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Estimation of Serum Interleukin-18 and Hs-CRP in Chronic Hepatitis C Infected Patients at Assuit Governorate, Egypt

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ABSTRACT

Hepatitis C virus (HCV) infection is a global health problem. Egypt has the highest prevalence of hepatitis C virus (HCV) in the world. Interleukin-18 (IL-18) as a critical multipotent inducer of innate and acquired immune responses.

This study aimed to estimate IL-18 and hs-CRP levels in patients at different stages of chronic HCV infection and to evaluate IL-18 as a non-invasive marker of the severity of liver damage in chronic hepatitis C patients, and to investigate the characteristics of hs-CRP and its correlation to chronic HCV infection.

This study included 50 chronic HCV patients and 20 healthy. They were subjected to history taking, liver function tests, real-time PCR test and other laboratory tests. Serum IL-18 levels were assayed by an enzyme-linked immunosorbent assay.

Our study was conducted on 70 subjects with their ages ranging between 9-70 years. 50 HCV chronically infected patients, were 42 (84.0%) males and 8 (16.0%) female. Twenty apparent healthy individuals without HCV infection used as a control, they were 18 (90%) males and 2 (10%) females. Serum IL-18 and Hs-CRP levels were higher in chronic HCV patients compared to healthy. Responders to interferon treatment had higher interleukin-18 levels than non-responders. However, Serum Hs-CRP were higher in non-responders than Responders to interferon treatment. Patients with hepatocellular carcinoma had higher interleukin-18 and Hs-CRP levels than those without hepatocellular carcinoma.

Serum IL-18 could be a significant predictor for the severity of HCV infection and monitoring of therapy response in chronic HCV patients with other tests.

Keywords: Assuit; Interleukin-18; Hs-CRP; Hepatitis C Virus

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Introduction

Hepatitis C virus (HCV) infection is a global health problem, being the second most common chronic viral infection in the world with a global prevalence of about 3% (about 180 million people) [1]. Egypt has the highest prevalence of HCV worldwide (22%) [2] and the highest prevalence of HCV- 4, which are responsible for almost 90% of HCV infections [3]. Patients infected with HCV have different clinical outcomes, ranging from from acute resolving hepatitis to chronic liver disease including liver cirrhosis or hepatocellular carcinoma [4].

Hepatocellular carcinoma (HCC) is a common malignant tumor, with high morbidity and mortality, which affects patients worldwide. Approximately 564,000 novel cases of HCC are diagnosed each year [5,6].

The currently recommended therapy of chronic HCV infection is the combination of pegylated interferon (IFN) alpha and ribavirin for a period 24-72 weeks. Sofosbuvir has become commercially available for the treatment of HCV in the US in late 2013 and several European countries in early 2014 [7].

Interleukin-18 **(**IL-18), previously known as an interferon-gamma-inducing factor, is a pleiotropic pro-inflammatory cytokine that is expressed mainly by peripheral blood mononuclear cells and macrophages. In chronic hepatitis C virus infection, a significant up-regulation of IL-18 occurs in the inflammatory infiltrate, suggesting a role of this cytokine in the chronic cellular immune response to hepatocytes in the course of the disease [8]. Viral infections are known to suppress the immune system; induction of IL-18 binding protein and inhibition of IL-18 [9]. About 20% of chronically infected patients develop cirrhosis with enhanced risk of hepatocellular carcinoma, because of the difficulty in eradicating HCV [10].

High-sensitivity C-reactive protein (Hs-CRP) is a nonspecific inflammatory marker. Like body temperature, it has great clinical utility but is too nonspecific to indicate a particular disease [11]. Hs-CRP plays a major role in the scenario of an activated systemic inflammatory response and is also a common feature of various chronic liver diseases, such as nonalcoholic fatty liver disease and nonalcoholic steatohepatitis [12,13].

This study aimed to determine serum IL-18 and Hs-CRP levels in patients at different stages of chronic HCV infection and to evaluate IL-18 as a non-invasive marker of the severity of liver damage in chronic hepatitis C patients, and to investigate the characteristics of Hs-CRP and its correlation to HCV infection with many factors such as cirrhotic liver patients, hepatocellular carcinoma patients and responders and non-responders patients for IFN therapy.

