*
Normal (<21)	3	1.3	234	98.7	
High (>21)	33	26.8	90	73.2	
Serum Albumin (38-51 g/L)					
Low (<38)	22	34.9	41	65.1	0.0001*
Normal (38-51)	13	4.5	273	95.5	
High (>51)	1	9.1	10	90.9	
Total serum protein (66-87 g/L)					
Low (<66)	15	37.5	25	62.5	0.0001*
Normal (66-87)	19	6.4	280	93.6	
High (>87)	2*	9.5	19	90.5	
Serum ALT (<40 IU/L)					
Low	-	-	-	-	0.0001*
Normal (<40)	6	2.0	298	98.0	
High (>40)	30	53.6	26	46.4	
Serum AST (<40IU/L)					
Low	-	-	-	-	0.0001*
Normal (<40)	10	3.2	304	96.8	
High (>40)	26	56.5	20	43.5	

*Significant difference between proportions using Pearson Chi-square test at 0.05 level.

The data we've gathered shows some notable differences in demographic and clinical traits among the various study groups. For instance, the renal dialysis patients tend to be older and mostly come from rural areas, whereas the thalassemia patients are primarily children. On the other hand, blood donors and healthy controls usually fall into the category of young adult males with higher education levels. It's interesting to see that all social factors displayed significant differences across the groups. Looking at the serological results, we found that 10% of participants tested positive for Anti-HCV antibodies. The biochemical markers varied quite a bit, reflecting the health differences among the participants. A significant gap in Anti-HCV positivity stood out between the groups, with renal dialysis patients showing the highest rate at 32.2%, followed by thalassemia patients at 7.8% that is agree with [13, 14]. that is show Anti-HCV positivity with renal dialysis patients. Blood donors and controls and agree with [12, 15] that show Haemodialysis (HD) requires blood exposure to infectious materials through the extracorporeal circulation for a prolonged period, and exposure to risk factors for nosocomial infections is always there.

In the renal dialysis units, hepatitis B (HBV) and hepatitis C (HCV) viral infections are significant causes of morbidity and mortality in haemodialysis (HD) patients, and management of such patients becomes complicated in lieu of these infections [16-18]. Most of the patients undergo dialysis for prolonged periods of time and are exposed to the various side effects occurring as a consequence of this procedure [19, 20]. The transmission of the virus to HD patients is generally nosocomial and potential risk factors include failure to disinfect devices between patients, sharing of single-use vials for infusion, improper aseptic techniques, contaminated dialysis equipment, and supplies and contamination by attending personnel. However, longstanding vascular exposure and manifold blood transfusions can also be major contributors [21, 22].

Also, HD patients are already immune-compromised due to irrevocable renal compromise, which contributes to infection by these viruses, however, all tested negative. When we checked the Anti-HCV positivity against social factors, age and education had significant connections, but gender and residence didn't seem to matter as much. The biochemical markers revealed that Anti-HCV positivity was closely linked to abnormal readings for bilirubin, albumin, total protein, ALT, and AST. Higher positivity rates were consistently seen in individuals with raised liver enzymes or abnormal liver-related blood values, underscoring the effect of HCV on liver function. All in all, these findings point to clear demographic differences among the study groups and emphasize the strong link between HCV positivity and both disease status and liver function indicators [23].

4. CONCLUSION

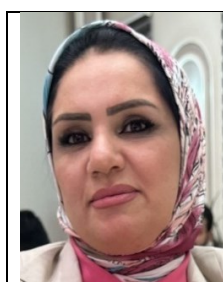
The current study revealed a relatively high prevalence of HCV infection among renal dialysis and thalassemia patients in Diyala Province, Iraq, no infection was found in blood donors neither healthy persons. The results also suggest that HCV infection is characterized by dramatic changes in the levels of biochemical markers as a measurable consequence on liver functioning progression, thus requiring active surveillance and preventive intervention among some risk groups.

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