Serum Clusterin In Patients With Chronic HCV infection Related Cirrhosis

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Abstract:

Cirrhosis is a condition in which the liver does not function properly due to long term damage. Typically, the disease comes on slowly over months or years. Cirrhosis is characterized by the replacement of normal liver tissue by scar tissue.

Clusterin (apolipoprotein J) is a 75 - 80 kDa disulfide-linked heterodimeric protein associated with the clearance of cellular debris and apoptosis**.**

Objective: We aimed to evaluate the diagnostic performance of serum CLU level in diagnosing chronic HCV infection-related cirrhosis, and comparing it to that of alpha fetoprotein (AFP).

Methods: Case control study was carried out in the Clinical Pathology Department, Faculty of medicine, Sohag university hospital.Twenty cases of apparently healthy subjects and 20 cases of chronic HCV infection-related cirrhosis (CHC cases) were included in this study. Serum CLU concentration was determined using a quantitative sandwich enzyme immunoassay technique.

Results: Serum clusterin level showed a significant decrease in the CHC group compared to the control group (88.3 ± 30.7 vs. 110.7 ± 30.1).

Conclusion: CLU is decreased in the serum of patients with established CHC.

Introduction

Cirrhosis is a condition in which the liver does not function properly due to longterm damage**.**This damage is characterized by the replacement of normal   liver tissue by scar tissue. Typically, the disease develops slowly over months or years. Early on, there are often no symptoms. As the disease worsens, a person may become tired, weak, itchy, have swelling in the lower legs, develop yellow skin, bruise easily, have fluid build up in the abdomen, or develop spider-like blood vessels on the skin. The fluid build-up in   the   abdomen   may become spontaneously infected. Other complications   include hepatic encephalopathy, bleeding from dilated veins   in   the   esophagus or dilated stomach   veins,   and liver cancer. Hepatic encephalopathy results in   confusion   and   may   lead to unconsciousness *(National Institute of Diabetes and Digestive and Kidney Diseases.,2015)*.

Cirrhosis is most commonly caused by alcohol, hepatitis   B, hepatitis   C, and non-alcoholic fatty liver disease *(Abubakar et al.,2015).*

Ideally, for timely treatment, the biomarker(s) that could recognize the fibrosis in the early stages of hepatic disorder to prevent the progression of cirrhosis   to   HCC   is   required *(Gangadharan et al.,2007).*

CLU   is   a   molecular   chaperone responsible for aiding protein folding of secreted proteins, and its three isoforms   have   been   differentially implicated in pro- or antiapoptotic processes. Through this function, CLU is involved in many diseases related to oxidative   stress,   including neurodegenerative   diseases,   cancers, inflammatory diseases, and aging (*Koltai, 2014 ,Lin et al., 2014 and Sansanwal et al., 2015)***.** CLU gene is

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highly conserved in species, and the protein is widely distributed in many tissues and organs, where it participates in a number of biological processes, including lipid transport, membrane recycling, cell adhesion, programmed cell death, and complement-mediated cell lysis (*Koltai, 2014 ,Lin et al., 2014 and Sansanwal et al., 2015).* CLU activity is also involved in infectious diseases, such as hepatitis C. CLU is induced by the stress of hepatitis C viral infection, which disrupts glucose regulation. The chaperone protein then aids hepatitis C viral assembly by stabilizing its core and NS5A units *(Lin et al.,2014).*

Objectives:

The aim of the present study was to determine serum CLU concentration in CHC, as well as assess the use of CLU measurement vs. AFP in the CHC patients.

Patients and methods:

Individuals in this study are classified into two groups based on clinical and laboratory characteristics:

- **Group (1)** which includes patients with chronic HCV infection-related cirrhosis (20) patients (CHC Group).
- **Group (2) or control group** which includes (20) apparently healthy age and sex-matched subjects with no history of previous liver disease.

All subjects (patients and controls) will be subjected to:

1- Complete history taking to retrieve information about health status, current medications, alcohol consumption, and history of viral or toxic hepatitis and general examination.

2- The following parameters are done in this study:

a. Routine investigations:
- Complete blood count.
- Prothrombin time and concentration.
- Liver function tests.
- Renal function tests.
- Random blood sugar.
- Serological testing for anti-HCV and hepatitis B virus surface antigen, anti-HIV and anti-bilharzial antibodies.

b. Specific investigations:
- AFP assays
- Serum Clusterin

Results of the study

This study was performed in Sohag University Hospital, on 20 patients known to have HCV ,in addition to 20 healthy individuals (age- and sex- matched). Data was analyzed using Statistical package for social sciences (IBM-SPSS), version 24 IBM-Chicago, USA (May 2016).Quantitative data was represented as mean, standard deviation, median and range. Data was analyzed using student t-test to compare means of two groups and ANOVA for comparison of the means of three groups or more.