Materials and methods:

Patients and data collection

This study was carried out during the period from April to December 2014. It included fifty unselected patients proved serologically as having chronic viral hepatitis C. They were admitted to Assiut University Liver Hospital and Dirout Fever hospital. Patients suffering from hepatitis B virus infection or any other cause of viral liver cirrhosis were excluded. A brief history of each case was taken as regard age, sex, chronic diseases, hepatitis C virus infection manifestations, and treatment uptake (interferon, liver supports or without any therapy). Twenty healthy persons from Dirout Town, having normal liver enzymes and free from

viral hepatitis markers were included as a control group.

Collection and processing of blood samples

Blood samples were collected from each subject by venipuncture of the cubital veins. The site was cleaned thoroughly using 70% isopropyl alcohol in water and 1% iodine for one minute and allowed to dry. Taking precautions to avoid contamination of the site, about 5 millilitres of blood was collected using a sterile syringe and needle and dispensed into a clean plastic tube. The blood samples were centrifuged at 4000 rpm for 10 minutes, and the serum obtained was stored at -70°C.

All patients were subjected to history taking, laboratory investigations including; HCV antibodies by ELISA. The other aliquot was stored at –70 ^{o}C till sera were subjected to ELISA technique for the quantitative determination of IL-18 and immunofluorescence technique for the quantitative determination of Hs-CRP.

Quantitative determination of IL-18 by ELISA technique:

Quantification of IL-18 levels in the sera of all studied subjects was performed as described by the manufacture with a commercially available Enzyme-linked Immunosorbent Assay Kit for IL-18 (Boster Biological Technology CO., Ltd 3942 B Valley Ave, CA, 94566, USA). Briefly, the assay uses two monoclonal antibodies against two different epitopes of human IL-18. In the wells coated with anti-human IL-18 monoclonal antibody, samples to be measured or standards were incubated. After washing, a peroxidase-conjugated anti-human IL-18 monoclonal antibody was added to the microwell and incubated. After another washing, the peroxidase substrate was mixed with the chromogen and allowed to incubate for an additional period. An acid solution was then added to each well to terminate the enzyme reaction and to stabilize the developed colour. The optical density (OD) of each well was then measured at 450 nm using a microplate reader. The concentration of serum IL-18 was calibrated from a dose-response curve based on reference standards.

Quantitative determination of Hs-CRP by immunofluorescence technique:

Hs-CRP was performed by kit supplied by i-CHROMA TM hs-CRP Boditech Med Inc. lot No. 100513- S, based on fluorescence immune assay technology. The CHROMA TM hs-CRP uses sandwich immune detection method by mixing the serum sample with detector buffer in a test vial, the fluorescence-labelled detector anti-CRP antibody in the buffer binds to CRP antigen on a blood specimen

Statistical analysis:

The data were analyzed using the statistical package SPSS (version 12.0, SPSS Inc., Chicago, IL). Statistical analysis was performed using a t-test, analysis of variance (ANOVA). Results are expressed as means ± SD or frequency. A p-value of less than 0.005 was considered to be significant.

3. Results:

Our study was conducted on 70 subjects with their ages ranging between 9-70 years. They were classified into two groups: **Group I**: Included 50 HCV chronically infected Egyptian patients from Assuit governorate, they were 42 (84.0%) males and 8 (16.0%) female mean aged 51.50±9.74 (mean ± SD) years. **Group II:** Included 20 healthy individuals without HCV infection, they were 18 (90%) males and 2 (10%) females mean aged 38.65±16.27 (mean ± SD) years **(Table 1&2).**

The highest distribution of the patients with liver supports treatment (73.7%) at the age group from 49 to 70 years and the lowest distribution of the patients without treatment (7.9%) at the same age group. The highest distribution of the cases with liver supports treatment (55.6%) at the age group from 29 to 48 years and the lowest distribution of the patients with interferon treatment (11.1%) at the same age group, While the lowest distribution of the cases with liver supports treatment (33.3%) at the age group from 9 to 28 years and the highest distribution of the cases without treatment (66.7%) at the same age group was recorded (Table 3).

The highest distribution of male patients with liver supports treatment (61.9%) as comparing the distribution of the male responder and non-responder patients to interferon treatment (4.8 % for each one). The highest distribution was recorded in male patients compared to a female of all groups **(Table 4).**

According to liver disease and response to treat patients, we investigated the following parameter and they're related to IL-18 and Hs-CRP levels: chronic hepatitis C patients compared with healthy, cirrhotic and non-cirrhotic patients, hepatocellular and non-hepatocellular carcinoma (HCC) patients, responders and non-responders to interferon treatment and patients with interferon treatment compared to non-treatment patients.

