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VARIATIONS IN LIVER BIOCHEMICAL TESTS IN PATIENTS WITH CHRONIC HCV AND HEPATITIS B IN LIBYA

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ABSTRACT

Chronic hepatitis is not a single disease, but rather a complex clinico-pathological syndrome with multiple causes, varying stages of necro-inflammatory and sclerosing liver damage, different prognoses and responses to treatment. This research was a comparative case control study.

It implicated about 120 blood samples from patients with chronic viral hepatitis, where 60 with HCV and 60 with HBV infection. The results of this study showed a highly significant difference regarding liver biochemical tests. The mean concentration of Total Bilirubin, Direct Bilirubin, Indirect Bilirubin and ALT enzyme in HCV and HBV patients (0.74 ±0.054, 0.26±0.24,0.48 ±0.033 mg/L and 23.45±1.18 u/L), (0.81±0.049, 0.30±0.024, 0.51±0.029 mg/dl and 26.20±1.38 u/L), (0.55 ± 0.039, 0.18 ± 0.17, 0.37 ± 0.026 mg/dl and 15.80 ± 1.54 u/L) when compared with the control group with *P* values (0.030, 0.036, 0.049 and 0.000), respectively.

KEYWORDS: Viral hepatitis, liver biochemical test, Libyan's patients.

INTRODUCTION

Hepatitis C is global ailment. The most affected regions are Central and East Asia, and North Africa.[1] There are multiple strains of the HCV virus and their distribution varies from region to region. African countries have the highest prevalence rates of HCV in the world, ranging from 1 to 26%.[1,2] Hepatitis B is the most prevalent in Sub-Saharan Africa and East Asia. Most people in these regions become infected with the virus during childhood and 5 – 10% of the adult population are chronically infected. High rates of chronic infection are also found in the Amazon and the southern parts of eastern and central Europe.[3,4] In Libya, comprehensive study was carried out on HCV genotypes. Hepatitis C virus genotype 4 was the most predominant, followed by HCV genotype 1 and then other less common genotypes.[3, 4] However, further studies are needed to clarify the magnitude and impact of HCV in Libya. The estimated number of chronic HBsAg carriers in Libya is 120 – 150 thousand individuals with a concentration of the disease in certain populations with high-risk behaviours associated with viral transmission.[5] The prevalence of HBsAg among the general population in Libya was found to be 2.2%. Liver diseases affect the normal functions of the liver. Abnormalities in the liver functions, however, are usually not apparent in most individuals with chronic liver disease until the disease is rather advanced.[6,7] The diseases that occur in the liver are infectious including viral hepatitis. The hepatitis is inflammation of the liver. The term is generally used to refer to the diseases caused by the hepatropic viruses including the diseases hepatitis A–E, and disease due to cytomegalovirus, Epstein-Barr virus, adenovirus, rarely herpes simplex virus and others.[6] Among the hepatitis viruses, only hepatitis B virus and hepatitis C virus are able to persist in the host and cause chronic hepatitis.[8] Descriptions are largely based on findings in transfused patients where the time of infection is certain. Clinical presentation of disease after other modes of transmission, such as intravenous drug addiction. The incubation period is about 7-8 weeks.[9] The symptoms resemble those of other forms of viral hepatitis. Serum Hepatitis C virus ribonucleic acid (RNA) becomes positive 1-2 weeks after infection. At 7-8 weeks, serum alanine and aminotransferase is moderately increased to about 15 times the upper limit of normal. Liver function tests (LFT) are a helpful screening tool, which are an effective modality to detect hepatic dysfunction. Since the liver performs a variety of functions, no single test is sufficient to provide complete estimations of liver function. Often clinicians are faced with reports that do not tally with the clinical condition of the patient and they face difficulty in interpreting the LFT.[9] In spite of receiving a lot of criticism for this terminology, the

