

The Effect of Chronic Liver Diseases on Some Biochemical Parameters in Patients Serum

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Abstract: Sixty two patients with chronic liver disease and 26 healthy individuals were included as normal controls for the study. Blood analysis was carried out for all of them which include serum bilirubin, total protein, creatinine and liver enzyme tests (GOT, GPT, ALP) levels. The result of blood parameters showed that: (total bilirubin and direct bilirubin ($\mu\text{mol/L}$)) have a higher level of bilirubin in viral groups (Chronic viral hepatitis) than in the cirrhosis and liver cancer patients, but all groups showed increased bilirubin levels more than in the control group ($p < 0.001$). Liver enzyme tests show the concentration of serum Glutamic Pyruvic Transaminase (GPT), serum Glutamic Oxaloacetic Transaminase (GOT) and serum Alkaline Phosphates (ALP) levels are increased highly significant in cirrhotic patients compared to other groups of control patients ($p < 0.001$). There was a decrease in total protein concentrations observed in cirrhosis and liver cancer (hepato-cellular Carcinoma) patients compared to the control group. The level of creatinine concentration for patients with cirrhosis and hepato-Cellular Carcinoma were increased highly significant ($p < 0.01$) than other groups with Chronic viral hepatitis and control groups.

Keywords: ALP, bilirubin, chronic liver diseases, cirrhosis, GOT, GPT, hepato-cellular carcinoma, serum bilirubin creatinine

INTRODUCTION

To identify the etiological relationship between homocysteine level and trace elements in blood with patients in chronic liver disease (cirrhosis, hepatocellular carcinoma, chronic viral hepatitis). Patients compared with normal individuals. To Measure the concentration of serum bilirubin, creatinine, liver enzymes (GOT, GPT, ALP) and total protein.

Bilirubin is produced by normal breakdown of pigment-containing proteins, especially hemoglobin from senescent red blood cells and myoglobin from muscle breakdown. Bilirubin released from such sources, tightly Albumin-bound, is delivered to the liver, where it is efficiently extracted and conjugated by hepatic glucuronidation and sulfation. Conjugated bilirubin is rapidly excreted into bile and removed from the body through the gut. Therefore, the amount of conjugated bilirubin, present in the serum of healthy subjects, is trivial (less than 10% of measured total bilirubin). Elevated levels of conjugated serum bilirubin imply liver disease. Because only conjugated bilirubin appears in urine the finding of bilirubinemia also implies liver disease (Blanckaert, 1997; Hickman *et al.*, 2004). Total bilirubin levels (the sum of the conjugated and non-conjugated portions). It sometimes useful to determine the fraction of total serum bilirubin that is

non-conjugated versus conjugated this is usually referred to as fractionation of bilirubin.

Hypoalbuminemia is noted in various liver disorders. Hepatic synthesis of albumin is decreased in patients with cirrhosis and liver cancer which with portal hypertension, Albumin leaks off the liver surface into the peritoneal cavity. This redistribution may contribute to the production of ascites by increasing oncotic pressure in the peritoneal cavity (Tredger and Sherwood, 1997).

AST, (GOT) and ALT (GPT) have been suggested for the two enzymes of greatest clinical significance in viral hepatitis and other forms of liver disease associated with hepatic necrosis serum AST (GOT) and ALT (GPT) levels which are elevated even before the clinical signs and symptoms of disease (such as jaundice) appear. Levels for both enzymes may reach values as high as the upper limits of the reference interval (Panteghini, 1990; Clark *et al.*, 2002).

The Amino-Transferase levels observed in cirrhosis with the level of AST activity are higher than that of ALT activity. Elevations of both enzymes occur in patients with primary or metastatic Carcinoma of the liver, with AST (GPT). But levels are often normal in the early stages of malignant infiltration of the liver (Schmidt, 1978).

The liver is the primary site of the synthesis of plasma proteins, with synthesis occurring on the rough endoplasmic reticulum of the hepatocytes (Globulins, Albumin and clotting factors) but not-globulin, which is produced by B-lymphocytes. The pattern of plasma protein alterations depends on the type, severity and duration of liver injury. The aim of this study was to obtain clear profiles of the changes by using biochemical parameters in chronic liver disease.