The means of IL-18 and Hs-CRP levels were higher among chronic hepatitis C patients than healthy (P = 0.173 & P = 0.407 respectively) **(Table 5).**

The means of IL-18 levels were higher among the non-cirrhotic than cirrhotic patients (P = 0.15), while the means of Hs-CRP levels were higher among the cirrhotic than the non-cirrhotic patients (P = 0.08) (Table 6).

The means of IL-18 and Hs-CRP levels were higher among the hepatocellular carcinoma than non-hepatocellular carcinoma patients **(Table 7).**

The means of IL-18 levels were higher among the responders than non-responders patients, while Hs-CRP levels were higher among the non-responders than responders patients. Also, the means of IL-18 levels were higher among the liver supports treatment patients and non-treatment patients than IFN treatment patients, while the means of Hs-CRP levels were higher among the liver supports treatment patients than IFN treatment and non-treatment patients **(Table 8).**

Also the means of liver function tests levels were higher among the non responders patients [ALT, AST and ALP] than among the responders patients. However, this was no statistically significant. [ALT (P = 0.66), AST (P = 0.36), ALP (P = 0.59), Total bilirubin (P = 0.01), Direct bilirubin (P = 0.02) and Indirect bilirubin (P = 0.01) **(Table 9).**

Discussion:

Interleukin (IL)-18, the IFN-γ-inducing cytokine, plays a critical role in the Th1 response [14], and administration of antibodies to IL-18 has been shown to prevent liver damage in an animal model [15]. IL-18 is synthesized by different cell types, including Kupffer cells, activated macrophages, monocytes and dendritic cells. The importance of IL-18 in immunity and host defence is only beginning to be appreciated. IL-18 may also induce macrophages to produce tumour

necrosis factor (TNF)-α and nitric oxide (NO) and account for the induction of cell death [16].

C-reactive protein (CRP), which is at the same time a major acute-phase protein, is produced by the liver; it may be a mediator of tissue damage and is a factor that activates the complement system. In humans, plasma levels of and/or fibrogenic activity, and markedly, as much as 1000-fold or more, after an acute inflammatory stimulus, largely reflecting an increased synthesis by hepatocytes [17,18].CRP is a nonspecific inflammatory marker. Like body temperature, it has great clinical utility but is too nonspecific to indicate a particular disease [11].

The present study showed that serum levels of IL-18 and hs- CRP were increased in chronic HCV patients than in healthy controls. Our results agreement with Heba et al., (2009) and Nadia *et al*., (2013) who found elevated plasma levels of IL-18 in HCV patients compared to healthy subjects [19,20]. Huang *et al*., (2009) who demonstrated that chronic hepatitis C patients had a higher hs-CRP level than healthy controls, were in agreement with the present study [21].

However, Schvoerer *et al*., (2003) reported lower levels of IL-18 in plasma and supernatants of stimulated peripheral blood mononuclear cells from patients with genotype 1 HCV infection than in those from normal controls [22]. Stevens *et al*. (2002) and Nascimento *et al*., (2005) showed that CRP levels were significantly higher in HCV negative patients compared to HCV positive patients [23,24].

Also, the present study showed that serum levels of IL-18 were increased in non-cirrhotic chronic HCV than in cirrhotic chronic HCV patients, however, serum levels of hs-CRP were increased in cirrhotic chronic HCV than in non cirrhotic chronic HCV patients. This study is in agreement with Nascimento et al., (2005) who found that a significant difference in the hs-CRP ratio in HCV positive patients might indicate that hepatocellular injury could affect CRP production in HCV positive patients [24]. This finding conflicts with studies of Ludwiczek et al. (2002) and Heba et al., (2009) who found that disease progression from non-cirrhotic to cirrhotic disease was accompanied by an increase in plasma IL-18 levels [19,25].

The present study showed that serum levels of IL-18 and hs-CRP were increased in HCC chronic HCV patients than in non-HCC chronic HCV patients. The current results were consistence with Abiru *et al*., (2002), Nascimento *et al*., (2005), and Tangkijvanich *et al*. (2007) who found that serum IL-18 and hs-CRP in patients with HCC were significantly elevated compared with those of the controls [22,24,26].