phrase 'liver function tests' is firmly entrenched in the medical lexicon. It might be argued that 'liver injury tests' would be a more appropriate terminology. Moreover, the clinical history and physical examination play important role to interpret the functions. The liver contains thousands of enzymes that have no particular function and behave as serum proteins. Aminotransferases (formerly transaminases) are the most frequently utilized and specific indicators of hepatocellular necrosis. Enzymes aspartate aminotransferase (AST, formerly serum glutamate oxaloacetic transaminase-SGOT) and alanine Aminotransferase (ALT, formerly serum glutamic pyruvate transaminase-SGPT) catalyze the transfer of aspartate and alanine amino acids, respectively, to the α-keto group of ketoglutaric acid. ALT is primarily localized to the liver but the AST is present in a wide range of tissues like the heart, skeletal muscle, kidney, and brain. Meanwhile, AST is present in both the mitochondria and cytosol of hepatocytes. The cytosolic and mitochondrial forms of AST are true isoenzymes and are immunologically distinct. About 80% of AST activity in human liver is contributed by the mitochondrial isoenzyme, while most of the circulating AST activity in normal people is derived from the cytosolic isoenzyme. Large increases in mitochondrial AST occur in serum after extensive tissue necrosis. Therefore, assay of mitochondrial AST has been advocated in myocardial infarction. Mitochondrial AST also increases in chronic liver disease. Their activity in serum at any moment reflects the relative rate at which they enter and leave circulation. There are abundant methods for this measurement, the most specific links the formation of pyruvate and oxaloacetate, products of the aminotransferase reactions, to their enzymatic reduction to lactate and malate. Virtually, no aminotransferases are present in urine or bile and hepatic sinusoids are the primary site for their clearance. Chronic HCV infection develops in 75-85% of persons, with persistent or fluctuating ALT elevations indicating active liver disease developing in 60-70% of chronically infected persons. No clinical or epidemiologic features among patients with acute infection have been found to be predictive of either persistent infection or chronic liver disease.[10] A recent study in Iraq has been done by Hussein RH, (2011), found that Liver function tests showed no significant difference between HBV and HCV patients, however, a significant difference regarding liver function tests was found among both sets of patients and the control group.[11] Rehman AU, *et al*. 2016, in Nigeria accomplished a study with a convenient sample of 156 HCV positive patients. To collect blood CP and ALT, a reduction of level data and other important information were collected from the patients at regular intervals. Finding was a significant reduction in ALT levels due to Pegylated interferon plus ribavirin therapy.[12] There was a significant reduction in ALT levels due to Pegylated interferon plus ribavirin therapy.[11,12] Therefore, the aim of this study is to assess the liver biochemical tests obtained infected patients with (HCV) and (HBV) in western region of Libya.

METHODS AND MATERIALS

Study Population

One hundred and twenty samples were involved in this study. There were 60 from HBV and/or HCV infected patients as well as 20 control samples from healthy people. Samples were collect from those infected with HCV and/or HBV as well as healthy individuals (as a control group) in the western region of Libya. We collected all samples from the Department of Communicable Diseases at Tripoli Central Hospital, and all tests were conducted at Sabratha Teaching Hospital, as described in our previse paper.[13]

Sample Collection and Liver Biochemical Tests

Three milliliters of venous blood were collected from each participant in plain tube. After coagulation, samples were centrifuged at 3500 RPM for 5 minute. Serum was then collected. For haematological parameters, 2 ml venous blood were collected in EDTA tubes from each participant. HBsAg and anti-HCV antibodies were detected by using commercially available enzyme linked immune-sorbent assay (ELISA) according to the manufacture instruction. After serum separation from each blood sample as described above the following liver biochemical tests were analysed by using ARCHITECT PLUS (C 4000):

1. Total Bilirubin Diazonium Salt

The total bilirubin assay is used for the quantitative analysis of total bilirubin in human serum of adults and neonates on the ARCHITECT c 4000 System. This method is based on the reaction of bilirubin with a diazo reaction which can be accelerated by the addition of various chemicals. Total (conjugated and unconjugated) bilirubin couples with a diazo reaction is accelerated by the addition of surfactant as a solubilizing agent. The increase in absorbance at 548 nm due to azobilirubin is directly proportional to the total bilirubin concentration.

2. Direct Bilirubin

The direct bilirubin assay is used for the quantitative analysis of direct bilirubin in human serum. Determination is generally based on the reaction of bilirubin with a diazotized sulfanilic acid, described by Ehrlich. In this method, direct (conjugated fractions) bilirubin couples with diazonium salt in the presence of sulfamic acid to form the colored compound azobilirubin. The increase in absorbance at 548nm due to azobilirubin is proportional to the direct bilirubin concentration.

3. Alanine Aminotransferase

NADH (p-5-p), the Alanine aminotransferase (ALT) assay, is used for the quantification of alanine aminotransferase in human serum. ALT present in the sample catalyzes the transfer of the amino group from L-alanine to α ketoglutarate, forming pyruvate in the

presence of NADH and lactate dehydrogenase (LD) is reduced to L-lactate. In this reaction NADH is oxidized to NAD.

4. Aspartate Aminotransferase (AST)

NADH (without p-5-p). The aspartate aminotransferase (AST) assay is used for the quantification of aspartate aminotransferase in human serum. AST present in the sample catalyzes the transfer of the amino group from L-aspartate to α ketoglutarate, forming oxaloacetate in the presence of NADH and malate dehydrogenase (MDH) is reduced to L-malate. In this reaction, NADH is oxidized to NAD.

5. Alkaline Phosphatase

Para-nitrophenyl phosphate for additional information on system and assay technology, refer to the ARCHITECT system operations manual. Alkaline phosphatase in the sample catalyzes the hydrolysis of colourless p-nitrophenyl phosphate (p-NPP) to givep-nitrophenol and inorganic phosphate. At the PH of assay (alkaline), the p-nitrophenol is in the yellow phenoxide form the rate of absorbance increase at 404 nm is directly proportion to the alkaline phosphatase activity in the sample.

Data Analysis

All the data of this study were analyzed using the application of statistical package of social science (SPSS) V. 25, biochemical parameters tested by Pearson's correlation test. Comparison of mean value of continuous data was tested by t-test and ANOVA test. Chi-square statistical analysis were done to detect significant value. *P* value of <0.05 and *P* value < 0.01were used to establish statistical significance.

