Atherosclerosis in chronic hepatitis C virus patients with and without liver cirrhosis

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Abstract Background: Chronic Hepatitis C virus (HCV) infection and liver cirrhosis may be associated with atherosclerosis and coronary artery disease (CAD). There are two phases to atherosclerosis, Subclinical and Clinical. Assessment of atherosclerosis may be started at its Subclinical phase by the evaluation of Epicardial Fat Thickness (EpFT) and Carotid Intima Thickness (CIMT).

Aim of the study: The aim of the study was to evaluate Clinical and Subclinical atherosclerosis in chronic HCV patients with and without liver cirrhosis by evaluating CIMT and EpFT and correlating the results with Child-Pugh functional scoring of cirrhosis as well as with ultrasound and laboratory parameters that define the severity of liver disease.

Patients and methods: This study involved 64 chronic HCV patients that were divided into two groups: 24 patients without liver cirrhosis and 40 patients with liver cirrhosis in addition to 20 apparently healthy volunteers serving as control.

All of the 84 subjects were subjected to the following: Clinical evaluation; Routine Laboratory Evaluation (CBC, Liver Function Tests, Renal Function Tests, Serum electrolytes, Cholesterol, Triglycerides, HBs antigen and HCV antibody); ECG; Abdominal ultrasound; Echocardiographic evaluation of segmental wall motion abnormalities and EpFT and B-Mode Carotid ultrasonography for evaluation of CIMT.

Results: In the cirrhotic HCV group, the CIMT and EpFT were both significantly increased [Compared to control group (p = 0.000), compared to the non-cirrhotic HCV group (p = 0.000)]. In the non-cirrhotic HCV group, the CIMT and EpFT were both significantly increased compared to the control group with a p-value of 0.003 for CIMT and 0.048 for EpFT. The CIMT and EpFT were...
Atherosclerosis is one of leading causes of mortality and morbidity throughout the world. It is a chronic, progressive, inflammatory disease with a long asymptomatic course. Atherosclerosis passes through two phases, a Pre-clinical (Sub-clinical) phase and a Clinical phase. The Subclinical phase refers to the early stages of atherosclerotic disease and confers an increased risk of cardiovascular disease which may eventually lead to coronary artery disease. A variety of imaging modalities have been used to assess subclinical atherosclerosis. Infectious agents including chronic hepatitis C virus (HCV) infection have been associated with atherosclerosis and a higher risk of coronary artery disease. Suggested mechanisms include the promotion of HCV endocytosis by low-density lipoprotein receptor and induction of oxidative stress by HCV core protein leading to the oxidation of lipoprotein in atherosclerotic plaques. It has also been associated with systemic vasculitides, increased concentration of soluble intercellular adhesion molecules and the presence of anti-endothelial antibodies.

HCV patients are also at increased risk of developing hepatic steatosis. Patients with hepatic steatosis may have underlying metabolic abnormalities including insulin resistance as well as increased inflammatory markers that may contribute to endothelial dysfunction. The presence of steatosis may contribute to the development of other metabolic diseases, such as hypertension and diabetes, all of which may exacerbate CAD risk in individuals with HCV. Adinolfi et al. performed a study on HCV patients with and without hepatic steatosis and found a significantly higher prevalence of cardiovascular disease in HCV patients compared to age- and sex-matched controls. In addition, even after matching for the presence of steatosis, a significantly higher prevalence of cardiovascular disease was seen in the HCV patients, supporting the notion that HCV may increase the risk of cardiovascular disease independent of steatosis.

It was believed that cirrhosis of the liver had a protective role for CAD. These observations were motivated by a theoretical protective role of some common features in cirrhotic patients like reduced circulating low-density lipoproteins and total cholesterol, decreased vascular resistance, low blood pressure, and high levels of circulating estrogens. Now end-stage liver disease (ESLD) could be considered to be a coronary artery risk equivalent, and it is therefore important to appropriately risk-stratify patients even in the absence of symptoms. It was found that the prevalence of coronary artery disease (CAD) in liver transplanted (LT) candidates can reach up to 26%. In turn, CAD is associated with increased post-LT mortality and a greater incidence of new cardiovascular morbidity.

