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**Effects of nigella sativa on clinical outcome of hepatitis C virus Egyptian patients**

Barakat EMF *et al.* A pilot study in HCV patients

Eman Mahmoud Fathy Barakat, Lamia Mohamed El Wakeel, Radwa Samir Hagag

**Eman Mahmoud Fathy Barakat,** Department of Tropical Medicine, Faculty of Medicine, Ain Shams University, Cairo 11566, Egypt

**Lamia Mohamed El Wakeel,** Department of Clinical Pharmacy, Faculty of Pharmacy, Ain Shams University, Cairo 11566, Egypt

**Radwa Samir Hagag,** Department of Clinical Pharmacy, Faculty of Pharmacy, Egyptian Russian University, Cairo 16686, Egypt

**Author contributions:** Barakat EMF contributed to study concept and design, acquisition of data, interpretation of data, critical revision of the manuscript for important intellectual content; El Wakeel LM contributed to acquisition of data, analysis and interpretation of data, critical revision of the manuscript for important intellectual content; Hagag RS contributed to data collection, material support; all of the authors contributed to drafting of the manuscript and final approval for version to be published.

**Correspondence to: Lamia Mohamed El Wakeel,** **PhD, Associate Professor** of Clinical Pharmacy, Faculty of Pharmacy, Ain Shams University, 4 street 292, New Maadi, Cairo 11566, Egypt. lamywak @yahoo.com

**Telephone:** +20-100-5201099 **Fax:** +20-100-5201099

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**Abstract**

**AIM:** To evaluate the safety, efficacy and tolerability of Nigella Sativa in the management of hepatitis C virus (HCV) patients non-eligible for interferon alpha (IFNα) therapy.

**METHODS:** Thirty HCV patients,who were non-eligible for IFN/ribavirin therapy, fulfilled the inclusion criteria and were included in the present study. Inclusion criteria included; all patients with HCV with or without cirrhosis, who either had a contraindication to IFNα therapy, have refused or had a financial constraint to IFNα therapy. Exclusion criteria included; patients on IFNα therapy, infection with hepatitis B virus or hepatitis I virus, hepatocellular carcinoma, other malignancies, major severe illness or non-compliance to treatment. Various parameters including; clinical parameters, complete blood count, liver functions, renal functions, plasma glucose, total antioxidant capacity and polymerase chain reaction, were all assessed at baseline and at the end of the study. Clinical assessment included; hepato and/or splenomegaly, jaundice, palmar erythema, flapping tremors, spider naevi, lower limb edema, and ascites. Nigella Sativa was administered for 3 successive months at a dose of (450 mg 3 times daily). Clinical responses, and adverse drug reactions’ incidence were assessed initially, periodically and at the end of the study.

**RESULTS:** Nigella Sativa administration significantly improved HCV patients’ viral load (380 808.7 ± 610 937 *vs* 147 028.2 ± 475 225.6, *P =* 0.001) and their total antioxidant capacity (1.35 ± 0.5 *vs* 1.612 ± 0.56, *P =* 0.001). After Nigella Sativa administration the following laboratory parameters improved; total protein (7.1 ± 0.7 *vs* 7.5 ± 0.8, *P =* 0.001), albumin (3.5 ± 0.87 *vs* 3.69 ± 0.91, *P* = 0.008), red blood cells count (4.13 ± 0.9 *vs* 4.3 ± 0.9, *P* = 0.001) and platelet count (167.7 ± 91.2 *vs* 198.5 ± 103, *P* = 0.004). Fasting blood glucose (104.03 ± 43.42 *vs* 92.1 ± 31.34, *P* = 0.001) and postprandial blood glucose (143.67 ± 72.56 *vs* 112.1 ± 42.9, *P* = 0.001) were significantly decreased in both diabetic and non-diabetic HCV patients. Patients with lower limb edema decreased significantly from baseline *vs* after treatment [53.30% (*n* = 16) *vs* 23.30% (*n* = 7), *P* = 0.004]. Adverse drug reactions were unremarkable except for few cases of epigastric pain and hypoglycemia that did not affect patient compliance.

**CONCLUSION:** Nigella Sativa administration in HCV patients was tolerable, safe, decreased HCV patients’ viral load, improved oxidative stress and clinical condition and glycemic control in diabetic patients.

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**Key words**: Hepatitis C virus; Nigella Sativa; Oxidative stress; Viral load

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**INTRODUCTION**

Egypt has the highest prevalence of hepatitis C virus (HCV) worldwide (15%) and the highest prevalence of HCV-4 (67%) with a predominance of subtype 4a (55%)[[1-4](#_ENREF_1)].