MATERIALS AND METHODS

This study was carried out in the specialized Center of Gastroenterology and Hepatology in Baghdad, during the period from October 2003 up to the end of November 2004. Sixty two patients were admitted to the center; all of them fulfill the criteria of chronic liver disease. From each patient, a detailed history was taken concerning the illness, the age at the onset of illness (age at first presentation of symptoms) and the age at presentation (age at which the patient consults his physician), complications of the disease, other associated diseases residency and family history of the same illness and their jobs or whether taking any drugs, smoking or working. These patients were subdivided to 3 groups:

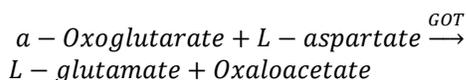
- **Liver cirrhosis:** includes 25 patients (15 males and 10 females), ages (7-73) years, (mean±S.D: (44.92±15.411) years)
- **Liver jaundice:** (HCV, HBV (hepatitis C and B viruses) chronic hepatitis, Gall Bladder Stones), includes 24 patients (16 males and 8 females) ages (17-73) years; mean±S.D.: (43.00±14.679)
- **Liver mass:** (Hepato-cellular Carcinoma) includes 13 patients (5 males and 8 females), ages (27-66) years, (mean±S.D.: (47.62±13.95) years)

Twenty-six healthy individuals were included as normal controls for the study (13 males and 13 females) ages (15-60) years: mean±S.D. (37.92±12.881) years.

Determination of serum bilirubin of chronic liver disease by using kit provided from SyRBio Company (Paris-France) and measured total bilirubin by absorbance at 578 nm and direct bilirubin at 546 nm.

Determination of serum Glutamic-Oxaloacetic Transaminase (GOT) using kit provided from Randox Company (United Kingdom).

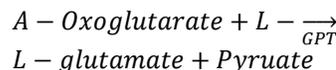
Glutamic-Oxaloacetic Transaminase is measured by monitoring the concentration of Oxaloacetate Hydrazone formed with 2, 4-dinitrophenylhydrazine.



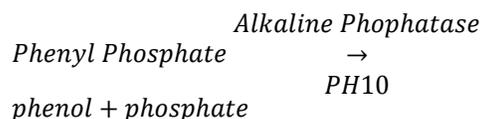
Read the absorbance at (540-550 nm).

Determination of Glutamic-pyruvic transaminase GPT by using kit provided from RANDOX Company (United Kingdom).

Glutamic-pyruvic transaminase is measured by monitoring the concentration of Pyruvate hydrazone formed with 2.4 Dinitrophenyl Hydrazine:



Determination of Alkaline Phosphates (ALP) by using kit provided from bio Mérieux/ France) colorimetric determination of alkaline phosphates activity according to the following reaction:



The phenol liberated is measured in the presence of amino-4-antipyrine and potassium ferri-Cyanide. The presence of sodium arsenate in the reagent stops the enzymatic reaction. The absorbance respect to the time of standard and serum sample and serum blank at 510 nm. Determination of Total protein by Biuret method.

RESULTS AND DISCUSSION

The results in Table 1 and 2 clearly show the differences in the total serum bilirubin (µmol/L) and the direct bilirubin concentrations of control the group and the patient groups with various types of chronic liver disease. The mean value differences were statistically and highly significant, increased (p<0.001) (Fig. 1 and 2).

In the present study the main clinical situation in which this is useful is when all of the standard liver tests are normal except the total bilirubin. This study

Table 1: Serum total bilirubin concentration (µmol/L) in patient group and control group

Group	N	Min	Max	Mean	S.D.	SEM
Cirrhosis	25	48.9	76.3	59.68	7.73	1.546
Chronic viral hepatitis	24	58.2	84.9	75.13	6.66	1.361
Hepatocellular carcinoma	13	48.2	56.9	52.36	3.34	0.928
Control	26	9.8	15.9	12.33	1.32	0.260

F: 341.904; p: 0.001 (significant); Min: Minimum; Max: Maximum

Table 2: Serum direct bilirubin concentration (µmol/L) in patients groups and control group

Group	N	Min	Max	Mean	S.D.	SEM
Cirrhosis	25	17.0	24.9	20.54	2.59	0.518
Chronic viral hepatitis	24	39.2	75.4	48.52	11.5	2.350
Hepatocellular carcinoma	13	18.9	24.4	21.63	2.24	0.623
Control	25	2.8	3.9	3.240	0.31	0.062