The present study showed that serum levels of IL-18 were increased in responders to IFN treatment than in non-responders patients. Also, found that serum levels of IL-18 were increased in liver supports treatment and non-treatment patients than in IFN treatment patients. However, serum levels of Hs-CRP were increased in non-responders to IFN treatment than in responders patients, while, the serum levels of Hs-CRP were increased in liver supports treatment than in IFN treatment and non-treatment patients. These findings are in agreement with Yoneda *et al*., (2011) who reported that cytokines IL-10, IL-12p40, and IL-18 all decreased during treatment and remained low in patients with an SVR [24]. Keeffe, (2005) demonstrated that serum hs-CRP significantly decreased after pegylated interferon and ribavirin combination therapy [27]. On the

other side, these findings are in disagreement with Nadia *et al*., (2013) who found higher baseline interleukin-18 levels in non-responders (513.9±119.4 pg/ ml) than responders (471.7±192.1 pg/ml) [20].

These data confirm previous studies of enhanced serum IL-18 in HCV patients and suggest its marked increase in chronic hepatitis C patients compared to HCV infected patients and controls. So our data suggest that IL-18 can be used as a non-invasive marker for detection of the chronicity and severity of liver inflammation in chronic hepatitis C.

Data illustrated in Table 9, showed that the various laboratory and clinical parameters were analyzed when compared between chronic HCV patients with liver supports treatment and chronic HCV patients without Interferon (IFN) treatment; the present study showed that a non-significant increased in patients with liver supports treatment than in patients with IFN treatment in their AST levels (p =0.36), ALT levels (p=0.66) and ALP levels (p =0.59).

However, the present study showed that a non-significant increased in patients with IFN treatment than in patients with liver supports treatment in their total bilirubin levels (p=0.01), direct bilirubin levels (p=0.02) and indirect bilirubin levels (p=0.01). Also, the present study showed that a non-significant increased in non-responders to IFN treatment patients than in responders patients in their AST levels (p =0.36), ALT levels (p=0.66) and ALP levels (p =0.59). These findings were in agreement with Nadia *et al*., (2013) who found ALT and AST levels were higher among non-responders than responders and it didn't reach statistically significance [20]. Persistently normal ALT is usually defined as ALT levels in the normal range during a 6-to-12 month period [28, 29] This definition found that stage and grade of liver disease were lower among patients with persistently normal serum ALT [30].

Conclusions

IL-18 is a non-invasive marker of the severity of liver damage in chronic hepatitis C patients. IL-18 is an important factor for the evaluation of HCV treatment by interferon regimens. Hs-CRP is a confirmed marker of the severity of liver damage in chronic hepatitis C patients and the evaluation of HCV treatment by interferon regimens.

Recommendations

1- Further studies using a large number of Egyptian patients with HCV-related chronic liver disease are required to confirm the association of IL-18 with the response to interferon therapy in HCV.

2- These investigations could help to identify patients with markedly increased risks of disease progression and could guide the design of individualized treatment strategies for chronic hepatitis C.

3- Identifying other predictors, especially host genetic factors, for treatment outcome in HCV patients may help in making appropriate treatment decisions.

Abbreviations: **ELISA**: Enzyme-Linked Immunosorbent Assay; **HCV**: Hepatitis C Virus; **Hs-CRP**: High-sensitivity C-reactive protein; **HCC**: Hepatocellular carcinoma; **IL-18**: Interleukin-18; **IFN**: interferon; **NO**: Nitric Oxide; **TNF**: Tumour Necrosis Factor; **Th**: T helper cells.

References

1.Zeuzem, S., Berg, T., Moeller, B., Hinrichsen, H. and Mauss S.(2009) Expert

opinion on the treatment of patients with chronic hepatitis C.J. Viral Hepat.; 16(2):75-90.

2. Smith, DB1., Bukh, J., Kuiken, C., Muerhoff, A.S., Rice, C.M., Stapleton, J.T. and Simmonds, P. (2014) Expanded classification of hepatitis C virus into 7 genotypes and 67 subtypes: updated criteria and genotype assignment web resource. Hepatology;59(1):318-27.

3.Faisal, N., Sidra S.H., Ghulam, M. and Shumaila, N. (2015) An overview on hepatitis C virus genotypes and its control. Egyptian Journal of Medical Human Genetics, 16 (4): 291–298.

