# Relatioship of Some Biochemical Variance with Hepatitis Patients

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## Abstract

Hepatitis is a viral disease; it has a wide distribution among peoples in different countries' and it has more than five common types named; A, B, C, D and E. The serum level of total protein, albumin and ferritin was estimated in patients with chronic hepatitis B and C, in male and female aged from 19 – 50 years; at the same time same variance was measured in apparently normal healthy volunteers. A patient with other liver disease was excluded from this search.

The average values for total protein, albumin, and ferritin for normal volunteers are (5.33, 4.724 and 51.485 g/ dl) respectively for male and 5.342, 4.733 and 51.667 g/ dl in the female. While its value in HBV was (3.399, 2.878 and 77.909 g/dl) for male and 3.429, 3.092 and 137.091g/dl in female respectively. While in HCV, the value for total protein, albumin and ferritin was (4.045, 3.462 and 151.182 g/dl) respectively in male and 3.838, 3.391 and 68.167 respectively in the female. Using statistical analysis, the results indicate that there is a significant value for all variances when compared with the control in both cases B and C. The p-value was highly significant its value in HBV male patients is 6.4E-19, 5.11E-17 and 1.96E-12 for total protein, albumin and ferritin respectively and 1.6E-18, 2.03E-15 and 3.65E-24 for female. While in type HCV male patients the p- values were 8.01E-10, 1.9E-10 and 5.13E-17 for total protein, albumin and ferritin respectively, in female the values are 1.79E-16, 3.36E-12 and 1.78E-05 respectively. The results indicate also a positive correlation between all of the variance in the two types of patients.

**Keywords**: chronic hepatitis, B, chronic hepatitis, C, total protein, albumin,and ferritin.

## الخلاصة

التهاب الكبد الفايروسي من الامراض واسعة الانتشار في معظم دول العالم , وتعرف له اكثر من خمسة أنواع هي : أي , بي , سي , دي , و أي. تم قياس مستوى البروتينات الكلية , الالبومين والفرتين في امصال المرضى المصابين بالتهاب الكبد الفايروسي الدائم للنوعين سي و بي لكلا الجنسين الرجال والنساء وللفئات العمريه من 19 الى 50 سنه, وبنفس الوقت تم اخذ دم من متطوعين اصحاء لاعتبارهم مجموعة سيطرة. وقد تم اهمال جميع المرضى المصابين بامراض الكبد المختلفه في هذا البحث.

معدل قيم كل من البروتين الكلي , الالبومين والفرتين في دم الاصحاء الذكور كانت بحدود 5.33 , 4.724 , و 51.485 على التوالي. في حين كانت قيم نفس المتغيرات في النساء 5.342 ,4.733 و51.667 على التوالي في حين كانت القيم في مرضى التهاب الكبد الفايروسي نوع بي للرجال بحدود 3.399 ,2.878 و 77.909 على التوالي اما في المرضى النساء فكانت بحدود 3.429 , 3.092 و 137.091 على التوالي . وفي مرضى النوع سي كانت القيم للمرضى الذكور 4.045, 3.462 و 151.182 وللنساء 3.838 , 3.391 و 68.167 لكل من البروتين الكلي والالبومين والفرتين على التوالي.

تم استخدام برنامج الإحصاء الإصدار 18 لتحليل النتائج أوضحت النتائج بان هناك قيم معنويه في جميع المتغيرات التي تم قياسها ولكلا النوعين من الالتهاب الفايروسي بي و سي وكانت القيم المعنويه للنوع بي في الرجال لكلمن البروتين الكلي , الالبومين والفرتينفي النوع بي بحدود ,6.4E−19و 5.11E−17 1.96E−12 وفي النساء 1.6E−18, 2.03E−15 و 3.65E−24 على التوالي. وفي النوع سي للرجال كانت القيم المعنويه 8.01E−10 , 1.9E−10 و 5.13E−17 وللنساء 1.79E−16, 3.36E−12 و 1.78E−05 على التوالي لنفس تسلسل المتغيرات اعلاة. كما لوحظ ان هناك علاقه ترابطيه موجبه بين جميع المتغيرات التي شملتها الدراسه.