F: 43.914; p: 0.001 (significant); Min: Minimum; Max: Maximum

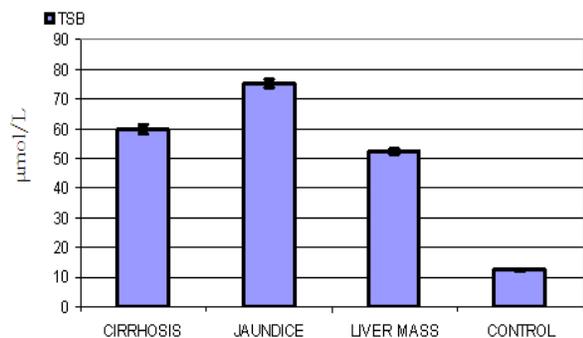


Fig. 1: Serum level of bilirubin in control and patients group is defined in the text

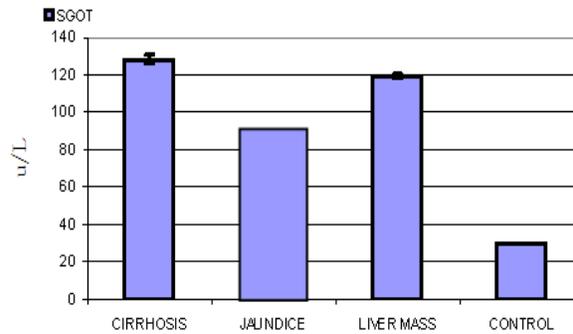


Fig. 3: Serum GOT concentration in patients groups and control

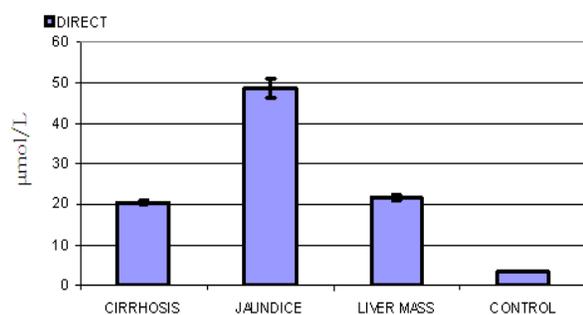


Fig. 2: Serum direct bilirubin level in patients groups and control subject

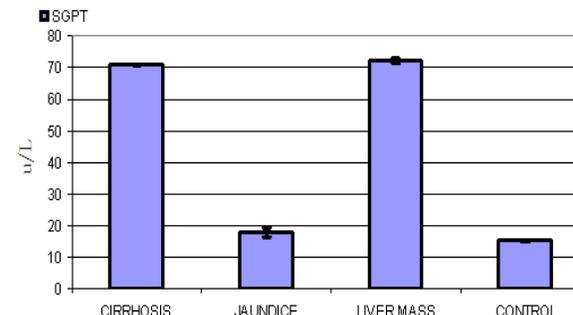


Fig. 4: Serum GPT concentration in patients groups and control

demonstrates that hyperbilirubinemia develops more in chronic viral hepatitis than in all patients.

Total bilirubin as well as the conjugated and non-conjugated portions; to make matters more confusing, the conjugated bilirubin is sometimes referred to as the direct-reacting bilirubin and the non-conjugated as the indirect-reacting bilirubin (Balistreri and Rej, 1994). Elevations of non-conjugated bilirubin, when the conjugated bilirubin remains normal, it may also indicate an increased load of bilirubin due to Haemolysis. The bilirubin level may be normal or elevated in each type of disorder. Viral hepatitis A (prototypical hepato-cellular disease) may frequently be associated with bilirubin levels that are quite high, whereas primary biliary cirrhosis (a prototypical cholestatic disorder) is associated with a normal serum bilirubin level except in a later stage of disease. Serum bilirubin levels should be disregarded when trying to decide if the liver test pattern is more suggestive of hepato-cellular or cholestatic disease (Doumas *et al.*, 1973; Kramer *et al.*, 2002).

The present study shows a higher level of bilirubin in viral group (jaundice) than the cirrhosis and liver cancer groups but all groups show the increase bilirubin (total and direct and indirect) level than in control group. In liver disease, bilirubin levels in blood can become high measurements of total and direct bilirubin can be helpful in diagnosing specific liver problems.