4. Lauer, G.M. and Walker, B.D. (2001) Hepatitis C virus infection. New Engl J Med; 345:41–52.

5. El-Serag, H.B. (2012) Epidemiology of viral hepatitis and hepatocellular carcinoma. Gastroenterology. 142:1264–1273.

6. Luk, J.M. and Liu, A.M. (2011) Proteomics of hepatocellular carcinoma in Chinese patients. OMICS. ;15:261–266.

7. **Pawlotsky, J.M. (2013)** Hepatitis C virus: standard-of-care treatment. Adv Pharmacol; 67:169-215.

8. Susan, L.S. (2001) Interleukin 18: Tipping the Balance towards a T Helper Cell 1 Response. J. Exp. Med. 194(3): F11–F14.

9. Kaser, D., Novick, M., Rubeinstein, B., Siegmund, B., Enrich, R. and Koch, W. (2002) Interferon-α induces interleukin-18 binding protein in chronic hepatitis C patients. Clin Exp Immunol., 129(2):332–338.

10. Zahida, B. and Charles, S. B. (2007) Adult systemic cat scratch disease associated with therapy for hepatitis C. BMC Infect Dis. 7: 8.

11-Pai, C.K., Shu-Chu, S.H. and Ta-Jen, W. (2006) Review: Serum C - reactive protein as a Marker for Wellness Assessment. Annals of Clinical & Laboratory Science, 36(2): 163- 169.

12-Riquelme, A., Arrese, M., Soza, A., Morales, A., Baudrand, R., Perez-Ayuso, R.M., Gonzalez, R., Alvarez, M., Hernandez, V., Garcia-Zattera, M.J., Otarola, F., Medina, B., Rigotti, A., Miquel, J.F., Marshall, G. and Nervi, F. (2009) Non-alcoholic fatty liver disease and its association with obesity, insulin resistance and increased serum levels of C-reactive protein in Hispanics. Liver Int; 29:82–88.

13.Kogiso, T., Moriyoshi, Y., Shimizu, S., Nagahara, H. and Shiratori, K. (2009) High-sensitivity C-reactive protein as a serum predictor of nonalcoholic fatty liver disease based on the Akaike Information Criterion scoring system in the general Japanese population. J Gastroenterol; 44:313–321.

14. Dinarello, C.A (1999) IL-18: a Th1-inducing, proinflammatory cytokine and new member of the IL-1 family. J Allergy Clin Immunol., 103:11–24.

15. Okamura, H., Tsutsui, H., Komatsu, T. et al (1995), Cloning of a new cytokine that induces IFN-γ production by T cells. Nature, 378:88–91.

16. Tsutsui, H., Matsui, K., Kawada, N., Hyodo, Y., Hayashi, N., Okamura, H., Higashino, K. and Nakanishi, K. (1997) IL-18 accounts for both TNF-alpha- and Fas ligand-mediated hepatotoxic pathways in endotoxin-induced liver injury in mice. J Immunol., 159:3961–7.

17. Ablij, H.C. and Meinders, A.E. (2002) C-reactive protein: history and revival. Eur J Int Med, 13(4): 412-22.

18.Volanakis, J.E. (2001) Human C-reactive protein: expression, structure and function. Mol Immunol, 38(2): 189-97.

19. Selim, H.S.1., El-Barrawy, M.A., Taha, H.A. and Abd El-Hafiz, D.A. (2009) Evaluation of Interleukin-18 as a Non-Invasive Marker of Liver Fibrosis among Chronic Hepatitis C Virus Patients. J Egypt Public Health Assoc.;84(5-6):391-403.

20-Mohran, Z.Y., Ali-Eldin, F.A. and Abdel Aal, H.A. (2011) Serum interleukin-18: does it have a role in the diagnosis of hepatitis C virus-related hepatocellular carcinoma? Arab J Gastroenterol.;12(1):29-33.

21. Huang, J.F., Chuang, W.L., Yu, M.L., Yu, S.H., Huang, C.F., Huang, C.I., Yeh, M.L., Hsieh, M.H., Yang, J.F., Lin, Z.Y., Chen, S.C., Dai, C.Y. and Chang, W.Y. (2009) Hepatitis C virus infection and metabolic syndrome—a community-based study in an endemic area of Taiwan. Kaohsiung J Med Sci; 25:299–305.