The natural history of HCV infection and disease progression are influenced by several factors such as age at infection onset, sex, duration of infection, co-infection with hepatitis B virus (HBV), the level of HCV viraemia and its genotype[[5](#_ENREF_5)].

HCV is an important etiologic factor for the development of hepatocellular carcinoma (HCC) as 23% of HCV patients develop HCC[[6](#_ENREF_6)]. It has been shown that there is an alarming increase in the incidence of HCC in HCV Egyptian patients[[7](#_ENREF_7)].

Presently, the only approved therapy for HCV are Pegylated interferon alpha (PEG-IFNα) and ribavirin treatment, and their success is heavily influenced by patients’ adherence to treatment, which correlates directly to the tolerance to their side effects[[8](#_ENREF_8)]. Moreover, the financial constraints for the combined therapy in many patients often contribute to their non-adherence to therapy, potentially lowering its success rates[[9](#_ENREF_9)].

Oxidative stress-related molecules may act as mediators modulating cellular events responsible for the progression to liver fibrosis[[10](#_ENREF_10),[11](#_ENREF_11)].It has been shown that an increased production of reactive oxygen species, in part catalyzed by iron overloading, is involved in HCV related liver damage through a pathway that involves DNA oxidative injury[[12](#_ENREF_12)] .

Silymarin is one of the alternative therapies that has been previously tested for the management of HCV patients who are not candidates for PEG-IFN; however, it did not show any appreciable effects on viral load[[13](#_ENREF_13)].

Nigella Sativa, which is used as a food condiment in the Middle East, its seeds/oil have shown to possess anti-inflammatory, antiviral and antineoplastic activity in various *in vitro* and *in vivo* studies[[14](#_ENREF_14)]. Nigella Sativa’s antioxidant effects have been shown in the essential oil obtained from six different extracts of Nigella seeds as well as from a commercial fixed oil[[15](#_ENREF_15)]. The crude Nigella Sativa oil and its fractions have shown potent *in vitro* radical scavenging activity[[16](#_ENREF_16)].

The effect of Nigella Sativahas been evaluated in animal studies. There are many reports on its biological activities including; immunpotentiation, anti-tumour, anti-inflammatory, analgesic, antihypertensive, anti-diabetic, respiratory stimulation, anti-bacterial, antifungal, anticestode and antinematode effects[[17-19](#_ENREF_17)].

A striking reduction of Murine cytomegalovirus virus titer in both spleen and liver was found in Nigella Sativa seed oil treated-mice compared with control mice[[20](#_ENREF_20)]. Moreover, oral feeding with Nigella Sativa extract suppressed chemically induced hepatic tumors in rats[[21](#_ENREF_21)].

Nigella Sativa treatment has been shown to ameliorate disturbed hematological parameters in diabetic rabbits through a modulation of lipid peroxide red blood cells (RBCs) membrane content leading to an increase in RBCs count[[22](#_ENREF_22)].

To date, no studies have addressed the use of Nigella Sativa in HCV patients and its potential benefits; hence we sought to evaluate the efficacy, safety, and tolerability of Nigella Sativa supplementation as an alternative therapy in the management of HCV patients who are non-candidates for IFNα therapy.

**MATERIALS AND METHODS**

This was a prospective, single-armed, self-controlled pilot study, conducted at the Tropical Medicine Department, El-Demerdash hospital, Ain Shams University, Cairo, Egypt.

***Patients***

All HCV patients presenting to the department were assessed for eligibility. Inclusion criteria included all patients diagnosed with HCV with or without cirrhosis who either had a contraindication to IFNα therapy[[23](#_ENREF_23)], have refused or had a financial constraint to IFNα therapy.

Exclusion criteria included; patients on IFNα therapy, infection with HBV or hepatitis I virus, hepatocellular carcinoma, other malignancies, major severe illness as; renal failure, congestive heart failure, respiratory failure or autoimmune disease or non-compliance to treatment. An informed consent was obtained from all patients, and the institutional ethical committee approved the study protocol, which conformed with the ethical guidelines of the 1975 Declaration of Helsinki.

