

## A Study of the Impact of Treatment Response to Direct Acting Antivirals on Serum Lipid Profile in Egyptian Patients with Chronic HCV Infection

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### Abstract

**Background:** Hepatitis C virus infection is a major etiological factor for liver cirrhosis, steatosis, and hepatocellular carcinoma. HCV is known to be closely related and associated with host and lipid metabolism.

**Aim of Study:** This study aimed at studying the changes in serum lipid profile with INF-free treatment regimens in chronic HCV infection.

**Patients and Methods:** This prospective observational cohort study was carried out on 301 chronic HCV-infected patients, who had fulfilled the inclusion & exclusion criteria and treated according to the National Committee of Control of Viral Hepatitis (NCCVH) Treatment Protocol, updated in December 2016, and completed their course of treatment and follow-up. They were divided into two groups. Group I included 264 patients, who achieved SVR and Group II included 37 patients who did not achieve SVR. Patients underwent additional investigations in the form of 12h fasting serum lipids profile (mg/dl) including: Serum cholesterol, serum triglycerides, Low Density Lipoprotein (LDL), High Density Lipoprotein (HDL). These tests were done before starting treatment (baseline), at the end of treatment.

**Results:** There was a statistically significant difference between responder and non-responder groups as regard baseline cholesterol ( $p=0.001$ ), LDL ( $p=0.001$ ), HDL ( $p=0.026$ ) and triglycerides levels ( $p=0.016$ ). These lipid parameters increased significantly over the course of the therapy and continued to be increased after treatment discontinuation in responders; while they showed non-significant changes in non-responders.

**Conclusions:** Viral eradication was associated with increased serum cholesterol, TG & LDL levels, which continued to be elevated after discontinuation of therapy.

**Key Words:** Serum lipid profile – Chronic HCV infection patients.

### Introduction

HCV infection is one of the leading causes of liver cirrhosis, Hepatocellular Carcinoma (HCC), and

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liver transplantation in Egypt. Hence, patients with HCV infection are in need for effective antiviral therapy to prevent the progression to these complications and reduce mortality [1].

Recently, a revolution has occurred with the availability of Direct-Acting Antivirals (DAAs) in the treatment of HCV, with different modes of action leading to high chance of cure and a good tolerance [2].

HCV infection is known to modify the host lipid metabolism; the virus uses host lipids in every step of its replication cycle from attachment and entry into hepatocytes, to replication and assembly of new viral particles [3].

The close relationship between serum level of LDL Cholesterol (LDL-C) and the chance of achieving SVR has been reported in patients under interferon based regimens [4].

Although higher SVR rates were achieved with DAAs treatment of HCV, special attention must be paid to unexpected adverse events based on host metabolic changes [5].

Eradication of HCV with DAAs increases total cholesterol levels, due to the removal of the effect of HCV on cholesterol metabolism, because these regimens have no influence on the nutritional status of the host [6].

Although DAAs rarely fail to achieve viral eradication, a subset of patients experience treatment failure [7]. So, the changes in host lipid metabolism may be helpful for prediction of treatment response.

## Patients and Methods

This cohort prospective study was performed at the Viral Hepatitis Treatment Unit in Tanta University Teaching Hospital from Jan. 2017-Jan. 2018 and included 301 patients with chronic HCV who started treatment with interferon free regimen and completed the course and follow-up period (according to the protocol of national committee for treatment of viral hepatitis), then they are further divided into 2 groups; Group I which included 264 patients who have achieved SVR and Group II which included 37 patients who did not achieve SVR.

**Exclusion criteria:** Patients with hepatitis B infection or other forms of chronic liver disease and patients on lipid lowering medications.

The demographic data, gender, and age were collected at initiation of therapy. HCV antibody & quantitative polymerase chain reaction for HCV RNA. Complete blood count, serum albumin, total and direct serum bilirubin, alanine transaminase, aspartate transaminase, international normalized ratio, serum creatinine, fasting blood glucose level, Hepatitis B surface antigen, Alpha fetoprotein and pregnancy test for females in the child-bearing period were included in the basic laboratory data.

Patients underwent further investigations in the form of 12h fasting serum lipid profile including: Serum cholesterol, triglycerides, Low Density Lipoprotein (LDL) and High Density Lipoprotein (HDL) detected by commercial kits (bio systems). These tests were done before starting treatment (baseline) and at the end of treatment (EOT).

**Statistical analysis:** Statistical presentation and analysis of our study was conducted, by IBM SPSS statistics® Version 22. *p*-value <0.05 was considered statistically significant.

## Results

A total of 301 chronic HCV-infected patients started interferon-free treatment with a combination of DAAs and RBV and were conducted on two groups according to achievement of SVR.