Epicardial fat thickness (EpFT), carotid intima media thickness (CIMT), and aortic stiffness index have been used to assess subclinical atherosclerosis. CIMT is a simple and inexpensive tool to assess the cumulative effect of atherosclerotic risk factors and is recommended by the American Heart Association for the noninvasive assessment of cardiovascular risk. Some studies have reported that HCV infected patients had higher CIMT compared to healthy control subjects and the mean CIMT of HCV infected patients was significantly greater than that of healthy controls; this difference cannot be attributed to a difference in age, gender, body mass index (BMI) or cardiometabolic risk factors.

Epicardial Fat Thickness (EpFT) may reflect the amount of visceral fat, which is associated with insulin resistance, inflammation and CAD. EpFT is a rich source of free fatty acids and a number of bioactive molecules and inflammatory cytokines. Some reports have suggested a crucial role of EpFT in the development of CAD through changes in adipokine expressions in EpFT, which promote pro-inflammatory characteristics, thereby facilitating the progression of coronary disease.
atherosclerosis.\textsuperscript{32–35} EpFT measured using transthoracic echocardiography significantly correlates with CAD and is considered a marker of severity of coronary lesions.\textsuperscript{36} EpFT has been considered to be a possible cardiovascular risk indicator.\textsuperscript{37,38} Assessment of EpFT by transthoracic echocardiography (TTE) could be a simple and practical tool for cardiovascular risk stratification in clinical practice.\textsuperscript{39,40}

2. Aim of the study

The study aimed to evaluate clinical and subclinical atherosclerosis in chronic hepatitis C virus patients with and without liver cirrhosis by evaluating carotid EpFT and CIMT as well as to correlate the degree of EpFT and CIMT with the Child-Pugh functional scoring of cirrhosis and with the ultrasound and laboratory parameters that define the severity of liver disease.

3. Patients and methods

3.1. Patients

3.1.1. Inclusion criteria

This study was carried out in 2014 and 2015 and included 64 patients with chronic hepatitis C virus from outpatient clinics and outpatient services of Theodor Bilharz Research Institute Hospital. These patients were divided into two groups according to findings of transient ultrasound elastography fibro-scan\textsuperscript{41–43} and abdominal ultrasound.

Group I: included 24 patients with chronic hepatitis C virus without liver cirrhosis, fibro-scan was F0-F1 and liver ultrasound showed no apparent cirrhosis.

Group II: included 40 patients with chronic hepatitis C virus with liver cirrhosis, fibro-scan was F4 and abdominal ultrasound showed shrunken and irregular liver surface, ascites and splenomegaly. They were classified according to the Child-Pugh functional scoring of cirrhosis.

Group III: included 20 apparently healthy volunteers as control group matched for age and sex and with normal liver ultrasound, normal liver function tests and negative hepatitis markers.

3.1.2. Exclusion criteria

Subjects with history of heart disease, diabetes mellitus, hypertension (blood pressure > 140/90 mmHg), smoking, hyperlipidemia, acute or chronic kidney disease, alcohol consumption, pregnancy, liver masses, and anemia with hemoglobin less 10 g% were excluded from the study.

3.2. Methods

3.2.1. General methods

All patients in the study were subjected to the following:

- Electrocardiogram was done to all subjects to detect any findings suggestive of CAD.
- Abdominal ultrasound scanning was performed to all subjects using a Toshiba Nemo 30 scanner equipped with a 3.5 MHz linear transducer by a member of the study team.

3.2.2. Transthoracic echocardiography

Echocardiography was performed using a Toshiba Nemo 30 scanner equipped with a 2.5 MHz linear transducer to detect segmental wall motion abnormalities suggestive of CAD and to measure epicardial fat thickness.

Epicardial fat thickness was identified as the echocardiographically free space between the outer wall of the myocardium and the visceral layer of the pericardium, and its thickness was measured perpendicularly to the free wall of the right ventricle at end-systole over 3 cardiac cycles. The mean value of 3 cardiac cycles from each echocardiographic view (including both parasternal long- and short-axis views) was recorded as the EpFT.\textsuperscript{44} These measurements were completed by three blinded members of the study team and measurements were averaged.

3.2.3. B mode carotid ultrasonography

High resolution B mode ultrasonography of both the common carotid arteries was performed using an ultrasound machine (Toshiba Memo 30 scanner) equipped with a 7.5 MHz high resolution transducer. Patients were examined in the supine position. The maximum CIMT was measured at the posterior wall of the common carotid artery, 2 cm before the bifurcation, as the distance between the first and second echogenic lines of the anterior and posterior arterial walls. CIMT was defined as a low-level echo gray band that does not project into the arterial lumen\textsuperscript{45} and was measured during end diastole. These measurements were completed by three blinded members of the study team and measurements were averaged.