***Methods***

Hepatitis markers were performed for all patients at enrolment, including: HB core IgG, hepatitis B surface antigen and HCV antibody. All eligible patients were subjected to the following at enrollment and after 3 months of therapy:

(1) Full clinical assessment stressing on; hepato and/or splenomegaly, jaundice, palmar erythema, flapping tremors, spider naevi, lower limb edema, and ascites; (2) Abdominal ultrasonography; (3)Laboratory investigations which included; a Complete blood count (CBC), liver functions [(aspartic transaminase (AST), alanine aminotransferase (ALT), total proteins, albumin, total and direct bilirubin, prothrombin time and international standard ratio (INR)], renal functions (S.Cr, BUN), serum alpha-fetoprotein, polymerase chain reaction (PCR) for HCV (lower detection limit; < 50 copies) and total antioxidant capacity (TAC); (4) The antioxidants assessed in the estimation of TAC included enzymes such as superoxide dismutase, catalase, glutathione peroxidase; macromolecules such as albumin, ceruloplasmin, ferritin; small molecules, including ascorbic acid, α-tocopherol, β-carotene, reduced glutathione, uric acid, and bilirubin; (5) The assay principle depended on the determination of the antioxidative capacity by the reaction of antioxidants in the sample with a defined amount of exogenously provide hydrogen peroxide (H2O2) The antioxidants in the sample eliminated a certain amount of the provided hydrogen peroxide. The residual H2O2 was determined colorimetrically by an enzymatic reaction which involved the conversion of 3,5,dichloro –2– hydroxy benzensulphonate to a colored product; (6) TAC was analyzed using TAC kit from Bio-diagnostic and measured spectrophotometrically using KENZA (Biolabo) analyzer; and (7) Real time PCR was done on COBAS TaqMan 48® PCR analyzer, using Roche COBAS Ampliprep Taqman Kit.

***Drug administration***

After performing the baseline evaluation, all patients received one capsule of Nigella Sativa seed oil 450 mg available as soft gelatin capsules (Baraka® Pharco pharmaceuticals) three times daily after meals continuously for 3 mo. Patients were followed up every 2 wk throughout the study period for assessing treatment adherence, tolerability and incidence of adverse reactions.

***Statistical analysis***

Statistical analysis was performed using Statistical Package for Social Sciences (SPSS) (vs. 17) software program. Numerical data were summarized using means and standard deviations or medians and ranges. Categorical data were summarized as percentages. Differences between numerical variables over 2 time measurements were tested using paired *t*-test or medians test for non-normally distributed data. Repeated measures analysis of variance was used to test differences between 3-time numerical normally distributed variables and Friedman test was used for non-normally distributed variables. McNemar’s test was used to compare categorical data overtime. All *P* values were two-sided, *P* values < 0.05 were considered significant. All authors had access to study data, reviewed and approved the final manuscript.

**RESULTS**

Thirty patients (16 males, 14 females) with a mean age of (47 ± 10.2 years), fulfilled the inclusion criteria and were enrolled in the study.Four of those patients (13.33%) were diabetics, while 26 (86.67%) were non-diabetics. Fifteen patients (30%) had chronic liver disease, 5 (16.7%) had compensated cirrhosis and 10 (33.3 %) patients had decompensated cirrhosis.

Patients’ clinical assessment data before and after treatment are represented in (Table 1). As shown in Table 1, after treatment, there was a significant decrease in the percentage of patients having lower limb edema while no change was observed in the percentage of patients with jaundice, palmar erythema, spider naevi or ascites. Laboratory parameters before and after treatment are represented in (Table 2).

***Liver functions tests***

After 3 mo of Nigella Sativa treatment, the mean HCV RNA levels (PCR) (147 028.2 ± 475 225.6) significantly decreased relative to their baseline levels (380 808.7± 610 937, *P* = 0.001), (Figure 1A). Table 3 represents the PCR responses after the 3 mo of treatment in patients with chronic liver disease, compensated and decompensated cirrhosis. Figure 2 presents individual patients’ HCV RNA (PCR) values before and after treatment. Table 4 represents the Child-Pugh score and PCR response at baseline and after 3 mo in patients with compensated and decompensated cirrhosis.

All cirrhotic patients (compensated and decompensated) showed either no change or an improvement in their Child-Pugh score, patients presented with variable Child-Pugh score, yet the proportions’ numbers were small for a valid statistical test.

Moreover, a significant increase in total protein and albumin levels was shown after treatment relative to their corresponding levels prior to treatment. However, there was no significant change in liver enzymes (AST, ALT), bilirubin, or INR. Renal functions didn’t show significant change from baseline. The total antioxidant capacity’s levels showed a significant increase after treatment (1.612 ± 0.56) relative to their baseline values (1.35 ± 0.05, *P* = 0.001, Figure 1B).