**الكلمات مفتاحية:** فرتين , البومين , البروتين الكلي, هبتايتس سي, هبتايتس بي.

## Introduction and literature review:

Hepatitis C virus (HCV) regard as one of the large health care burden in different countries in the world. Nearly, 130–170 million living persons are with chronic HCV infection [Rodalia M**.** *etal* 2014, Global B. 2004]**,** when this disease left without treatment, may develop to cancer or cirrhosis of the liver. In Arabic regain Egyptrecognize as the greatest burden of HCV infection, about 10% prevalence of chronic HCV infection (CHC) among a population with age ranged from 15 to 59 years [El-Zanaty *etal* 2009]. From this percent about 70–80% reported as HCV infections persist and about 30% of individuals weredeveloped to severe liver disease, comprising hepatocellular carcinoma and cirrhosis [David *et al* 2002].Variability in theresponse of virology depends on diverse patient factors as well as histological and virological factors in addition to the interaction between these factors [Feuerstadt *et al* 2010].These factors include age, duration of infection, gender, mode of acquisition, HCV genotype, ethnicity the degree of fibrosis of the liver, viral load [Thio & Thomas DL. 2010],and the degree iron overload in hepatic [Metwally *et al* 2004,Fujita *et al*2007].

One of the most proper marker for liver iron deposition is serum ferritin (SF) for this reason in addition to a relatively low-cost it has been widely used as noninvasive tool to monitor iron status [Tavill, 2001] and can be used as an indirect marker for the estimation of hepatic iron concentration, since hepatic iron controls the production of serum ferritin (SF) [Knovich , *etal 2009*].Many researchers have mentioned that, the increase of serum ferritin concentration in patients with either, type 2 diabetes, or steatosis,and metabolic syndrome [Vari & Forouhi]**,** which are often found in CHC patients [Adinolfi *et al* 2001; Asselah *etal* 2006].

Patients with chronic infection are asymptomatic or have only mild, nonspecific symptoms as long as cirrhosis is not present [Lauer *et al*., 2001; Merican *et al*., 1993]. Patients with the above symptoms suffered from one or more from these frequent complications fatigue, weakness, nausea, myalgia, weight loss, and arthralgia [Lauer *et al*., 2001].The aminotransferase enzyme also can be used for monitoring of chronic hepatitis C and B. In case of patients infected with chronic hepatitis B, there are nearly 400 million cases in the worldwide this number represents nearly10- 20% of the total infected patients and about one million died / year because of chronic hepatitis B viral infection[McMahon *et al*., 2005; Custer *et al*., 2004]. The differences in age at the time of infection, following acute hepatitis B and developed to chronic type proportion inversely with age, about 35% of this infection happened in young years (1-5 years old), while less than ten percent of adult[WHO, 2011]. Viral hepatitis B can be detected in different body fluids and blood such as; saliva, semen, and nasopharyngeal fluids), there are four major modes of transmission of this virus:

- Abnormal sexual practices.
- Transmission from mother to her child during the period of pregnancy and at birth (perinatal).
- Blood transfusion.
- Contact or sharing of infected items through close personal. This mode of transmission is seen mainly in early childhood. In low-endemic areas, the highest incidence of HBV infection is among teenagers and young adults.

The sexual transmission is the most common modes in addition tothe transmission from unsafe injecting practices [Liaw *et al*., 2010]. Many papers claimed that viral

hepatitis B and viral hepatitis I, have common routes of transmission and endemic areas, but viral hepatitis B is about 100 times more infectious[Tarantola *et al*., 2006]. Other papers claimed that about 1/3 of all infected people with viral hepatitis I, have a blood marker of past or present viral hepatitis B infection. At higher rates of viral hepatitis B /viral hepatitis, I happeninmen who have sex with men (MSM) [Thio *et al*., 2002]. The risk of chronic hepatitis B is greater in congenital and causesuppression of the immune system, including HIV infection, and due to usage the suppressant drugs of the immune system or chronic hemodialysis [Wang *et al*., 2010].

30% of the patients with chronic active viral hepatitis B, will progress to liver cirrhosis after about 30 years, in about 1/4 of cirrhotic patients with hepatitis B will be developed to hepatic decompensation over a five-year period; while about 5–10% will develop liver cancer. If the patients were Left untreated, approximately 15% of them will die within 5 years due to cirrhosis[Liang TJ. 2009].The total body stored iron usually reflect by serum ferritin levels since its secretion into circulation is proportional to the amount of cellular iron in the form of cellular ferritin [Finch *et al*., 1986].

## Experimental part:

### Blood samples:

Blood samples were collected from patients with chronic hepatitis type B (28 males and 22 females) and chronic hepatitis type C (30 males and 23 females), with ages between 17 and 50 years old for both males and females. In addition, blood samples were collected from apparently healthy volunteers (20 males and 20 females), from Marjane hospital in IRAQ – Babylon city. After collection, the samples were centrifuged for 15 minutes at 4000 RPM using Hettich – EBA20 centrifuge, serum was collected in clean dried tubes.