4. Ethics

All patients were provided by informed consent, and the ethical committee of the hospital approved this study which was conducted in accordance with the Helsinki Declaration (1975).

5. Statistical analysis

Statistical analysis was performed using SPSS version 17. Data were expressed as the mean ± standard deviation (SD) for numerical variables. \( P \leq 0.05 \) was considered to be statistically significant and \( P \leq 0.01 \) was considered to be highly statistically significant. The univariate correlations of the carotid intima-media thickness and epicardial fat thickness with other parameters were also done.

6. Results

The demographic data of the patient groups and the control group revealed mean ages of 41.3 ± 9.95 years for the non-cirrhotic hepatitis C group, 42.5 ± 8.75 years for the cirrhotic group and 43.2 ± 7.85 years for the control group. In the non-cirrhotic hepatitis C group 16 patients (66.7%) were males and
8 patients (33.3%) were females; in the cirrhotic group 24 patients (60%) were males and 16 patients (40%) were females and in the control group, there were 13 males (65%) and 7 females (35%). There were no significant differences between the patient groups and the control group regarding age and gender (Table 1).

Hepatitis C patients with cirrhosis had a statistically significant decrease in systolic blood pressure together with a statistically significant increase in heart rate compared to the controls. Also, they had a statistically significant increase in their heart rate compared to hepatitis C patients without cirrhosis (Table 1).

The electrocardiographic findings showed inverted T and depressed ST segment in 2 patients (8.33%) having hepatitis C without cirrhosis, in 5 patients (12.5%) having hepatitis C with cirrhosis and in no subjects of the control group. These results were neither statistically significant between the patient groups and control nor between both patient groups (Table 1).

Regarding the echocardiographic findings, segmental wall motion abnormalities at rest were found in 1 patient (4.17%) having hepatitis C without cirrhosis, in 4 patients (10%) having hepatitis C patients with cirrhosis and in no subjects of the control group. These results were neither statistically significant between the patient groups and control nor between both patient groups (Table 1).

Regarding the laboratory data, hepatitis C patients with cirrhosis had statistically significant decrease in serum sodium, albumin and platelets count together with statistically significant increase in serum potassium, total bilirubin, INR and CRP compared to the control group and to the non-cirrhotic hepatitis C patients group. Also, hepatitis C patients with cirrhosis had statistically significant increase in ALT and AST compared to control group. Non-cirrhotic hepatitis C patients had F0-F1 fibro-scan with no apparent cirrhosis by abdominal ultrasonography (100%). Regarding the cirrhotic group, 32 patients (80%) had cirrhosis, ascites and splenomegaly by abdominal ultrasound while all the patients had F4 fibro-scan (Table 3). The 2 patients having asciites in the non-cirrhotic group were females with ruptured ovarian cysts.

Carotid intima-media thickness and epicardial fat thickness were significantly increased in the cirrhotic hepatitis C group compared to both the control group and the non-cirrhotic group. Also, the non-cirrhotic hepatitis C group showed statistically significant increase in the carotid intima-media thickness and epicardial fat thickness compared to control group (Table 4).

There was a statistically significant increase in CIMT in Child class B patients compared to Child class A patients and in Child class C patients compared to Child class B patients (Table 5).

There was a statistically significant increase in EpFT in Child class B patients compared to Child class A patients and in Child class C patients compared to Child class B patients (Table 6).

CIMT was positively correlated with AST, total bilirubin, INR%, CRP, spleen span, portal vein diameter and EpFT and negatively correlated with albumin, platelets count and liver span in HCV infected patients (Table 7).