Hematologic functions varied significantly after 3 mo of Nigella Sativa treatment. There was a significant increase in RBCs (*P* = 0.001) and platelets (*P* = 0.004) counts and a significant decrease (*P* = 0.013) in white blood cells count.

***Blood glucose***

There was a significant decrease in both fasting and postprandial blood glucose after treatment relative to their corresponding levels prior to treatment (*P* = 0.001).

***Incidence of side effects and drug interactions***

The reported side effects throughout the study period were; gastritis in one patient (3.33%) and hypoglycemia in 5 patients (16.76 %); of whom 2 were diabetic on insulin therapy, while the other 3 patients had advanced liver cirrhosis with possible glycogen depletion. Both side effects were treated and did not hinder completion of therapy. The only reported drug interaction was the reported hypoglycemia due to the concurrent use of insulin together with Nigella which aggravated its hypoglycemic effects.

**DISCUSSION**

The main findings of our study is that administration of Nigella Sativa significantly decreased HCV viral load, increased total anti-oxidant activity, total protein and albumin levels, lowered blood glucose levels and improved patients’ lower limb edema as compared to their prior to treatment levels.

Nigella Sativa’s anti-inflammatory, antiviral and antineoplastic activity have been previously documented in various *in vitro* and *in vivo* studies[[14](#_ENREF_14)].

In the current study, Nigella Sativa administration resulted in a significant decrease in patients’ viral loads with 16.67% of patients becoming sero-negative, and 50% showing a significant decrease in the quantitative viral count, of those 66.7 % were cirrhotic patients and 33.3 % were chronic liver disease patients, implying an evident antiviral activity. In the current study, patients with compensated and decompensated cirrhosis, either improved or were maintained on their baseline clinical condition and viral load and none of them deteriorated, which can signify the potential beneficial effects of Nigella administration as reflected by improvement in HCV RNA responses and clinical condition reflected in patients’ Child Pugh class. Although the sub-category of cirrhotic patients was not large enough to detect significance, we recommend larger studies to be conducted in cirrhotic patients to confirm the potential beneficial effects offered by Nigella, which might improve patients’ overall outcome. To our knowledge, this is the first human study that evaluates Nigella’s effects on viral loads in HCV patients. Our findings of improved viral load, could be explained by the findings of a previous animal study of Murine CMV infected mice[[20](#_ENREF_20)], that showed a significant increase in macrophages and CD4+ T cells with a significant decrease in viral titer and an increased serum IFN-γ levels in Nigella treated animals versus control[[24](#_ENREF_24)].

Oxidative stress related molecules have shown to modulate cellular events responsible for the progression of liver fibrosis[[10](#_ENREF_10),[11](#_ENREF_11)]. Moreover, HCV related fibrosis, cirrhosis and liver failure were found to be the result of an adaptive immune response to HCV infected cells[[25](#_ENREF_25)] which is mediated by an induction of endoplasmic reticulum and oxidative stress and the down-regulation of anti-apoptotic proteins NF-kB and Bcl-xl in infected hepatocytes[[26](#_ENREF_26)].

In our study, Nigella administration significantly increased the total antioxidant capacity of HCV patients, implying the potential protective effect of Nigella by halting oxidative stress that contributes to the disease progression. Furthermore it may be tempting to propose that increasing the antioxidant capacity with its cytoprotective role contributed to decreasing the viral loads.

The antioxidant effects of Nigella Sativa have been previously elaborated in animal models of liver ischemia, where it improved the antioxidant capacity and reduced oxidative stress[[27](#_ENREF_27)]. Moreover, Nigella increased hepatic glutathione and reduced elevated hepatic serum enzymes in carbon tetra chloride treated mice, ameliorating its hepatotoxic potential[[15](#_ENREF_15),[28](#_ENREF_28)].

A proportion of patients with acute and chronic liver disease develop diabetes mellitus[29-[30](#_ENREF_30)].HCV infection may also contribute to the development of diabetes where diabetes was observed in 21% of HCV-infected patients[[31](#_ENREF_31)] and glucose intolerance was observed in patients with HCV infection compared with controls suffering liver diseases[[32-35](#_ENREF_32)].

Insulin resistance is one of the pathological features in patients with HCV which may be associated with life threatening complications, making HCV-associated insulin resistance a therapeutic target at any stage of HCV infection[[36](#_ENREF_36)].