RESULTS

Total of 120 blood samples from patients with chronic viral hepatitis, 60 of them with HBV infection, 60 with HCV infection and 20 apparently healthy age and gender matched subjects were included as a control group. Out of the 60 patients with HBV, 43 (36%) were males and 17 (14%) were females. Whereas, 38 (32%) patients with HCV were males and 22 (18%) were females as described in our previse paper [12]. Result of biochemical parameters of male patients infected with HCV and HBV showed that ALT (U/L) were 16.80 ± 2.39, 23.58 ± 1.49, and 26.53 ± 1.56 among control group, HCV and HBV patients respectively. Whereas result of AST (U/L) were 25.30±1.68, 25.50 ± 1.22, and 25.37 ± 1.10, among control group, HCV and HBV patients respectively. ALP (U/L) were 76.90 ± 3.93, 80.95 ± 3.91, and 76.44 ± 2.69, among control group, HCV and HBV patients respectively. Direct bilirubin results (mg/L) were 0.19 ± 0.031, 0.27 ± 0.03, and 0.33±0.03 among control group, HCV and HBV patients respectively. While indirect bilirubin results (mg/L) were 0.36 ± 0.04, 0.51 ± 0.04, and 0.56 ± 0.03 among control group, HCV and HBV patients respectively. Total bilirubin results (mg/L) were 0.55 ± 0.06, 0.78 ± 0.07, and 0.88 ± 0.06 among control group, HCV and HBV patients respectively. Regarding results of females biochemical parameters of control group, patients infected with HCV and HBV showed that, ALT (U/L) was 14.80 ± 2.00, 23.23 ± 1.99, and 25.35 ± 2.94 respectively. At the same time results of AST (U/L) were 22.00 ± 2.06, 25.91±2.33, and 26.59 ± 2.29 respectively. From the other hand results of ALP (U/L) was 83.80 ± 7.52, 89.23 ± 5.10, 74.88 ± 6.16 respectively. Direct bilirubin results were 0.17 ± 0.015, 0.25 ± 0.040, and 0.22 ± 0.035 respectively. Indirect bilirubin results were 0.37 ± 0.037, 0.43 ± 0.051, and 0.39 ± 0.051 respectively. Total bilirubin results were 0.54 ± 0.048, 0.68 ± 0.086, and 0.61 ± 0.083 respectively.

Table 1- Biochemical parameters of male and female patients infected with HCV.

Groups / Parameters	Males Patients	Females Patients
	Mean ± SE	**Mean ± SE**
ALT	23.58 ± 1.49	23.23 ± 1.99
AST	25.5 ± 1.222	25.91 ± 2.33
ALP	80.95 ± 3.91	89.23 ± 5.10
Direct Bil	0.27 ± 0.03	0.25 ± 0.04
Indirect Bil	0.51 ± 0.04	0.43 ± 0.05
Total Bil	0.78 ± 0.07	0.677 ± 0.09

(*) significant difference compared to male patients infected with HCV group (P < 0.05).

(**) highly significant difference compared to male patients infected with HCV group (P < 0.01).

Table 2 - Biochemical parameters of male and female patients infected with HBV.

Groups / Parameters	Males Patients	Females Patients
	Mean ± SE	**Mean ± SE**
ALT	26.53 ± 1.561	25.35 ± 2.943
AST	25.37 ± 1.104	26.59 ± 2.29
ALP	76.44 ± 2.692	74.88 ± 6.16
Direct Bil	0.326 ± 0.0289	0.224 ± 0.0349*
Indirect Bil	0.558 ± 0.0318	0.388 ± 0.0514**
Total Bil	0.884 ± 0.0568	0.612 ± 0.0831*

(*) significant difference compared to male patients infected with HBV group (P < 0.05).

(**) highly significant difference compared to male patients infected with HBV group (P < 0.01).

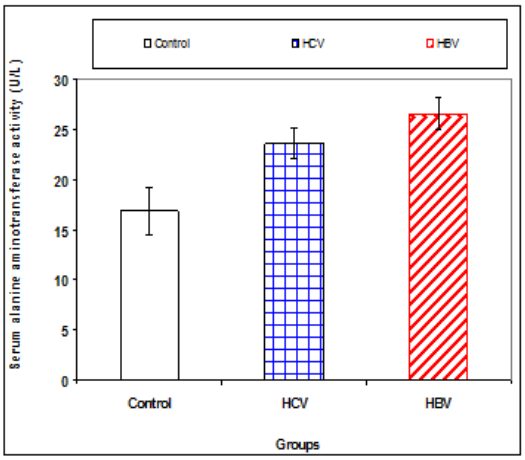

Figure 1 - Serum alanine aminotransferase activity of male patients infected with HCV and HBV.