The mean age in group I is 53.2 years, SD 4.09 years and the range is (45 –62years), the mean age in group II is 53.8 years, SD 5.5 years and the range is (46 –66years). No significant difference was found in comparing diseased group with controls (P=0.77) as in figure (1).
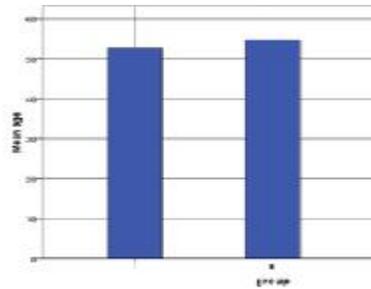
Figure (1):Comparison between the groups according to age.

All subjects in the control group had normal liver biochemistry but in CHC group was significantly higher (Table 1). As expected, serum AFP (ng/ml) was significantly higher in the CHC patients compared to the control group. Serum CLU (ng/ml) level showed a significant decrease in the CHC group compared to the control group (Table 2).

Table (1): Liver function tests in the studied groups

	Group I LC (n= 20)	Group II Control (n= 20)	P-value
	Mean ± SD	Mean ± SD	I versus II
ALT (IU/ L)	73.9 ± 27.4 45-123	27.45 ± 6.6 15-39	0.02(**Sig**)
AST (IU/ L)	81.65± 24.67 35-138	32.55 ± 6.5 18-41	<0.001(**HS**)
TP (g/ dL)	6.1 ± 0.48 5.3-6.9	7.29 ± 0.35 6.6-7.9	<0.001(**HS**)
Albumin(g/ dL)	2.2 ± 0.41 1.5-2.8	4.55 ± 0.46 3.9-5.2	<0.001(**HS**)
T.Bil (mg/ dL)	3.3 ± 0.95 1.6-4.9	0.7 ± 0.16 0.5-1	0.04 (**Sig**)
ALP (IU/ L)	111.6 ± 95.37 35-361	53.1 ± 14.9 35-92	0.03(**Sig**)

Table (2):AFP and CLU in the studied groups

	Group I LC (n= 20)	Group II Control (n= 20)	P value
	Mean ± S.D	Mean ± S.D	I versus II
AFP (ng/ mL)	**21.49 ± 21.17 0.9-60.2**	**2.28 ± 1.33 0.5-5.2**	**<0.01(HS)**
CLU (µg/ mL)	**88.3 ± 30.7 32-150**	**110.7 ± 30.1 60-164**	**=0. 03(Sig)**

Discussion

According to *WHO 2012*, every year, 3-4 million people are infected with the hepatitis C virus. About 150 million people are chronically infected and at risk of developing liver cirrhosis and/or liver cancer. More than 350000 people die from hepatitis C-related liver diseases every year.

AFP showed statistically significant elevation in group I LC patients as regard controls(P <0.01).The elevation of AFP occurs in hepatocyte regeneration, hepatocarcinogenesis, and embryonic carcinomas*(Malaguarnera, 2010).*

Serum CLU, Serum CLU showed a significant decrease in the LC group as regards controls. This may point to a possible protective role of CLU against liver cell fibrogenesis which ultimately ends in cirrhosis. Such an assumption was similarly postulated in renal

fibrosis by *Jung et al*., *2012*, who suggested that up regulation of clusterin during renal injury, in a mouse model, has a protective response against the development of renal fibrosis.

This is further sup-ported by the fact that clusterin exists as both an intracellular truncated form and an extracellular heterodimeric secreted glycoprotein, making clusterin the only known chaperone protein to be secreted*(Chi et al., 2008).*

CLU is a Golgi molecular chaperone involved in BAX- antiapoptotic processes, activation of the phosphatidylinositol 3-kinase/ protein kinase B pathway, promotion of angiogenesis, mediation of the nuclear factor kappa B (NF-κB) pathway and modulation of extra-cellular signal-regulated kinase (ERK) signaling. A number of biological processes, including programmed cell death (Down regulation allows for p53activation and cell death), lipid transport, membrane recycling and cell adhesion *(Sansanwal et al.,2015, Lin et al.,2014).* Serum clusterin was introduced as more specific and sensitive biomarker than AFP in distinction of HBV-cirrhosis with HCC base on HBV-cirrhosis *(Wang et al.,2010).* It also has been shown that clusterin may be a useful marker in the evaluation of prognosis of patients with alcoholic cirrhosis and severity of liver disease *(Ehsani Ardakani et al.,2016).* CLU involved in BAX-antiapoptotic processes and it's down regulation allows p53 activation and cell death *(Norouzinia et al.,2012).*

Conclusion

CLU is decreased in the serum of patients with established CHC.

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