Our study has shown that Nigella treatment significantly decreased blood glucose levels in HCV patients implying that Nigella might offer a potential modulatory effect on HCV induced glucose intolerance. This effect was beneficial in the control of diabetic HCV patients as it allowed lowering Insulin requirements. Similar results have been previously shown in a prior study of diabetic patients, where the administration of Nigella (2 gm/d) caused significant reductions in fasting blood glucose and two hour postprandial blood glucose, and HbA1c and improved insulin resistance[[37](#_ENREF_37)].

HCV infection itself can induce autoimmune hemolytic anemia, leukopenia, and thrombocytopenia, even in the absence of IFN-α treatment[[38-42](#_ENREF_38)]. Hematopoietic growth factors modulating these complications have shown beneficial role in HCV patients[[43](#_ENREF_43)].

Nigella therapy in our study has significantly improved HCV patients’ RBCs and platelets counts relative to their prior treatment counts, indicating a potential amelioration/prevention of HCV induced hematologic disorders, hence may positively affect HCV patients’ clinical outcome.

Nigella’s ability to improve hematologic indices has been also reported in prior animal studies where it increased both the packed cell volume and hemoglobin in treated rats[[18](#_ENREF_18)]as well as increased RBCs count in treated diabetic rabbits[[44](#_ENREF_44)]. The increased RBC count was attributed to lowering the RBC’s membrane lipid peroxide level leading to a decreased susceptibility of RBC hemolysis.

Serum albumin is the most abundant plasma protein[[45](#_ENREF_45)] and is essential for maintaining oncotic pressure of the vascular system[[46](#_ENREF_46)]. Chronic HCV patients may suffer a decrease in serum albumin level[[47](#_ENREF_47)] where improvement in hypoalbuminaemia has been shown to improve prognosis[[48](#_ENREF_48)] and quality of life[[49](#_ENREF_49)]. Concentrations of < 30 g/L were associated with 85% chance of liver-related complications at five years and a three-year mortality of 70%[[50](#_ENREF_50)] and was predictive of morbidity and mortality for patients with liver cirrhosis[[51](#_ENREF_51),[52](#_ENREF_52)].

In the current study, Nigella administration significantly increased serum albumin levels and significantly reduced lower limb edema indicating an improvement in patients’ clinical condition. Prior animal studies have shown similar effects in rats[[53](#_ENREF_53)] and in broiler chicks[[54](#_ENREF_54)] in a dose dependent manner[[55](#_ENREF_55)].

Nigella Sativa is used in Arab folk medicine as a diuretic plant[[56](#_ENREF_56)], the mechanism that can also contribute to its efficacy in decreasing lower limb edema, and its resolution in many patients.

In our study, although, the number of patients with ascites decreased after treatment, yet the change was not significant, and the change in ascites severity could not be totally denied, as the degree of ascites was not assessed sonographically after therapy. We hence recommend assessment of ascites incidence and severity in future studies to confirm these results.

The safety and tolerabilityof Nigella Sativa has been previously documented in various clinical trials[[57-60](#_ENREF_57)]. However, to date, clinical studies addressing Nigella Sativa efficacy, safety and tolerability in HCV patients are lacking. Our study has shown that Nigella Sativa administration was tolerable to all patients, and the only side effects reported were; one patient with epigastric pain that was controlled with antacids and five patients complained of hypoglycemia two of whom were diabetic and were receiving concomitant insulin and the hypoglycemia did not recur after decreasing the insulin dose. Of note, the dose of Nigella Sativa used in the current study was (1.35 gm/d) slightly lower than other studies; 2 gm/d used by Bamosa *et al*[[37](#_ENREF_37)] as this dose was the available strengths in the Egyptian market that was the closest to the doses previously used. On the other hand, although Nigella in such patients had significantly positive effects on many parameters, perhaps higher doses or longer durations of therapy may accentuate such appreciable effects. Further studies are needed to confirm such findings.

It can hence be concluded that Nigella sativa administration can have a potential beneficial effect on HCV disease progression and outcome through its prominent antiviral, antioxidant, immunomodulatory effects and can minimize HCV related hematological complications.

Study Limitations. This is the first clinical study to be performed in HCV patients and larger studies are required to confirm the results of the current study. We did not assess all patients for the amount of ascites after therapy sonographically, as such favorable effect of nigella was not anticipated. Hence, in view of significant improvement of serum albumin, this effect of Nigella on the amount of ascites needs further studies. Liver biopsy was not performed, as included patients were either non-eligible or refused the procedure.