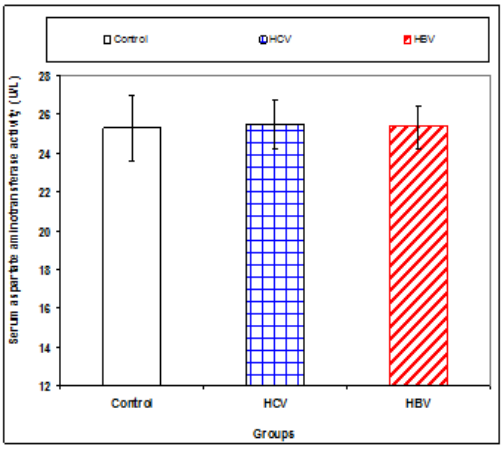

Figure 2 - Serum aspartate aminotransferase activity of male patients infected with HCV and HBV.

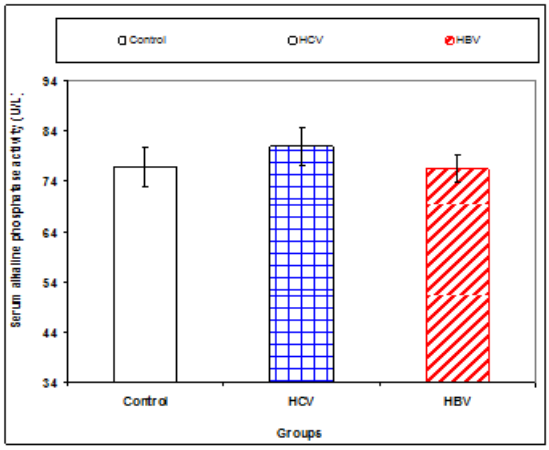

Figure 3 - Serum alkaline phosphatase activity of male patients infected with HCV and HBV.

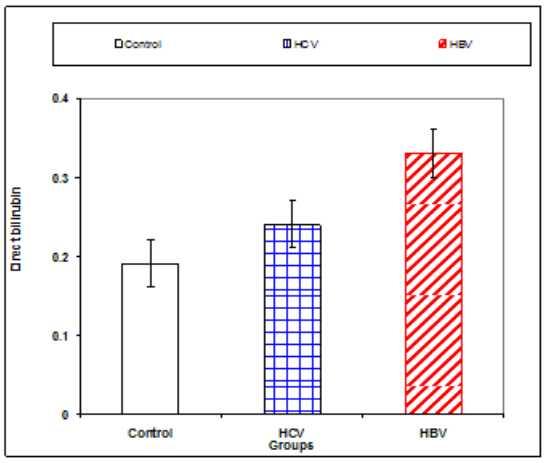

Figure 4 - Serum direct bilirubin concentration of male patients infected with HCV and HBV.

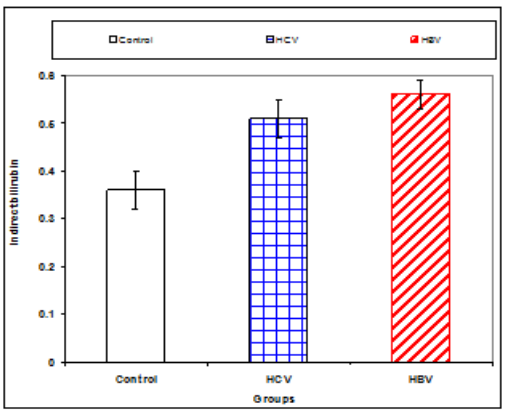

Figure 5 - Serum indirect bilirubin concentration of male patients infected with HCV and HBV.

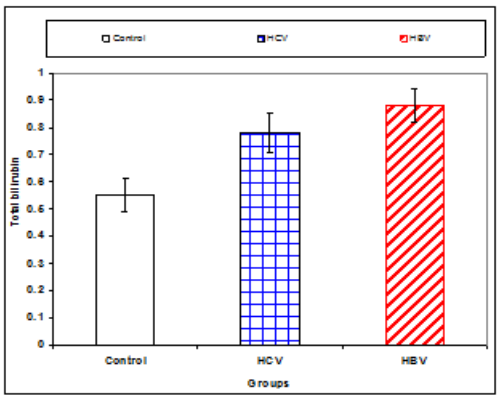

Figure 6 - Serum total bilirubin concentration of male patients infected with HCV and HBV.

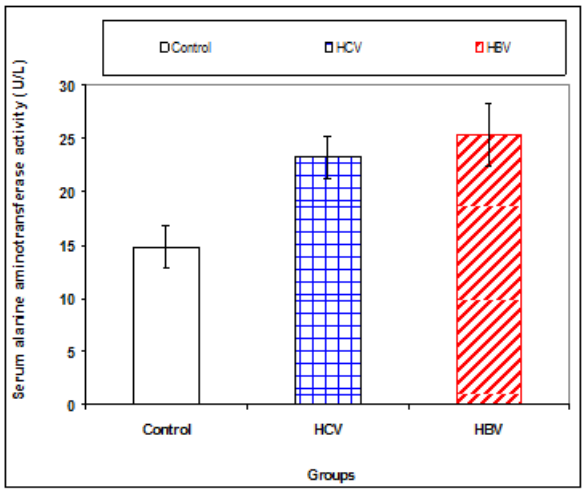

Figure 7 - Serum alanine aminotransferase activity of female patients infected with HCV and HBV.

