Vitronectin, Integrin Vitronectin Receptor Other Biochemical Markers to Assess the Development of Liver Cirrhosis

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Abstract

Aim of study: To evaluate some fibrosis markers such as vitronectin, serum vitronectin receptor (VNR) and the amino terminal propeptide of type III procollagen (P-III-P) in the sera of liver cirrhosis patients and to monitor the behavior of these markers in disease progression. Material and Methods: About (60) volunteer participated in current study; 30 patients and 30 patients. The patients were divided according to severity of the disease by Child-Pugh Score to three classes: (10) patients class A, (10) class B patients and (10) class C. Procollagen III peptide, serum vitronectin and vitronectin receptor were measured in serum by ELISA. Results: A Significant increases were found in serum vitronectin, receptor and sP-III-P in all patients' sub-groups when compared to control. VNR seems to detect early liver cirrhosis and distinguish between patients (A and B classes) and advanced disease with cirrhosis. Conclusion: According to our data, the area under curve of serum VNR is greater than vitronectin and Sp-III-P so it can be act as a marker of liver cirrhosis and also could be used to monitor the severity of disease.

Introduction

Cirrhosis is the severe damage at the liver accompanied by significant loss of liver functions seen at the end stages of chronic liver disease. Cirrhosis gets due to several causes like viral infections or long-term exposure to toxins such as alcohol or fatty liver developed to cirrhosis or cryptogenic cirrhosis. The liver is located in the upper part of the abdomen in the right side. It is a very important part of the body where it performs many important functions such as storing sugar, detoxification, and producing clotting proteins [1].

The liver secrete a number of glycoproteins that play an important role in clinical diagnosis example ceruloplasmin, beta globulins, alpha antitrypsin and others[2]. Vitronectin is a glycoprotein that produced by the liver. It is one of the major cell adhesion proteins in blood. It is also companioned to plasminogen activator inhibitor-1 (PAI-1), and makes it more stable.

Thus vitronectin also control the proteolysis process which begins by plasminogen activation [3]. Vitronectin effect by numerous pathologic processes in liver involve persistent inflammation with excessive healing, leading at times to cirrhosis [4]. The amino terminal of type III procollagen peptide (sP-III-P) is a peptide produced in the transforming of type III procollagen to type III collagen. It has been consider as an index of collagen amount in the liver. Increased levels occur as a consequence of tissue repair [5]. Chronic liver disease is associated with increased synthesis the components of connective tissue, with significant increase in collagens types I & II. Inactive form from collagen (procollagen) converted to active form by specific proteases which deliver amino- and carboxy-terminal peptides.'

The resulting collagen molecule is then combined to collagen fibrils. In type III collagen, the amino-terminal peptides released by deliver into blood stream but some are rejoined to the surface of collagen fibrils [6]. Aim of the study was that; in order to assess the biochemical markers which measured in the study to diagnose the disease and to determine if it can be used to monitor the development of disease.

Subjects and Methods

Current study was carried out on 60 individuals (patients& healthy). 30 patients (16 males & 14 females) were selected from
outpatient and inpatient clinic of gastroenterology & hepatology center at Baghdad Teaching Hospital. Their ages ranged from 50-67 years. They are classified according to Child-Pugh Score as follows;

Group A: (10) score A in Child-Pugh score, Group B: (10) score B score in Child-Pugh score, Group C: (10) score C in Child-Pugh score.

All patients and healthy subjects were fasting for 12 hours at least.

Measurements

Platelet count, PCV and INR tests were down directly, vitronectin was done by using enzyme linked immune sorbent assay ELISA, (Ray biotech Co., USA), as well as, vitronectin receptor (CLOUD-CLONE CORP, USA) and sP-III-P (Biocompare Co., USA). Biochemical testing included total protein, GOT, GPT, total billirubin and serum albumin were done by spectrophotometer technique (Spinreact Co., Morocco).

Statistical Analysis

Statistical analysis was performed using SAS (Statistical Analysis System - version 9.1) and MedCalc-5 program was used also to estimate sensitivity and specificity and cutoff point using the Receiver Operation Characteristics (ROC curve). To find the significant difference between more than two means, ANOVA analysis was done.

Results

The clinical characteristics of liver cirrhosis patients and healthy subjects summarized in Table 1.

Table1: Clinical characteristics

<table>
<thead>
<tr>
<th>Age (years)</th>
<th>Vitronecin</th>
<th>Vit. receptor</th>
<th>sP-III-P</th>
<th>ALT</th>
<th>ALP</th>
<th>Platelet count</th>
<th>INR</th>
<th>Total protein</th>
<th>Albumin (g/dL)</th>
<th>Bilirubin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Class A</td>
<td>57.70±0.29b</td>
<td>0.17±0.01b</td>
<td>8.78±0.28c</td>
<td>67.90±4.05c</td>
<td>130.30±2.50b</td>
<td>1.56±0.06b</td>
<td>6.93±0.11ab</td>
<td>4.17±0.07b</td>
<td>1.34±0.11c</td>
<td></td>
</tr>
<tr>
<td>Class B</td>
<td>57.80±0.99b</td>
<td>0.15±0.09bc</td>
<td>9.74±0.27b</td>
<td>90.75±3.55b</td>
<td>127.20±3.20b</td>
<td>1.65±0.05b</td>
<td>31.00±1.44a</td>
<td>35.20±1.75a</td>
<td>2.40±0.15b</td>
<td></td>
</tr>
<tr>
<td>Class C</td>
<td>60.80±1.17a</td>
<td>0.14±0.08c</td>
<td>10.63±0.20a</td>
<td>125.10±5.77a</td>
<td>125.40±3.36b</td>
<td>1.63±0.05b</td>
<td>31.00±1.20a</td>
<td>35.96±1.39a</td>
<td>2.89±0.12d</td>
<td></td>
</tr>
</tbody>
</table>

Means with different subscript letters refer to significant difference (P<0.05)

There was a significant difference in the means of serum vitronectin levels in all patients sub-groups when compared to control (P<0.05), as well as there was a significant difference (P<0.05) when compare between patients group (class A, B and C).