***Screening for hepatitis:-***

Serum sample was taken for detection of (HBs –Ag and Anti- HCV antibody) for confirmation of hepatitis infection in the case study and for exclusion of hepatitis infection in the control group by ELISA- Biotech- ELx50 technique, according to the principle and procedure of BIOMERIEUX Hepanostika, Micro Elisa system [Boseind, 2011].

***Single Radial Immunodifusion (SRID):-***

***Principle:-***

Equal volumes of reference sera and sample are added to the wells in agarose gel containing monospecific antiserum, the sample diffuses radially through the gel and the antigen form precipitation ring with the monospecific antiserum. The result can be calculated easily from the table of diameters provided with plates [Biomagreb, 2004].

***Procedure of SRID:-***

1. All Endo plates, reference, and test sera brought to room temperature.
2. 5 microliter of serum and control were dispensed into the wells of the plate by using a suitable pipette (sample should be mixed gently before use).
3. The plate Center covered by the wet cotton to avoid affairs dehydration.
4. The plate was tightly closed and incubated at room temperature ($23^{+/-}2^{`0}$C) on a level surface for equal or more than 48 hours.

5. After incubation, the diameter of each ring measured with 0.1 mm precision using a suitable viewer.
6. The results directly evaluated by using the results of the control samples and a reference table that is provided with RID plates [Biomagreb, 2004].
7.

## Results and discussion

The data collected from patients indicate there are decreasing in total protein concentration in both male and female with respect to that of control groupsas shown in fig -1.

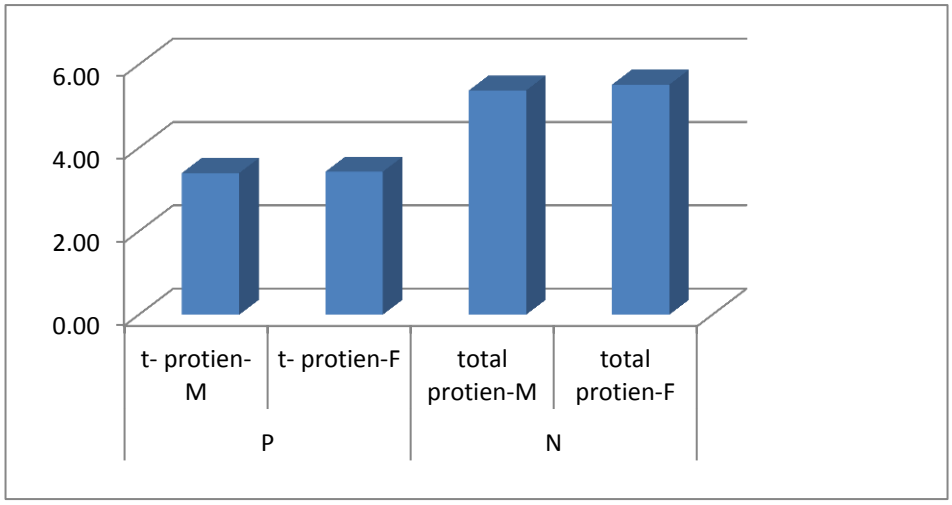


**Fig- 1: relationship between total protein value in normal and patients with chronic HB**

The results also show a decrease in albumin concentration in chronic patients compared with normal volunteers (control group), will there be in increasing value in ferritin concentration in patient group but the increase in themale is greater than in females. This result was in agreeing with many published papers, which indicate there are an increase in ferritin concentration and decreased in protein and albumin level in serum of patients with hepatitis[Wang and Han, 2010].

The statistical study using spss program version 18 to show the correlation between the biochemical variance in chronic hepatitis B, it was shown that there is a high correlation between total protein and albumin (0.8288) wears there is no significant correlation between others. In case of age, the results indicate there is a negative correlation between total protein and albumin with age, while there is a positive correlation with ferritin. As appear in table –1.

**Table-1: Correlation between variance in chronic hepatitis (b) patients**

| | *Total protein* | *albumin* | *ferritin* | *Age* |
|---|---|---|---|---|
| Total protein | 1 | | | |
| albumin | 0.828885 | 1 | | |
| ferritin | 0.254072 | 0.375496 | 1 | |
| Age | -0.1798 | -0.38547 | -0.217302443 | 1 |

In patients with chronic hepatitis C, almost the same results as in chronic B with the exception of the correlation of age with all variance are weakly positive values, as in table – 2.