- Group I included 264 patients who have achieved SVR.
- Group II included 37 patients who did not achieve SVR.

As regards the demographic and baseline characteristics, all tested parameters showed non-significant differences between the two studied groups as shown in (Table 1).

Table (1): The demographic and baseline characteristics in the studied groups.

Variables	Group I SVR (n=264)		Group II Non-SVR (n=37)		$\chi^2$	<i>P</i>
	N	%	N	%		
<b>Age in years:</b>					<i>t</i> =	
• Range					0.834	0.405
<b>Sex:</b>						
• Male	107	89.2	13	10.8	0.394	0.530
• Female	157	86.7	24	13.3		
<b>Treatment status:</b>						
• Naïve	251	87.2	37	12.8	FE	0.3 81
• Experienced (PegINF + RBV)	13	100.0	0	0.0		
<b>Treatment regimen:</b>						
• SOF + DAC + RBV	159	89.3	19	10.7	1.058	0.304
• SOF + DAC	105	85.4	18	14.6		

$\chi^2$ : Chi square. FE: Fisher exact test. *t*: Student *t*-test.

As regards baseline laboratory data between studied groups, all tested parameters showed non-significant differences between the two studied groups, except for the AFP. It was  $11.30 \pm 21.16$  (ng/mL) in Group I, while it was  $17.28 \pm 21.01$  (ng/mL) in Group II with significant difference between both groups (*p*=0.001) as shown in (Table 2).

Table (2): Baseline laboratory data between studied groups.

Variables	Group I SVR (n=264)	Group II Non-SVR (n=37)	<i>t</i> *	<i>P</i>
	Mean ± SD	Mean ± SD		
• HCV RNA (X10 <sup>6</sup> )	1.108±1.821	1.531±2.156	1.278	0.202
• <b>CBC:</b>				
Hemoglobin (mg/dl)	12.79±1.52	13.021±1.46	1.584	0.114
WBC count (X10 <sup>3</sup> )	5.96±1.89	5.66±1.63	0.898	0.370
Platelet count (X10 <sup>3</sup> )	176.2±67.9	159.4±60.1	1.435	0.152
• Serum albumin (g/dl)	4.02±0.49	3.89±0.49	1.491	0.137
• Total bilirubin (mg/dl)	0.87±0.40	0.94±0.46	1.089	0.277
• International normalized ratio	1.13±0.13	1.14±0.17	0.572	0.268
• Alpha fetoproteins (ng/mL)	21.16±11.30	21.01±17.28	3.894	0.001*
• ALT (IU/L)	51.68±33.96	46.84±30.42	0.821	0.412
• AST (IU/L)	52.31±34.24	51.34±31.27	0.163	0.870
• Serum creatinine (mg/dl)	0.77±0.18	0.81±0.20	1.031	0.303
• Fasting blood sugar (mg/dl)	109.41±48.27	106.48±40.20	0.404	0.687
• Glycosylated hemoglobin (%)	7.05±1.09	7.03±1.14	0.049	0.687

- *t*-Student *t*-test or Man Whitney were used according to data distribution.

\*: Statistically significant at *p*≤0.05.

Comparison of changes in serum lipid levels between responders to antivirals Vs. relapsers: All tested parameters showed significant differences between both groups before treatment and at the end of treatment, as shown in (Table 3).

Table (3): Changes in serum lipid profile levels between studied groups.

	Group I SVR (n=264)	Group II Non-SVR (n=37)	t	p
<i>Serum cholesterol (mg/dl):</i>				
Before treatment (baseline)	120.99±27.21	87.14±22.41	7.231	0.001*
End of treatment (EOT)	200.94±28.31	92.65±14.12	37.316	0.001*
<i>Serum triglycerides (mg/dl):</i>				
Before treatment (baseline)	75.21±27.83	63.73±20.36	2.418	0.016*
End of treatment (EOT)	156.26±39.84	68.70±18.89	22.126	0.001*
<i>Serum high density lipoproteins (mg/dl):</i>				
Before treatment (baseline)	53.64±16.40	46.92±21.69	2.238	0.026*
End of treatment (EOT)	53.36±15.65	44.97±19.93	2.457	0.018*
<i>Serum low density lipoproteins (mg/dl):</i>				
Before treatment (baseline)	52.16±25.93	27.35±11.25	10.157	0.001*
End of treatment (EOT)	116.08±29.57	33.78±15.22	26.598	0.001*

t-Student t-test or Man Whitney were used according to data distribution.

\*: Statistically significant at p≤0.05.