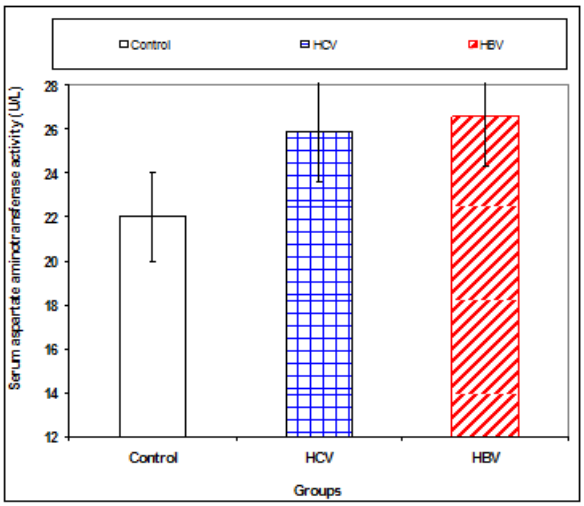

Figure 8 - Serum aspartate aminotransferase activity of female patients infected with HCV and HBV.

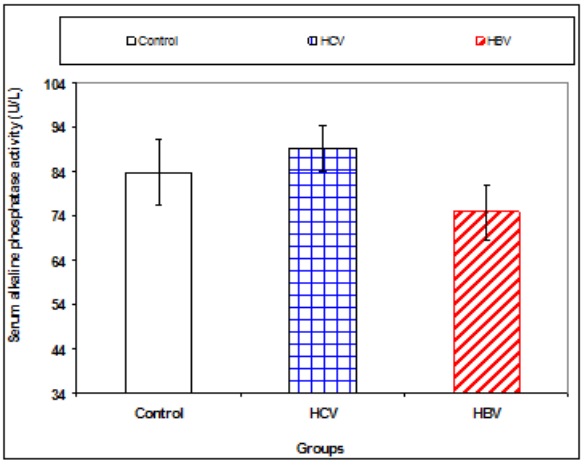

Figure 9 - Serum alkaline phosphatase activity of female patients infected with HCV and HBV.

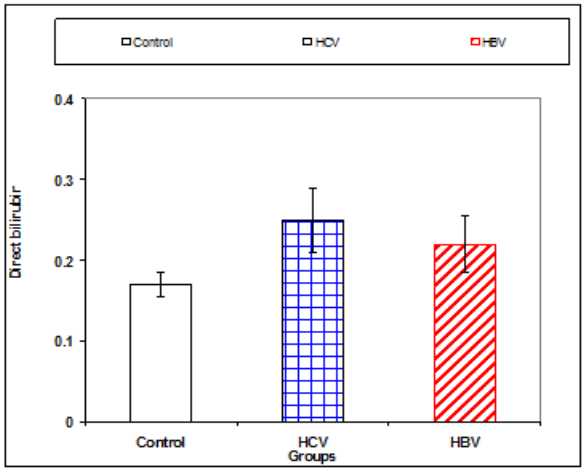

Figure 10 - Serum direct bilirubin concentration of female patients infected with HCV and HBV.

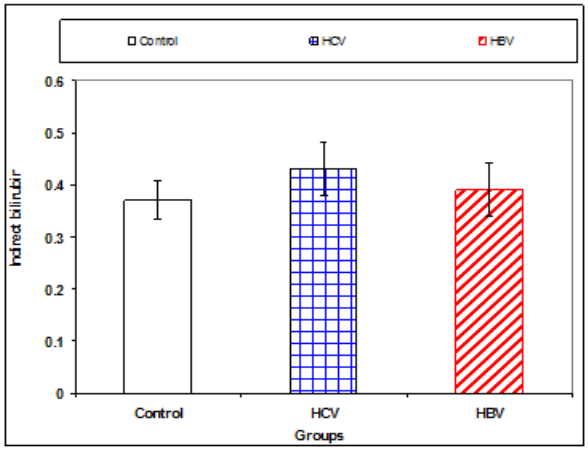

Figure 11 - Serum indirect bilirubin concentration of female patients infected with HCV and HBV

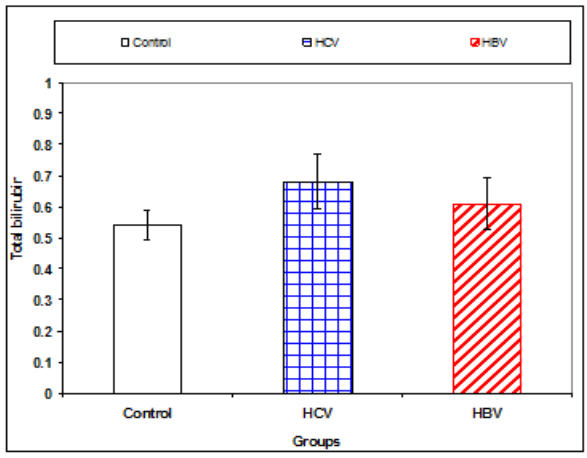

Figure 12 - Serum total bilirubin concentration of female patients infected with HCV and HBV.