In conclusion, Nigella Sativa administration in HCV patients, is safe and tolerable and results in a significant improvement in patients’ viral load, oxidative stress and laboratory markers. Moreover, the clinical improvement and better glycemic control in diabetic patients indicate a potential role for Nigella Sativa in improving the clinical outcome of HCV patients.

Further larger controlled multicenter randomized studies for longer periods for evaluation of Nigella sativa potential beneficial role in HCV patients with and without the concurrent IFN therapy are recommended.

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**COMMENTS**

***Background***

Hepatitis C virus (HCV) is an important etiologic factor for the development of hepatocellular carcinoma. Pegylated interferon-α (PEG-IFNα) and ribavirin treatment are the only currently approved therapy for HCV with variable response rate, and a success that is heavily influenced by patients’ response rate, adherence to treatment, tolerance to side effects. Moreover, the financial constraints for the combined therapy in many patients often contribute to their non-adherence to therapy, potentially lowering its success rates. Nigella Sativa,a food condiment used in the Middle East, have shown to possess anti-inflammatory, antiviral, antioxidant and anticancer activity in various *in vitro* and *in vivo* studies. Till today, no studies have addressed the use of Nigella Sativa in HCV patients and its potential benefits on HCV patients.

***Research frontiers***

Nigella Sativa a natural food supplement, has shown beneficial antioxidant, antiviral, anticancer and immunopotentiating properties in various *in vitro* and *in vivo* studies, but HCV studies are lacking. In exploring the potential role of Nigella Sativa on improving HCV patients’ clinical outcome, the research hot spot is the beneficial effects that Nigella Sativa administration had on reducing patients’ HCV viral load, improving their antioxidant capacity, alleviating their hematologic parameters and improving their blood glucose control especially the diabetics. All of which could have a potential beneficial effect on HCV patients’ responses and amelioration of HCV related complications.

***Innovations and breakthroughs***

No prior clinical trials in HCV patients have evaluated the use of Nigella Sativa and its potential beneficial roles. No studies have addressed any alternative treatments to IFN non-eligible patients or those who refuse or can’t tolerate IFN therapy. Nigella Sativa treatment offered potential hope for a safe tolerable alternative to those patients who can’t tolerate or have a contraindication to IFN use. Moreover, it showed a potential beneficial improvement in patients’ clinical outcome. It showed a preliminary alarming improvement in patients’ viral load and body antioxidant levels that could enlighten the dark path to HCV potential cure. Nigella Sativa administration also improved HCV patients’ hematologic profile, total protein and albumin levels, that contribute to HCV induced-complications. Moreover, the administration decreased blood glucose levels, and hence decreased diabetic patients’ insulin requirements.

***Applications***

The study results suggest that Nigella Sativa is a potentially beneficial, safe and tolerable alternative in IFN non-eligible HCV patients. It can improve patients’ clinical outcome, ameliorate HCV induced hematologic and diabetic complications and can improve patients’ lower limb edema.

***Terminology***

Viral load, also known as viral burden or viral titer, is a measure of the severity of a [viral](http://en.wikipedia.org/wiki/Virus) infection, and can be calculated by estimating the [amount of virus](http://en.wikipedia.org/wiki/Virus_quantification) in an involved body fluid, *e.g.*, [RNA](http://en.wikipedia.org/wiki/Ribonucleic_acid) copies/ml of [blood plasma](http://en.wikipedia.org/wiki/Blood_plasma). Human serum albumin is the most abundant [protein](http://en.wikipedia.org/wiki/Protein) in [human](http://en.wikipedia.org/wiki/Human) [blood plasma](http://en.wikipedia.org/wiki/Blood_plasma). It is produced in the [liver](http://en.wikipedia.org/wiki/Liver) and constitutes about half of the blood serum protein. It transports hormones, fatty acids, and other compounds, buffers pH, and maintains [osmotic pressure](http://en.wikipedia.org/wiki/Osmotic_pressure), among other functions.Total antioxidant capacity measures collectively the amount of antioxidant components of the body that reflects the body’s capacity to combat oxidative stress.

***Peer review***

This is an interesting study in which the authors have treated HCV patients with Nigella Sativa a food condiment used in the Middle East.