22-Schvoerer, E., Navas, M.C., Thumann, C., Fuchs, A., Meyer, N., Habersetzer, F. and Stoll-Keller, F. (2003) Production of inter-leukin-18 and interleukin-12 in patients suffering from chronic hepatitis C virus infection before antiviral therapy. J Med Virol;70:588-593.

23- Lin, Z.Y., Chuang, W.L., Dai, C.Y., Yu, M.L., Chen, S.C., Hsieh, M.Y., Wang, L.Y., Tsai, J.F. and Chang, WY (2002) Clinical application of serum C-reactive protein measurement in the detection of bacterial infection in patients with liver cirrhosis. Kaohsiung Journal of Medical Sciences, 18: 121-126.

24-Nascimento, M.M., Bruchfeld, A., Suliman, M.E., Hayashi, S.Y., Pecoits-Filho, R., Manfro, R.C., Pachaly, M.A., Renner1, L., Stenvinkel, P., Riella1, M.C. and Lindholm, B. (2005) Effect of hepatitis C serology on C-reactive protein in a cohort of Brazilian hemodialysis patients. Brazilian Journal of Medical and Biological Research; 38: 783-788.

25-Ludwiczek, O., Kaser, A. and Novick, D. (2002) Plasma levels of interleukin-18 and interleukin-18 binding protein are elevated in patients with chronic liver disease. J Clin Immunol.; 22:331–337.

26-Abiru, S., Kato, Y., Hamasaki, K., Nakao, K., Nakata, K. and Eguchi, K. (2002) Spontaneous regression of hepatocellular carcinoma associated with elevated levels of interleukin 18. Am J Gastroenterol; 97:774–5.

27. Keeffe, E.B., (2005) Chronic hepatitis C: management of treatment failures. Clin Gastroenterol Hepatol; 3:S102–S105.

28. Nutt A.K., Hassan H.A., Lindsey J., Lamps L.W. and Raufman J.P. (2000): Liver biopsy in the evaluation of patients with chronic hepatitis C who have repeatedly normal or near-normal serum alanine aminotransferase levels. Am J Med; 109:62-4.

29. Strader D.B., Wright T., Thomas D.L., Seef L.B. and AASLD (2004): Diagnosis, management and treatment of hepatitis C. Hepatol; 39:1147-71.

30. Taraneh D., Nasser E.D., Mohammad B., Ali H., Babak H., Ali S. and Madjid S.H. (2005): Relationship between serum alanine aminotransferase levels and liver histology in chronic hepatitis C-infected patients. Indian J Gastroenterol; 24:49-51.

Table (1): Distribution of Patients with HCV and apparent healthy as regard to age groups

Age groups (years)	Studied groups					
	Patients with HCV (N = 50)		Healthy (controls) (N = 20)		Total	
	N	%	N	%	N	%
9-28	3	42.9	4	57.1	7	100
29-48	9	50.0	9	50.0	18	100
49-70	38	84.4	7	15.6	45	100
Mean (SD)	51.50 ± 9.74		38.65 ± 16.27		70	100

$X^2 = 10.59$ P = 0.005

Table (2): Distribution of Patients with HCV and apparent healthy as regards to gender

Gender	Studied groups					
	Patients with HCV (N = 50)		Healthy (controls) (N = 20)		Total	
	N	%	N	%	N	%
Male	42	70.0	18	30.0	60	100
Female	8	80.0	2	20.0	10	100
Total	50	100	20	100	70	100

P = 0.713

Table (3): Distribution of the patients as regard to therapy concerning age groups

Age groups (years)	Therapy											
	Liver Supports treatment patients (N = 34)		Non-Responders patients (N = 2)		Responders Patient (N = 2)		Patients With interferon treatment (N = 4)		Non-Treatment patients (N = 8)		Total	
	N	%	N	%	N	%	N	%	N	%	N	%
9-28	1	33.3	0	0.0	0	0.0	0	0.0	2	66.7	3	100
29-48	5	55.6	0	0.0	0	0.0	1	11.1	3	33.3	9	100
49-70	28	73.7	2	5.3	2	5.3	3	7.9	3	7.9	38	100