Table 3 - Biochemical parameters of male patients infected with HCV and HBV.

Groups / Parameters	Control n= 10 Mean ± SE	HCV N=38 Mean ± SE	HBV n=43 Mean ± SE	F	P Value
ALT (U/L)	16.80± 2.39[bc]	23.58 ± 1.49[a]	26.53 ± 1.56[a]	4.36	0.016
AST (U/L)	25.30 ± 1.68	25.50 ± 1.22	25.37 ± 1.10	0.01	0.995
ALP (U/L)	76.90 ± 3.93	80.95 ± 3.91	76.44 ± 2.69	0.53	0.590
Direct Bili mg/dl	0.19 ± 0.031[c]	0.27 ± 0.03	0.33 ± 0.03[a]	2.54	0.085
Indirect Bil mg/dl	0.36 ± 0.04[c]	0.51 ± 0.04	0.56 ± 0.03[a]	3.11	0.049
Total Bil mg/dl	0.55 ± 0.06[c]	0.78 ± 0.07	0.88 ± 0.06[a]	3.16	0.047

[a] = Refers to the relation of control group with other groups, [b] = Refers to the relation of HCV group with other groups, [c] = Refers to the relation of HBV group with other groups. Variation between similar single letters in each components is significant at P <0.05.

Table 4 - Biochemical parameters of female patients infected with HCV and HBV.

Groups / Parameters	Control n= 10 Mean ± SE	HCV n=22 Mean ± SE	HBV n=17 Mean ± SE	F	P Value
ALT (U/L)	14.80 ± 2.00[bc]	23.23 ± 1.99[a]	25.35 ± 2.94[a]	3.74	0.031
AST (U/L)	22.00 ± 2.06	25.91 ± 2.33	26.59 ± 2.29	0.77	0.467
ALP (U/L)	83.80 ± 7.52	89.23 ± 5.10	74.88 ± 6.16	1.65	0.201
Direct Bili mg/dl	0.17 ± 0.015	0.25 ± 0.040	0.22 ± 0.035	0.83	0.443
Indirect Bil mg/dl	0.37 ± 0.037	0.43 ± 0.051	0.39 ± 0.051	0.37	0.691
Total Bil mg/dl	0.54 ± 0.048	0.68 ± 0.086	0.61 ± 0.083	0.57	0.571

[a] = Refers to the relation of control group with other groups, [b] = Refers to the relation of HCV group with other groups, [c] = Refers to the relation of HBV group with other groups. Variation between similar single letters in each components is significant at P < 0.05.

Table 5 - Biochemical parameters of male and female patients infected with HCV and HBV.

Groups / Parameters	Control n= 20 Mean ± SE	HCV n=60 Mean ± SE	HBV n=60 Mean ± SE	F	P Value
ALT (U/L)	15.80 ± 1.54[bc]	23.45 ± 1.18[a]	26.20 ± 1.38[a]	8.80	0.000
AST (U/L)	23.65 ± 1.35	25.65 ± 1.14	25.72 ± 1.02	0.54	0.582
ALP (U/L)	80.35 ± 4.20	83.98 ± 3.12[c]	76.00 ± 2.57[b]	2.03	0.135
Direct Bili mg/dl	0.18 ± 0.017[c]	0.26 ± 0.024	0.30 ± 0.024[a]	3.42	0.036
Indirect Bil mg/dl	0.37 ± 0.026[c]	0.48 ± 0.033	0.51 ± 0.029[a]	3.09	0.049
Total Bil mg/dl	0.55 ± 0.039[bc]	0.74 ± 0.054a	0.81 ± 0.049[a]	3.60	0.030

[a] = Refers to the relation of control group with other groups, [b] = Refers to the relation of HCV group with other groups, [c] = Refers to the relation of HBV group with other groups. Variation between similar single letters in each components is significant at P < 0.05.

General Correlation

Generally, to confirm the speculation about the presence of significant correlation between different parameters in both groups of patients (HCV and HBV), we made a general comparison between those parameters in both conditions as compared with the control group. This study found a highly significant correlation between TB and direct, indirect bilirubin and AST (P<0.01) whereas there was no correlation between TB and other parameters (ALP, ALT and Hb) (Table 8). Inversely, in patients infected with HBV, there was a significant correlation between Hb and other biochemical parameters such as correlation of haemoglobin with total, direct & indirect bilirubin activities (P value < 0.05). Distinctly, there was a significant correlation between TB with direct and indirect bilirubin (P < 0.01) (Table 7). Generally, this result confirms our speculation about the presence of a significance correlation between these parameters in both groups of patients (HCV and HBV). In order to achieve this relation, this study made a general comparison between those parameters in both conditions and found a highly significant correlation between total bilirubin, direct bilirubin, indirect bilirubin and AST (P< 0.01) whereas there was no correlation between TB and other parameters (ALP, ALT and Hb) (Table 8).