Glutamic-Oxaloacetic Transaminase (GOT) and Glutamic Pyruvic Transaminase (GPT): The study of liver enzyme shows highly significant differences in liver enzyme concentrations between the patients groups and control group. Table 3 shows the concentrations of serum Glutamic-Oxaloacetic Transaminase (AST) (GOT) unit/Liter is highly increased in cirrhosis and hepatocellular carcinoma groups more than in the control and chronic viral hepatitis groups ($p < 0.001$)

Table 3: Serum GOT concentration (u/L) in patients groups and control

Group	N	Min	Max	Mean	S.D.	SEM
Cirrhosis	25	111.3	142.6	128.3	11.2	2.244
Chronic viral hepatitis	24	73.1	99.9	85.04	8.40	1.716
Hepatocellular carcinoma	13	110.4	124.3	119.3	4.38	1.216
Control	26	24.3	35.5	29.77	2.85	0.559

F: 289.143; p: 0.001 (significant); Min: Minimum; Max: Maximum

Table 4: Serum GPT concentration (u/L) in patients groups and control

Group	N	Min	Max	Mean	S.D.	SEM
Cirrhosis	25	68.2	74.4	70.94	1.52	0.304
Chronic viral hepatitis	24	8.5	37.2	17.67	7.47	1.526
Hepatocellular carcinoma	13	68.4	78.3	72.14	2.90	0.805
Control	26	13.0	17.5	15.05	1.23	0.241

F: 77.647; p: 0.001 (significant); Min: Minimum; Max: Maximum

Table 5: Serum ALP concentration (u/L) in patients groups and control group

Group	N	Min	Max	Mean	S.D.	SEM
Cirrhosis	25	135	160	145.74	6.43	1.28
Chronic viral hepatitis	24	85	99	92.51	4.01	0.82
Hepatocellular carcinoma	13	118	132	126.24	4.60	1.27
Control	26	60	85	76.03	7.61	1.49

F: 386.418; p: 0.001 (significant); Min: Minimum; Max: Maximum

Table 6: Serum total protein level (g/L) in patients groups and control group

Group	N	Min	Max	Mean	S.D.	SEM
Cirrhosis	25	48.9	69.7	56.36	5.82	1.165
Chronic viral hepatitis	23	57.2	77.8	67.63	5.85	1.219
Hepatocellular carcinoma	13	50.8	64.7	57.52	4.96	1.377
Control	26	61.2	89.5	69.16	5.30	1.039

F: 32.148; p: 0.001 (significant); Min: Minimum; Max: Maximum

(Fig. 3). Table 4 shows the concentrations of serum Glutamic Pyruvic Transamine serum (ALT) (GPT) unit/Liter is increased highly in cirrhotic patients and other groups with the control (p<0.001) (Fig. 4).

In various forms of liver disease, serums of numerous cytosolic, mitochondrial and membrane-associated enzymes are increased; the elevation of specific enzymes varies with the type of disease. These iso-enzymes are genetically distinct and different in their amino and composition, kinetic behavior, electrophoretic mobilities and immune chemical properties (Panteghini, 1990; Rej and Horder, 1983; Rej, 1984). In the present study, the increased serum ALT and AST enzyme levels were observed in cirrhotic, hepatocellular carcinoma and chronic viral hepatitis when compared to the control subjects and the results are similar to screening and diagnostic evaluation for liver diseases.

But the liver is a complex organ with interdependent metabolic, excretory and defense functions. No single or simple test assesses over all liver functions; sensitivity and specificity are limited. The use of several screening tests improves the detection of hepato-biliary abnormalities, helps differentiate the basis for clinically suspected disease and determines the severity of liver disease. Many tests are available, but relatively few improve patient care. In this study, the comparison of means between control the group and the patient groups showed highly increasing levels in serum ALP. Hepatic alkaline phosphates are most densely represented near the canalicular membrane of the hepatocyte. Obstructive diseases, bile duct obstruction, primary biliary cirrhosis are some examples of diseases in which elevated alkaline phosphates levels are often predominant over Transaminase elevation.