EpFT was positively correlated with AST, total bilirubin, INR%, CRP, spleen span, and portal vein diameter; and CIMT negatively correlated with albumin, platelets count and liver span in HCV infected patients (Table 8).

| Table 1 | Demographic data, ECG and echocardiographic findings suggestive of CAD of the studied groups. |
|---------|---------------------------------------------------------------|
| Age     | 41.3 ± 9.95                                                   |
| Years   | 42.5 ± 8.75                                                   |
| Gender  | 43.2 ± 7.85                                                   |
| Male    | 0.492                                                         |
| Female  | 0.764                                                         |
| Systolic BP, mmHg | 0.617                             |
| Diastolic | 43.2 ± 7.85                                                   |
| BP, mmHg | 13 (65%)                                                      |
| Pulse beat/min | 0.569                             |
| ECG findings suggestive of CAD | 0.703                             |
| Echo findings suggestive of CAD | 0.904                             |
| P1 value | 0.115                                                         |
| P2 value | 0.493                                                         |
| P3 value | 0.013                                                         |

P1 value, between groups 1 and 3; P2 value, between groups 2 and 3; P3 value, between groups 1 and 2. P ≤ 0.05, statistically significant; P ≤ 0.01, highly statistically significant. ECG: electrocardiogram; CAD: coronary artery disease; Echo: echocardiography.
7. Discussion

Atherosclerosis is a chronic, progressive and inflammatory disease with a long asymptomatic prelude.\(^1\) Atherosclerosis has two phases: preclinical (subclinical) and clinical. Subclinical atherosclerosis refers to the early stages of the atherosclerotic disease, and may lead to coronary artery disease. A variety of imaging modalities have been used to assess subclinical atherosclerosis.\(^1,2\)

O’Leary et al. in their study concluded that CIMT is a simple and inexpensive tool to assess the cumulative effect of atherosclerotic risk factors and is an independent predictor of future cardiovascular risk.\(^25\) The ultrasound-based measurement of CIMT has become a standard for assessing arteriosclerosis and is recommended by the American Heart Association for the noninvasive assessment of cardiovascular risk.\(^26,27\)

Ahn et al. found that EpFT is strongly related to the development of coronary artery disease.\(^30\) Also Shemirani et al. found that increased EpFT is correlated with the severity of

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**Table 2** Laboratory data of the studied groups.

|                      | Hepatitis C without cirrhosis | Hepatitis C with cirrhosis | Control group | \(P1\) value | \(P2\) value | \(P3\) value |
|----------------------|-------------------------------|----------------------------|---------------|--------------|--------------|--------------|
| Na, mEq/L            | 136.85 ± 5.44                 | 130.16 ± 6.74              | 141.64 ± 2.04 | 0.001        | 0.000        | 0.000        |
| K, mEq/L             | 4.05 ± 0.24                   | 4.78 ± 0.56                | 4.14 ± 0.22   | 0.206        | 0.000        | 0.000        |
| Creat, mg/dL         | 0.95 ± 0.3                    | 1.0 ± 0.2                  | 0.92 ± 0.2    | 0.705        | 0.150        | 0.427        |
| ALT, U/L             | 61.78 ± 33.85                 | 51.65 ± 20.29              | 23.85 ± 2.06  | 0.000        | 0.000        | 0.139        |
| AST, U/L             | 53.8 ± 16.89                  | 74.96 ± 57.76              | 23.65 ± 4.04  | 0.000        | 0.000        | 0.086        |
| T bil., mg/dL        | 1.56 ± 0.36                   | 3.32 ± 3.79                | 0.54 ± 0.11   | 0.000        | 0.000        | 0.027        |
| Albumbin, g/dL       | 3.51 ± 0.48                   | 2.47 ± 0.49                | 4.21 ± 0.7    | 0.000        | 0.000        | 0.000        |
| Platelets count 10\(^9\)/L | 164.2 ± 32.7              | 62.6 ± 57.3                | 224.7 ± 58.7  | 0.000        | 0.000        | 0.000        |
| INR%                 | 1.13 ± 0.24                   | 1.69 ± 0.5                 | 1.01 ± 0.13   | 0.000        | 0.012        | 0.000        |
| LDL, mg/dL           | 106.4 ± 18.5                  | 102.6 ± 17.5               | 109.6 ± 16.4  | 0.551        | 0.071        | 0.331        |
| HDL, mg/dL           | 44.2 ± 15.4                   | 43.4 ± 14.6                | 46.2 ± 11.6   | 0.635        | 0.458        | 0.836        |
| T Choles., mg/dL     | 151.2 ± 34.3                  | 148.2 ± 31.3               | 153.2 ± 41.3  | 0.603        | 0.862        | 0.722        |
| TG, mg/dL            | 112.3 ± 46.5                  | 110.6 ± 40.5               | 121.9 ± 27.7  | 0.422        | 0.267        | 0.878        |
| CRP, mg/L            | 5.6 ± 2.1                     | 7.2 ± 2.5                  | 2.3 ± 0.4     | 0.000        | 0.000        | 0.111        |

\(P1\) value, between groups 1 and 3; \(P2\) value, between groups 2 and 3; \(P3\) value, between groups 1 and 2.