Table 6 - Correlation between Hb, and total, direct & indirect bilirubin, ALT, AST, and ALP activities in patients infected with HCV.

Parameters		Hb %	Total Bil	Direct Bil	Indirect Bil	ALP	AST	ALT
Correlation of hemoglobin with total, direct & indirect bilirubin, ALT, AST, and ALP activities	Pearson r	-	0.07	0.037	0.088	-0.022	0.029	0.196
	p value (two tailed)	-	0.594	0.779	0.501	0.87	0.826	0.134
	p value summary	-	ns	ns	ns	ns	ns	Ns
Correlation of total bilirubin with direct& indirect bilirubin, ALT, AST, and ALP activities	Pearson r	0.07	-	0.932	0.965	0.08	0.378	0.096
	p value (two tailed)	0.594	-	0.000	0.000	0.543	0.003	0.467
	p value summary	ns	-	**	**	ns	**	Ns
Correlation of direct bilirubin with total& indirect bilirubin, ALT, AST, and ALP activities	Pearson r	0.037	0.932	-	0.805	0.099	0.280	0.075
	p value (two tailed)	0.779	0.000	-	0.000	0.453	0.03	0.567
	p value summary	ns	**	-	**	ns	*	Ns
Correlation of indirect bilirubin with total& direct bilirubin, ALT, AST, and ALP activities	Pearson r	0.088	0.965	0.805	-	0.06	0.417	0.102
	p value (two tailed)	0.501	0.000	0.000	-	0.65	0.001	0.437
	p value summary	ns	**	**	-	ns	**	Ns
Correlation of ALP activity with total, direct & indirect bilirubin, ALT, and AST activities	Pearson r	-0.022	0.08	0.099	0.06	-	-0.047	0.119
	p value (two tailed)	0.87	0.543	0.453	0.65	-	0.719	0.366
	p value summary	ns	ns	ns	ns	-	ns	Ns
Correlation of AST activity with total, direct & indirect bilirubin, ALT, and ALP activities	Pearson r	0.029	0.378	0.280*	0.417	-.047	-	0.440
	p value (two tailed)	0.826	0.003	0.03	0.001	0.719	-	0.000
	p value summary	ns	**	*	**	ns	-	**
Correlation of ALT activity with total, direct & indirect bilirubin, AST, and ALP activities	Pearson r	0.196	0.096	0.075	0.102	0.119	0.440	-
	p value (two tailed)	0.134	0.467	0.567	0.437	0.366	0.000	-
	p value	ns	ns	ns	ns	ns	**	-

****: Highly significant correlation (p<0.01), *: Significant correlation (p<0.05), ns: non significant.**

Table 7 - Correlation between Hb, and Bil, ALT, AST, and ALP activities in pts infected with HBV.

Parameters		Hb %	Total Bil	Direct Bil	Indirect Bil	ALP	AST	ALT
Correlation of haemoglobin with total, direct & indirect bilirubin, ALT, AST, and ALP activities	Pearson r	-	0.296	0.269	0.292	-0.043	-0.100	0.083
	p value (two tailed)	-	0.022	0.039	0.024	0.745	0.447	0.53
	p value summary	-	*	*	*	ns	ns	Ns
Correlation of total bilirubin with direct& indirect bilirubin, ALT, AST, and ALP activities	Pearson r	0.296	-	0.933	0.955	-0.018	0.023	0.018
	p value (two tailed)	0.022	-	0.000	0.000	0.894	0.863	0.892
	p value summary	*	-	**	**	ns	ns	Ns
Correlation of direct bilirubin with total & indirect bilirubin, ALT, AST, and ALP activities	Pearson r	0.269	0.933	-	0.788	0.013	0.016	0.013
	p value (two tailed)	0.039	0.000	-	0.000	0.923	0.902	0.924
	p value summary	*	**	-	**	ns	ns	Ns
Correlation of indirect bilirubin with total & direct bilirubin, ALT, AST, and ALP activities	Pearson r	0.292	0.955	0.788	-	-.040	0.026	0.026
	p value (two tailed)	0.024	0.000	0.000	-	0.76	0.844	0.842
	p value summary	*	**	**	-	ns	ns	Ns
Correlation of ALP activity with total, direct & indirect bilirubin, ALT, and AST activities	Pearson r	-0.043	-0.018	0.013	-0.040	-	-0.070	-0.124
	p value (two‑tailed)	0.745	0.894	0.923	0.76	-	0.597	0.346
	p value summary	ns	Ns	ns	ns	-	ns	Ns
Correlation of AST activity with total, direct & indirect bilirubin, ALT, and ALP activities	Pearson r	-0.100	0.023	0.016	0.026	-0.070	-	0.551
	p value (two tailed)	0.447	0.863	0.902	0.844	0.597	-	0.000
	p value summary	ns	Ns	ns	ns	ns	-	**
Correlation of ALT activity with total, direct & indirect bilirubin, AST, and ALP activities	Pearson r	0.083	0.018	0.013	0.026	-0.124	0.551	-
	p value (two tailed)	0.53	0.892	0.924	0.842	0.346	0.000	-
	p value	ns	Ns	ns	ns	ns	**	-

****: Highly significant correlation (p<0.01), *: Significant correlation (p<0.05), ns: non significant.**

Table 8 - Correlation between Hb, and Bil, ALT, AST, and ALP activities in patients infected with HCV & HBV.