$X^2 = 10.529$ p = 0.225

Table (4): Distribution of the patients as regard to therapy concerning gender

Gender	Therapy											
	Liver Supports treatment patients (N = 34)		Non-Responders patients (N = 2)		Responders Patient (N = 2)		Patients With interferon treatment (N = 4)		Non-Treatment patients (N = 8)		Total	
	N	%	N	%	N	%	N	%	N	%	N	%
Male	26	61.9	2	4.8	2	4.8	4	9.5	8	19.0	42	100
Female	8	100	0	0.0	0	0.0	0	0.0	0	0.0	8	100

X^2= 4.482 **P = 0.344**

Table (5): Levels of IL-18 and Hs-CRP among Chronic Hepatitis C patients and Healthy controls

Variables	Studied groups		Significant	
	Patients with HCV (N = 50)	Healthy (controls) (N = 20)	T-value	P-value
IL-18 (pg/ml)	194.40±75.05	175.46±75.33	-0.953	0.173
Hs-CRP (mg/I)	2.05±3.02	1.87±2.64	-0.233	0.407

Table (6): Levels of IL-18 and Hs-CRP among Cirrhotic and Non-cirrhotic Chronic Hepatitis C patients

Variables	Patients with HCV		Significant	
	Cirrhotic cases (N = 22)	Non-cirrhotic cases (N =28)	T-value	P-value
IL-18 (pg/ml)	182.29±63.60	203.92±82.84	-1.04	0.15
Hs-CRP (mg/I)	2.51±3.76	1.37± 1.04	-1.381	0.08

Table (7): Levels of IL-18 and Hs-CRP among Hepatocellular carcinoma (HCC) and Non-Hepatocellular Carcinoma patients

Variables	Patients with HCV		Significant	
	Cases having HCC (N = 2)	Cases having no HCC (N =48)	T-value	P-value
IL-18 (pg/ml)	376.86±7.96	186.80±66.30	-17.119	0.000*
Hs-CRP (mg/l)	3.25±0.21	1.82±2.68	-3.45	0.0009**

* Significant ** High Significant

Table (8): Levels of IL-18 and Hs-CRP among Chronic Hepatitis C patients according to therapy modalities

Variables	Patients with HCV					Significant	
	Liver supports treatment patients (N = 34)	**Non-Responders patients (N = 2)**	**Responders patient (N = 2)**	**Patients with interferon treatment (N = 4)**	**Non-treatment patients (N = 8)**	F-value	P-value
	Mean ± SD	**Mean ± SD**	**Mean ± SD**	**Mean ± SD**	**Mean ± SD**		
IL-18 (pg/ml)	201.12 ± 80.004	163.22 ± 59.503	234.55 ± 136.95	177.82 ± 53.092	171.92 ± 56.34	0.504	0.73
Hs-CRP (mg/l)	2.127 ± 3.079	3.075 ± 3.273	1.030 ± 0.042	1.440 ± 1.327	0.946 ± 0.273	0.48	0.74

Table (9): Distribution of the studied cases as regard to means of liver function tests according to therapy modalities

Variables	Therapy					Significant	
	Liver supports treatment patients (N = 34)	Non-Responders patients (N = 2)	Responders patient (N = 2)	Patients with interferon treatment (N = 4)	Non-treatment patients (N = 8)	F-value	P-value
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD		
ALT (< 40 U/L)	55.59 ± 40.03	41.50 ± 20.506	40.00 ± 21.213	37.75 ± 7.632	35.63 ± 13.049	0.74	0.66
AST (< 37 U/L)	65.38 ± 40.098	45.00 ± 28.284	38.50 ± 24.749	44.75 ± 21.884	42.38 ± 11.363	1.11	0.36
ALP (up to 240 U/L)	205.24 ± 94.479	189.50 ± 127.986	151.50 ± 65.761	142.75 ± 88.289	169.38 ± 63.058	0.70	0.59
Total bilirubin	1.155 ± 0.481	1.000 ± 0.000	0.900 ± 0.141	3.975 ± 5.382	1.025 ± 0.399	3.56	0.01
Direct bilirubin	0.355 ± 0.214	0.200 ± 0.000	0.200 ± 0.141	1.175 ± 1.693	0.237 ± 0.130	3.10	0.02
Indirect bilirubin	0.800 ± 0.2994	0.800 ± 0.000	0.700 ± 0.000	2.800 ± 3.689	0.787 ± 0.274	3.765	0.01