Table 5 shows the concentrations of serum Alkaline Phosphates (ALP) unit/Liter are highly increased

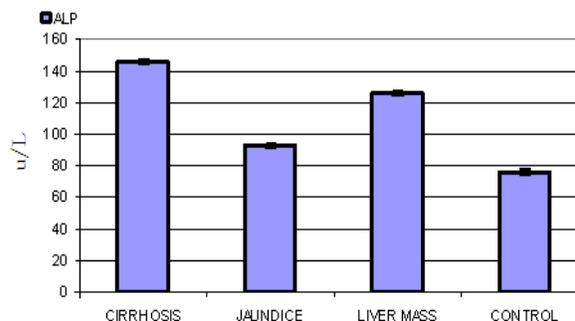


Fig. 5: Serum ALP concentration in patients groups and control

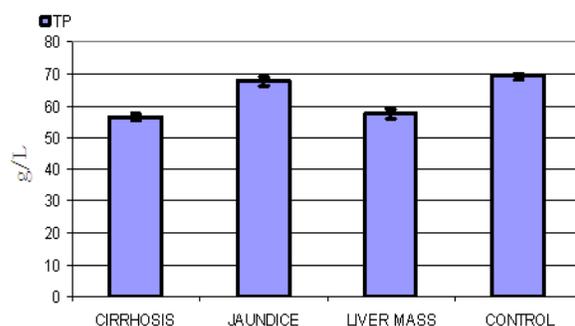


Fig. 6: Serum total protein level in patients groups and control group

significant in cirrhosis and hepatocellular carcinoma and chronic viral hepatitis groups more than the control groups (p<0.001) (Fig. 5). (EC.3.1.3.1) Ortho phosphoric-Monoester Phosp hohydrolase Alkaline Phosphates (ALP) is an enzyme found in the liver and bone. For many years, it was believed that ALP reaching the liver from other tissues (especially bone) was excreted into the bile and that the elevated serum enzyme activity found in hepato-biliary disease was a result of a failure to excrete the enzyme through the bile. However, it is now known that the response of the liver to any form of biliary tree obstruction is to synthesize more ALP. The main site of new enzyme synthesis is the hepatocytes adjacent to the biliary. Some of the newly formed enzymes enter the circulation to raise the enzyme level in serum (Smit *et al.*, 1987).

The detailed mechanism by which enzymes released from the Cytosol and mitochondria of hepatocytes into the blood stream is not completely known. Clinical observations and experimental studies have shown that subtle membrane changes are sufficient to allow passage of intracellular enzymes to the intracellular space (Tredger and Sherwood, 1997). A very large concentration gradient between the hepatocyte and the sinusoidal space usually exists for enzymes. Cell damage increases membrane permeability, causing cytosolic iso-enzymes to spill into

the sinusoids and from there into the peripheral blood. Permeability of mitochondrial membranes is also increased (Freidel *et al.*, 1979).

The difference in total protein is shown in Table 6 and Fig. 6: the mean decrease of total protein (g/L) concentration was more observed in cirrhosis and hepatocellular carcinoma groups than in the control and chronic viral hepatitis groups.

The mean value difference of serum Albumin level is more highly decreased in cirrhosis and than hepatocellular carcinoma in the control and chronic viral hepatitis groups. Decreased ($p < 0.001$).

The results of this study indicate that the total serum protein decreases significantly ($p < 0.001$) in cirrhosis and hepatocellular carcinoma patient groups that means impaired hepatic function. Because the liver reserves capacity and the relatively long half-life of these proteins, significant decreases will not become apparent except in severe or long-standing hepatic disease (Hutchinson *et al.*, 1981) a total protein test measure is used as a screening process to diagnose a number of illnesses, but the most important to screening the liver disease is serum Albumin level. Because the Albumin is synthesized exclusively by the liver; it is secreted from the golg: apparatus across the sinusoidal membrane of the hepatocyte (Castelino and Salern, 1997) in some, inflammatory conditions the release of tumor necrosis factor inhibits Albumin synthesis but induces the synthesis of proteins of the acute phase response. In this study, the serum Albumin level is decreased in cirrhosis and hepatocellular carcinoma more significantly ($p < 0.001$) than in the control group.

CONCLUSION

It can be concluded that is an elevation of blood bilirubin in all groups of study is compared to the control group. There is an elevation of liver enzyme (GOT, GPT, ALP) is highly significant in cirrhotic patients compared to control group ($p < 0.001$). The levels of serum total protein g/L were significantly decreased in cirrhosis and liver cancer groups patients more than the control and jaundice groups.

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