Serum vitronectin receptors significantly increase (P<0.05) in class C patients when compare to other patients group and also when compare to control group serum N-terminal procollagen type III peptide (sP-III-P) levels significantly increase in patients class C when compared to control (P<0.05) and to other patients sub-groups (P<0.05).

In class B patients, the levels of (sP-III-P) significantly increase (P<0.05) when compared to class A patients and also when compared to control group. There was a significant increase in (sP-III-P) levels when compared between patients class A and control (P<0.05), as shown in table (1). There were no significant difference (P > 0.05) in both AST & ALT when compared between all patients group and control group. There was a significant difference (P<0.05) between patients class C and both class A and control groups. There was a significant increase in class B group when compared to class A group and control group (P<0.05) but there was no significant difference between class B and C (P > 0.05).

The mean of platelet count significantly decrease in all patients group when compared to control, but there was no significant difference between patients class groups (P>0.05). There was a significant difference in INR when compared between patients class C with class A&B (P<0.05)
and also with control group (P<0.05). There was significant difference between class C when compare to class B in total protein levels.

Both serum albumin and total bilirubin significantly differ in class C group when compare to others (P<0.05), also when compare between class A and b groups with other groups (P<0.05). To distinguish between cirrhosis patients and control by using studied parameters, the ROC analysis was used. It’s used to find the sensitivity and specificity to every parameter. The ROC analysis revealed the descending order (VNR =1.000, sP-III-P=0.936, vitronectin = 0.814) of parameters that showed a significant variation. As shown in the figure VNR was more sensitive parameter than the other.

<table>
<thead>
<tr>
<th>Variable</th>
<th>AUC</th>
<th>SE</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vitronectin</td>
<td>0.814</td>
<td>0.0435</td>
<td>0.718 to 0.888</td>
</tr>
<tr>
<td>VNR</td>
<td>1.000</td>
<td>0.000</td>
<td>0.960 to 1.000</td>
</tr>
<tr>
<td>sP-III-P</td>
<td>0.936</td>
<td>0.0249</td>
<td>0.863 to 0.976</td>
</tr>
</tbody>
</table>

**Receiver Operator Curve (ROC)**

**Analysis for the Investigated Parameters in Patients and Controls**

**Discussion**

The Child–Pugh score (CPS) is applied to evaluate the development of chronic liver disease, mainly cirrhosis. Its depend on the measurement of total bilirubin, μmol/L, serum albumin( g/dL), prothrombin time, as cites and the grade of hepatic encephalopathy. CPS classified the cirrhosis patients into three grades; class A (Less severity), class B (Moderate severity), class C (severe) [7]. This study was design to assess novel parameters to monitor the severity of liver cirrhosis. In the current study, serum vitronectin show significant decrease in cirrhosis patients in all classes when compare to control groups. Vitronectin cling to the extracellular matrix by binding to vitronectin receptors on the cellular surface, thus enabling the cells to spread. It’s also complaining to the thrombin-ant thrombin III complex to inhibit thrombin activity [8]. Naji & Aziz suggested that the severely depressed in vitronectin levels that observed in liver cirrhosis patients levels. The redistribution may be accrue in vitronectin
cannot be excluded in patients with cirrhosis by ascites formation, and by the deposition of the two proteins in fibrotic liver tissue [8]. The depression in the vitronectin levels in chronic liver diseases it may be due to decrement in the size of liver parenchymal cells and the consuming of vitronectin levels due to its precipitation in the extracellular matrix [9]. The present study show that there was significant difference in the integrin vitronectin receptor level in patients when compare to healthy group and its exhibit high sensitivity to development of disease. Vitronectin receptor is a part of the integrin superfamily of adhesion molecules. This molecule has many of general structural and functional properties of integrin.

It changes cell adhesion to extracellular matrix by distinguish the conserved arg-gly-asp (RGD) sequence of several plasma and matrix proteins. Vitronectin receptor is an integrin that has a major role in cell-cell, VNR in serum and found that its concentration was high in liver diseases. In general, Integrin's receptors, which act as mechanoreceptors by transfer the information from cell to cell, and also transfer it from the extracellular matrix (ECM) to the cell interior and vice versa. Since integrin vitronectin receptors bind to ECM ingredients and change its 3D structure according to binding, so its play an important role in development and progression the liver cirrhosis [10].

The current study recorded that; there was a significant increase in the sP-III-P levels in patient's class and the difference was more increase between patient’s classes. Type III procollagen peptide are responsible for synthesis type III procollagen molecule during fibrogenesis. The sP-III-P levels reflects type I collagen metabolism, which is one of many part of fibrosis. In several hepatic diseases, type III collagen was stimulated early in hepatic injury so the plasma sP-III-P increase [11]. Recent study suggest that serum PIIIP was significantly higher in CHC patients with fibrosis than in control subjects [12].

Manolagas SC recorded that fibrotic livers are one of the important sources of type I collagen turnover and it is important to know the relative contribution P-III-P in each tissue. The bone matrix consists of approximately 90% type I collagen, while the remaining 10% is consist of proteoglycans and numerous non-collagenous proteins [13]. The present study conclude that vitronectin, VNR and sP-III-P significantly elevated in all patients classes and the diagnostic value of serum VNR is greater than vitronectin and sP-III-P so it can be act as a marker of liver cirrhosis and also could be used to monitor the severity of disease.

References


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