Parameters		Hb %	Total Bil	Direct Bil	Indirect Bil	ALP	AST	ALT
Correlation of haemoglobin with total, direct & indirect bilirubin, ALT, AST, and ALP activities	Pearson r	-	0.160	0.144	0.157	-0.096	-0.010	0.117
	p value (two tailed)	-	0.059	0.091	0.063	0.257	0.903	0.168
	p value summary	-	Ns	ns	ns	ns	ns	Ns
Correlation of total bilirubin with direct& indirect bilirubin, ALT, AST, and ALP activities	Pearson r	0.160	-	0.933	0.961	0.026	0.234	0.130
	p value (two tailed)	0.059	-	0.000	0.000	0.762	0.005	0.125
	p value summary	ns	-	**	**	ns	**	Ns
Correlation of direct bilirubin with total & indirect bilirubin, ALT, AST, and ALP activities	Pearson r	0.144	0.933	-	0.797	0.041	0.183	0.118
	p value (two tailed)	0.091	0.000	-	0.000	0.634	0.031	0.168
	p value summary	ns	**	-	**	ns	*	Ns
Correlation of indirect bilirubin with total & direct bilirubin, ALT, AST, and ALP activities	Pearson r	0.157	0.961	0.797	-	0.013	0.253	0.131
	p value (two tailed)	0.063	0.000	0.000	-	0.879	0.003	0.123
	p value summary	ns	**	**	-	ns	**	Ns
Correlation of ALP activity with total, direct & indirect bilirubin, ALT, and AST activities	Pearson r	-0.096	0.026	0.041	0.013	-	-0.052	-0.010
	p value (two tailed)	0.257	0.762	0.634	0.879	-	0.540	0.910
	p value summary	ns	Ns	ns	ns	-	ns	Ns
Correlation of AST activity with total, direct & indirect bilirubin, ALT, and ALP activities	Pearson r	-0.010	0.234	0.183	0.253	-0.052	-	0.483
	p value (two tailed)	0.903	0.005	0.031	0.003	0.54	-	0.000
	p value summary	ns	**	*	**	ns	-	**
Correlation of ALT activity with total, direct & indirect bilirubin, AST, and ALP activities	Pearson r	0.117	0.130	0.118	0.131	-.010	0.483	-
	p value (two tailed)	0.168	0.125	0.168	0.123	0.91	0.000	-
	p value	ns	Ns	ns	ns	ns	**	-

****: Highly significant correlation (p<0.01), *: Significant correlation (p<0.05), ns: non significant.**

DISCUSSION

Viral hepatitis continues to be a disease of major significance, in terms of both morbidity and mortality. Chronic viral hepatitis is an important health problem worldwide, where hepatitis B virus (HBV) or hepatitis C virus (HCV) infections are the main causes of liver insufficiency.[10,12]

It is well established that ALT and AST are crucial biological markers widely used for liver diseases. All types of hepatitis and cirrhosis have been reported to cause liver damage that can elicit elevations in serum ALT and AST activities.[13,14] All patients infected with HCV and HBV had significantly higher biochemical parameters (ALT, AST, Direct and Indirect Bilirubin). However, ALP levels were of lower significance, as compared with control group (Table 5). These results had *P* values of < 0.01 and < 0.05, respectively. These are in confirmation with many other studies.[14, 15]

Biochemical parameters for those infected with both HCV and HBV were significantly high in contrast to those for the control subjects. In divergence to HBV-infected individuals, HCV patients showed slightly lower

parameters, particularly for ALT (*P* value < 0.001), and direct, indirect and total bilirubin (Table 5). Interestingly, there was a noticeable difference in liver function tests among the two infected groups. One remarkable variation was regarding ALP, where it was higher in those infected with HCV than HBV patients. This also accords with earlier observations, which showed that HCV-infected patients presented more liver inflammation (higher ALP) than HBV-infected patients.[16]

To identify any associations between Hb concentration in patients with HCV and HBV and other biochemical parameters, this current study strived to apply statistical analysis to determine such a connection. Table 7 portrays the correlation aspects between the concentration of Hb and all other biochemical parameters in both HBV- and HCV-infected patients.

Contrary to expectations, this study did not find a correlation between Hb concentration and the other biochemical parameters (0.196 Pearson and 0.134 two-tailed). However, the observed correlation between AST activity and total bilirubin, indirect bilirubin, and ALT (P < 0.01) was high. Moreover, a slightly low correlation was also perceived between AST activity and direct bilirubin (P < 0.05). Adversely, no statistical correlation between the parameters, Hb concentration and ALP was contemplated.

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