*Changes in serum lipid levels in responders:*

Serum lipid parameters in GI showed highly significant increase in all tested parameters, except for serum HDL-C levels which showed non-significant changes between the baseline levels and at the end of treatment as shown in (Table 4).

*Changes of serum lipid in non-responders:*

Serum lipid parameters in Group II showed non-significant difference between the baseline levels and at the end of treatment as shown in (Table 5).

Table (4): Serum lipid parameters at baseline and at the end of treatment in G I.

	SVR		t	p
	Baseline	EOT		
<i>Cholesterol:</i>				
Mean ± SD	120.99±27.207	200.938±28.306	-44.792	<0.001*
<i>TG:</i>				
Mean ± SD	75.206±27.833	156.261 ±39.838	-33.354	<0.001*
<i>HDL:</i>				
Mean ± SD	53.644±16.396	53.362±15.649	0.730	0.466
<i>LDL:</i>				
Mean ± SD	52.159±25.929	116.080±29.568	-36.173	<0.001*

t : Student t-test.

\*: Statistically significant at p≤0.05.

Table (5): Serum lipid parameters at baseline and at the end of treatment in G II.

	Non SVR		Paired Samples Test	
	Baseline	EOT	t	p
<i>Cholesterol:</i>				
Mean ± SD	87.135±22.412	92.649±14.117	-1.956	0.058
<i>TG:</i>				
Mean ± SD	63.730±20.364	68.703±18.894	-1.938	0.061
<i>HDL:</i>				
Mean ± SD	46.919±21.686	44.973±19.926	1.904	0.065
<i>LDL:</i>				
Mean ± SD	27.351±11.248	33.784±15.221	-1.962	0.058

**Discussion**

Egypt has a very high burden of infection and disease: Egypt ranks the 5 th amongst all countries for the burden of disease from viral hepatitis C [8]. The availability of DAAs with excellent efficacy and good tolerability profiles offer a unique opportunity to achieve HCV elimination worldwide. However, viral clearance with DAAs has led to a global worsening of lipid profile.

In this study, we investigated the changes in serum lipid profile with INF-free treatment regimens in Egyptian patients with chronic HCV infection, its association with the treatment outcome.

Regarding patients' gender, sixty percent of patients were females and 40% were males. The female predominance in our study differs with El-Serag, [9] who reported male predominance, which was attributed to reluctance of the Egyptian rural female population to participate in national health programs. However, the female predominance in our study might be due an increased awareness about viral hepatitis through a nationwide screening campaigns that encourage female population to participate and receive treatment.

As regard previous treatment status, 4% of treated patients were treatment-experienced (PegINF + RBV); all of them (100%) have achieved SVR. This is concordant with Wyles et al., [10] who reported SVR 98% of patients previously treated with an interferon-based regimen on SOF plus DAC + RBV, but differs with Nelson et al., [11] who reported SVR 86% in patients using SOF plus DAC + RBV regimen. The difference in findings across these studies might be due to different number of treatment-experienced patients included in our study and theirs and different genotypes.

In the current study, 264 patients (88%) have achieved SVR. Our results are in agreement with Nelson et al., [11] who stated that a 12-week regimen of SOF plus DAC + RBV was associated with an overall SVR rate of 89% [11]. However, Fontaine et al., [12] reported SVR 100% in treatment of genotype 4 HCV using the same regimen. The difference might be attributed to higher number of patients included in our study (301 vs. 47).

In this study, the mean HCV RNA level in Group I was  $1.108 \pm 1.821$  ( $\times 10^6$ ) IU/mL, while in Group II was  $1.531 \pm 2.156$  ( $\times 10^6$ ) IU/mL with no significant difference between both groups. This is in accordance with Nelson et al., [11] who reported that there is no link between baseline HCV viral load and SVR, but differs with Kowdley et al., [13] who found that a very high baseline viral level (>6 million IU/ml) was shown to be associated with reduced likelihood of SVR. The difference might be due to different genotype, treatment regimen and duration of therapy (12w vs. 8w) between our study and theirs.

Regarding serum alpha fetoproteins in our study, its level ranged from 0.3-237 with mean  $21.16 \pm 11.30$  (ng/mL) in Group I, while it ranged from 1-104 with mean  $21.01 \pm 17.28$  (ng/mL) in Group II with a significant difference between both groups. This was similarly reported by Akuta et al., [14], who found that high level of AFP is a pretreatment predictor of poor virological response to therapy.

We proposed a real-life study with analysis of the changes in lipid profile. In our study, there was a statistically significant difference between responder and non-responder group as regard pretreatment cholesterol, LDL, HDL and triglycerides levels. We found that these baseline lipid parameters were higher in Group I compared to Group II as regard LDL-C level, total cholesterol level, and serum triglycerides. This is in agreement with the findings of Ogawa et al., [15] who found that the higher the baseline lipid levels, the greater the chance of curing hepatitis C.