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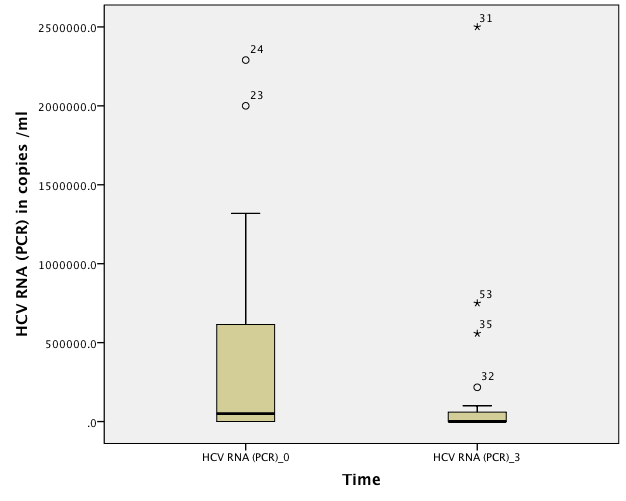
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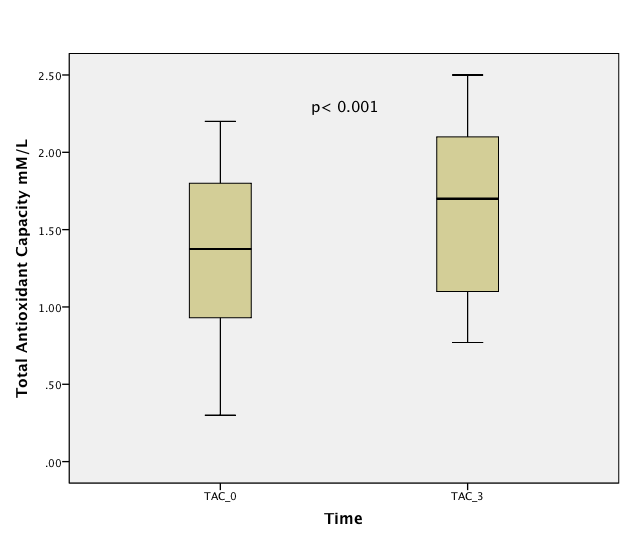
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**P-Reviewers** Montalto G, Anand BS **S-Editor** Song XX **L-Editor** **E-Editor**

A

**B**

**Figure 1 Box Plot for hepatitis C virus RNA (polymerase chain reaction) levels (A), total antioxidant capacity (B) before and after treatment. A:** Hepatitis C virus (HCV) RNA [polymerase chain reaction (PCR)] 0: polymerase chain reaction values of patients before treatment;HCV RNA (PCR) 3: polymerase chain reaction values of patients after 3 mo of treatment.Median test (equivalent to Wilcoxon matched pairs test), *P* < 0.001 (significant); **B:** Total antioxidant capacity (TAC) 0: total antioxidant capacity levels of patients before treatment;TAC 3: total antioxidant capacity levels of patients after 3 mo of treatment. Paired *t*-test, *P* < 0.001 (significant).

**Figure 2 Line Plot for hepatitis C virus RNA levels (polymerase chain reaction) in individual patients at baseline and after 3 mo of treatment.** Series 1 (dark dots): Hepatitis C virus (HCV) RNA levels [polymerase chain reaction (PCR)] in all patients at baseline; Series 2 (pale dots): HCV RNA levels (PCR) in all patients after 3 mo of treatment.

**Table 1 Clinical assessment data at baseline and after treatment *n* (%)**

|  |  |  |  |
| --- | --- | --- | --- |
| Characteristic | Baseline  Percent % (n) | After treatment  Percent % (n) | Significance (p) |
| Hepato and/or splenomegaly | 19 (63.30) | 19 (63.30) |  |
| Jaundice | 8 (26.70) | 5 (16.70) | 0.25 |
| Palmar erythema | 10 (33.30) | 8 (26.70) | 0.5 |
| Spider naevi | 8 (26.70) | 4 (13.30) | 0.125 |
| Lower limb edema | 16 (53.30) | 7 (23.30) | 0.004 |
| Clinically detected Ascites | 13 (43.30) | 8 (26.70) | 0.063 |

After treatment: 3 moof nigella sativa treatment.Mc Nemar’s test was used to compare categorical data overtime.