**Table-2: Correlation between variance in hepatitis (c) patients**

| | *Total protein* | *albumin* | *ferritin* | *Age* |
|---|---|---|---|---|
| Total protein | 1 | | | |
| albumin | 0.68282295 | 1 | | |
| ferritin | 0.09989819 | 0.1345631 | 1 | |
| Age | -0.31540139 | -0.3859691 | -0.184882505 | 1 |

The correlation between the same variance in normal volunteers the data show a high correlation between T- protein and albumin (0.9787), and the low positive correlation between other parameters, as in table -3.

**Table-3: Correlation between variance in normal volunteers**

| | T-protein | albumin | ferritin | age |
|---|---|---|---|---|
| T-protein | 1 | | | |
| Albumin | 0.97876928 | 1 | | |
| Ferritin | 0.00539911 | 0.0436392 | 1 | |
| Age | 0.12873169 | 0.1232478 | 0.419690978 | 1 |

To study the significant(p-value)between the biochemical variables between patients and normal, healthy volunteers the results show there is a highly significant value between all variables in both chronic hepatitis type B &C, with the exception of age the value was not significant with respect to ferritin but in the case of protein and albumin the data reviled a high difference in p- values, in hepatitis –B than in C. As appears in table – 4.

**Table-4: p-value between variance in normal and patient**

| | t-protein | albumin | ferritin | Age |
|---|---|---|---|---|
| HIV-B | 1.11357E-05 | 4.30714E-05 | 2.28816E-06 | 0.0911161 |
| HIV-C | 0.000141556 | 0.000475561 | 8.30635E-06 | 0.079178952 |

To establish the effect of sex on biochemical variance used in this study in patients with chronic hepatitis B, a statistical analysis was done to verify p-value, slandered deviation and average values, tables – 5,6;the sex has highly effective on ferritin, and to a less extent in protein and albumin.

**Table – 5: effect of sex on variance in chronic HBV for male**

| | total protein | | albumin | | ferritin | |
|---|---|---|---|---|---|---|
| | N | P | N | P | N | P |
| average | 5.333 | 3.399 | 4.724 | 2.878 | 51.485 | 77.909 |
| std | 0.696 | 0.459 | 0.721 | 0.562 | 6.704 | 14.390 |
| the p-value | | 6.43E-19 | | 5.11E-17 | | 1.96E-12 |

**Table – 6: effect of sex on variance in chronic HBV for female**

| | total protein | | albumin | | ferritin | |
|---|---|---|---|---|---|---|
| | N | P | N | P | N | P |
| the p-value | | 1.6E-18 | | 2.03E-15 | | 3.65E-24 |
| average | 5.342 | 3.429 | 4.733 | 3.092 | 51.667 | 137.091 |
| std | 0.696 | 0.363 | 0.721 | 0.343 | 6.704 | 20.395 |

In case of patients with chronic hepatitis –C, nearly the results were found as in chronic hepatitis B, highly significant in ferritin compared with other parameters, tables 7,8.

**Table – 7: effect of sex on variance in chronic HCV for male**

| | t- protein | | albumin | | ferritin | |
|---|---|---|---|---|---|---|
| | N | P | N | P | N | P |
| the p-value | | 8.01E-10 | | 1.9E-10 | | 5.13E-17 |
| average | 5.333 | 4.045 | 4.724 | 3.462 | 51.485 | 151.182 |
| std | 0.696 | 0.753 | 0.721 | 0.627 | 6.704 | 36.365 |

**Table -8: effect of sex on variance in chronic HCV female**

| | total protein | | albumin | | ferritin | |
|---|---|---|---|---|---|---|
| | N | P | N | P | N | P |
| the p-value | | 1.79E-16 | | 3.36E-12 | | 1.78E-05 |
| average | 5.342 | 3.838 | 4.733 | 3.391 | 51.667 | 68.167 |
| std | 0.676 | 0.383 | 0.705 | 0.649 | 6.655 | 19.411 |

Since albumin is synthesized in the liver, and low serum albumin may be indicative of liver failure or diseases such as cirrhosis or chronichepatitis. Hypoalbuminemia can also present as part of the nephrotic syndrome, in which protein is lost in the urine due to kidney damage. Low albumin levels can be an indicator of chronic malnutrition or protein-losing enteropathy [Anderson, 2000]. Our results go online with that found by [Sargsyants,2009]. Elevated ferritin level was revealed in 34,3% of patients, and it was significantly higher in men than in women (326,0±38,4 vs 108,2±44,7 ng/dl, p <0,05).

## Conclusion

Chronic hepatitis is a disease with many types, with respect to type B and C have aneffect on liver functions, and this phenomenon will affect the serum level of ferritin increase in male very high than female and decrease total protein and albumin level nearly of the same value in both male and female. Therefore, the development of disease and management of drugs and others can be followed can be followed easily.

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