We demonstrated in our study that viral clearance due to DAAs led to significant rise from the baseline levels as regard total cholesterol, triglycerides and LDL-C. We also observed that Total Cholesterol (TC), Triglycerides (TG) and LDL-C significantly increased gradually over the course of the therapy, which continued after treatment discontinuation. No meaningful increase in HDL-C level in responders was found. These findings are consistent with Hashimoto et al., [16] and Mauss et al., [17] who reported that the increase in total

cholesterol and LDL but not in HDL levels following SVR with DAAs therapy thought to reflect the role of the intrahepatic cholesterol biosynthetic pathway in HCV replication cycle. Also, Hashimoto et al., [16] suggested that DAAs in addition to have an anti-HCV effect, they themselves might have pharmacological effects on serum lipid levels, especially on LDL-C [16].

Hashimoto et al., 2016 suggested that the rapid suppression of HCV core protein by DAA might result in a decrease of lipid droplet production in HCV-infected liver cells, leading to a massive rebound of circulating LDL [16].

In our study, we noticed that in patients who failed to achieve treatment response, there were non-significant changes in the all tested lipid profile at baseline, and after discontinuation of treatment. Our results were in agreement with Meissner et al., [18] who stated that mean serum LDL was lower in patients who relapsed versus achieved SVR at baseline and post-treatment.

This study has some limitations. Other parameters that might alter the results, such as a change of body weight, dietary modifications, and the amount of exercise were not available.

#### *Conclusion:*

In this study, we could conclude that eradication of HCV with DAAs was associated with increased level of LDL, TC and TG levels.

#### *Conflict of interest:*

No conflicts of interest declared.

#### *Acknowledgments:*

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## دراسة تأثير الإستجابة للعلاج بمضادات الفيروسات المباشرة على مستوى الدهون بالدم فى المرضى المصريين المصابين بالالتهاب الكبدى سى المزمن

تعتبر مصر من بين البلدان ذات أعلى معدلات إنتشار العدوى بالفيروس الكبدى (سى) فى العالم، حيث يعتبر النمط الجينى ٤ هو النمط الوراثى السائد فى مصر. وقد أظهرت دراسات متعددة وجود صلة وثيقة بين فيروس (سى) والتمثيل الغذائى للدهون. ولذلك، القضاء على الفيروس نتيجة إستخدام مضادات الفيروسات المباشرة يؤدى إلى تدهور فى مستوى الدهون بالدم.

الهدف من الدراسة: دراسة التغيرات فى مستوى الدهون فى الدم مع نظم العلاج الخالية من الإنترفيرون فى المرضى المصريين المصابين بالالتهاب الكبدى (سى) المزمن وإرتباطه بنتائج العلاج.

طرق البحث: أجريت هذه الدراسة فى وحدة علاج إلتهاب الكبد الفيروسات بمستشفى طنطا التعليمى الجامعى. وقد شملت الدراسة ٣٠١ مريض مصاب بعدوى إلتهاب الكبد سى المزمن، الذين تم علاجهم وفقاً لبروتوكول العلاج القومى لعلاج إلتهاب الكبدى الفيروسى (سى) المزمن فى ذلك الوقت، والذين قد أكملوا دورة العلاج والمتابعة وقد تم تقسيمهم إلى مجموعتين وفقاً لإستجابة العلاج.

النتائج: فيما يتعلق بالخصائص الديموغرافية الأساسية للمرضى وتحليل ما قبل العلاج، أظهرت النتائج عدم وجود فرق إحصائى بين المجموعتين بإستثناء دلالات أورام الكبد (ألفا فيتو بروتين). وقد أظهرت النتائج أن القضاء على الفيروس مرتبط بزيادة ملحوظة فى مستويات الكوليسترول، الدهون الثلاثية والدهون منخفضة الكثافة بالدم عند نهاية العلاج. وقد تبين عدم وجود فرق إحصائى فى مستوى الدهون بالدم فى المرضى الذين لم يحققوا إستجابة للعلاج.

الإستنتاج: إن نظم العلاج بإستخدام سوفوسبوفير بالإضافة إلى دكلاتزفير مع أو بدون ريبافيرين لمدة ١٢ إسبوع، كان لها نتائج إيجابية فى علاج المرضى المصريين المصابين بعدوى فيروس (سى) من النمط الجينى ٤. وقد إرتبط القضاء على الفيروس بزيادة مستويات الكوليسترول، الدهون الثلاثية، والدهون منخفضة الكثافة بالدم.