**Table 2 Laboratory data assessment at baseline and after treatment**

|  |  |  |  |
| --- | --- | --- | --- |
| Parameter | Base line | After 3 mo treatment | *P* |
| Hb (g %) | 11.8 ± 2.1 | 12.2 ± 2.2 | 0.1 |
| RBCs (× 106/µL) | 4.13 ± 0.9 | 4.3 ± 0.9 | 0.001 |
| WBCs (×103/µL) | 6.4 ± 2.1 | 5.6 ± 2.2 | 0.013 |
| Platelets (PLT) (× 103/µL) | 167.7 ± 91.2 | 198.5 ± 103 | 0.004 |
| Hematocrit (%) | 35.5 ± 6.3 | 37.3 ± 6.3 | 0.056 |
| ALT (iu/L) | 35.0 ± 15.7 | 41 ± 24.4 | 0.255 |
| AST (iu/L) | 40.9 ± 30.4 | 46.8± 32.2 | 0.307 |
| Total protein (g/dL) | 7.1 ± 0.7 | 7.5 ± 0.8 | 0.001 |
| Albumin (g/dL) | 3.5 ± 0.9 | 3.69 ± 0.9 | 0.008 |
| Direct bilirubin (mg/dL) | 0.5 ± 0.8 | 0.57 ± 1.5 | 0.745 |
| Total bilirubin (mg/dL) | 1.46 ± 1.5 | 1.36 ± 1.3 | 0.428 |
| PT (s) | 14.1 ± 2.7 | 13.8 ± 2.2 | 0.562 |
| INR | 1.18 ± 0.2 | 1.2 ± 0.2 | 0.974 |
| BUN (mg/dL) | 13.5± 6.2 | 14.1 ± 5 | 0.540 |
| Creatinin (mg/dL) | 0.99 ± 0.4 | 0.88 ± 0.2 | 0.102 |
| Serum α-fetoprotein (iu/mL) | 5.07 ± 1.8 | 4.67 ± 2.3 | 0.194 |
| Sodium (mmole/L) | 135.5 ± 6.1 | 133.5 ± 6 | 0.064 |
| Potassium (mmole/L) | 4.1 ± 0.5 | 4 ± 0.5 | 0.350 |
| Total antioxidant capacity (mmol/L) | 1.35 ± 0.5 | 1.61 ± 0.6 | 0.001 |
| Fasting blood sugar (mg/dL) | 104.03 ± 43.4 | 92.1 ± 31.3 | 0.001 |
| Post prandial blood Sugar (mg/dL) | 143.67 ± 72.6 | 112.1 ± 42.9 | 0.001 |
| PCR (copies) | 380 808.7 ± 610 937 | 147 028.2 ± 475 225.6 | 0.001 |

Paired *t*-test for all parameters, Median test (equivalent to Wilcoxon matched pairs test) for polymerase chain reaction levels. Hb: Hemoglobin; RBCs: Red blood cells; WBCs: White blood cells; ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; PT: Prothrombin time; INR: International normalized ratio; BUN: Blood urea nitrogen; PCR: Polymerase chain reaction.

**Table 3 Polymerase chain reaction response after treatment**

|  |  |
| --- | --- |
| Response | *n* (%) |
| Total Responders  Chronic liver disease  Compensated cirrhosis  Decompensated cirrhosis | 5 (16.67)  3  1  1 |
| Total Partial responders  Chronic liver disease  Compensated cirrhosis  Decreased 1 log  Decreased 2 log  Decompensated cirrhosis | 15 (50)  5  4  1  3  16 |
| Total Non-responders  Chronic liver disease  Compensated cirrhosis  Decompensated cirrhosis | 10 (33.33)  7  3 |

1Patients decreased polymerase chain reaction (PCR) but in same log. Non- responders: Patients did not show a decrease or showed an increase in PCR after 3 mo of treatment with Nigella Sativa; Responders: Patients became seronegative after three months treatment with Nigella Sativa; Partial responders: Patients showed a decrease in PCR but were still seropositive after 3 mo of treatment with Nigella Sativa.

**Table 4 Child-Pugh score at baseline and after 3 mo in patients with compensated and decompensated cirrhosis**

|  |  |  |  |
| --- | --- | --- | --- |
| Patients | Child-Pugh score at baseline | Child-Pugh score after 3 mo of treatment | HCV RNA (PCR) response |
| 1 | B | B | Partial responder |
| 2 | C | B | Partial responder |
| 3 | A | A | Partial responder |
| 4 | B | B | Partial responder |
| 5 | A | A | Partial responder |
| 6 | C | C | Partial responder |
| 7 | C | B | Non- responder |
| 8 | C | B | Partial responder |
| 9 | A | A | Responder |
| 10 | B | B | Non-responder |
| 11 | C | B | Partial responder |
| 12 | B | A | Responder |
| 13 | C | B | Non-responder |
| 14 | A | A | Partial responder |
| 15 | A | A | Partial responder |

HCV: Hepatitis C virus; PCR: Polymerase chain reaction.