Na: serum sodium; K: serum potassium; Creat: creatinine; ALT: alanine aminotransferase; AST: aspartate aminotransferase; T bil.: Total bilirubin; INR: international normalized ratio; LDL: low density lipoprotein; HDL: high density lipoprotein; T Choles: total cholesterol; TG: Triglyceride; CRP: C reactive protein.

**Table 3** Fibro-scan and abdominal ultrasound data of the studied groups.

|                      | Hepatitis C without cirrhosis | Hepatitis C with cirrhosis | Control group | \(P1\) value | \(P2\) value | \(P3\) value |
|----------------------|-------------------------------|----------------------------|---------------|--------------|--------------|--------------|
| Fibro-scan           | FO-F1 (24) (100%)             | F4 (40) (100%)             | –ve           | 0.000        | 0.000        | 0.000        |
| Coarse liver         | –ve                           | + ve (32) (80%)            | –ve           | 0.539        | 0.002        | 0.016        |
| Liver span, cm       | 14.3 ± 2.7                    | 12.4 ± 3.1                 | 14.7 ± 1.1    | 0.187        | 0.000        | 0.000        |
| Ascites              | + ve (2) (8.33%)              | + ve (34) (85%)            | –ve           | 0.000        | 0.000        | 0.000        |
| Spleen span, cm      | 14.2 ± 2.4                    | 16.3 ± 3.7                 | 8.2 ± 2.3     | 0.000        | 0.000        | 0.016        |
| PV diameter, mm      | 8.5 ± 2.6                     | 13.7 ± 3.4                 | 7.1 ± 1.2     | 0.032        | 0.000        | 0.000        |

\(P1\) value, between groups 1 and 3; \(P2\) value, between groups 2 and 3; \(P3\) value, between groups 1 and 2.

\(P \leq 0.05\), statistically significant; \(P < 0.01\), highly statistically significant.

**Table 4** Carotid intima-media thickness and epicardial fat thickness of the studied groups.

|                      | Hepatitis C without cirrhosis | Hepatitis C with cirrhosis | Control group | \(P1\) value | \(P2\) value | \(P3\) value |
|----------------------|-------------------------------|----------------------------|---------------|--------------|--------------|--------------|
| CIMT, mm             | 8.2 ± 1.4                     | 11.9 ± 1.8                 | 6.2 ± 2.7     | 0.003        | 0.000        | 0.000        |
| EpFT, mm             | 4.4 ± 1.8                     | 6.7 ± 2.1                  | 3.2 ± 1.3     | 0.048        | 0.000        | 0.000        |

\(P1\) value, between groups 1 and 3; \(P2\) value, between groups 2 and 3; \(P3\) value, between groups 1 and 2.

\(P \leq 0.05\), statistically significant; \(P < 0.01\), highly statistically significant.

CIMT: carotid intima-media thickness and EpFT: epicardial fat thickness
Many other previous studies have reported that epicardial fat has a significant association with CIMT, coronary artery calcification, increased ApoB/ApoA1 ratio and cardiovascular risk according to the Framingham score. Abaci et al. also demonstrated a significant correlation between epicardial fat and CIMT. Rego et al. examined 300 asymptomatic subjects and found that epicardial fat had a significant association with CIMT, coronary artery calcification, increased ApoB/ApoA1 ratio and cardiovascular risk according to the Framingham score. The prevalence of carotid plaque was significantly greater in those with EpFT thickness ≥ 5 mm who had low Framingham risk scores compared with those with EpFT thickness < 5 mm.

In the current study, in spite of excluding patients having history of ischemic heart disease, the electrocardiogram showed inverted T wave and depressed ST segment in 2 (8.33%) patients having non-cirrhotic hepatitis C and in 5 patients (12.5%) having cirrhotic hepatitis C. These results were neither statistically significant between the patient groups and control nor between both patient groups. Echocardiography showed wall motion abnormalities at rest in 1 patient (4.17%) having cirrhotic hepatitis C and in 4 (10%) patients having non-cirrhotic hepatitis C. These results were not statistically significant between patient groups and control or between both patient groups. Our study showed statistically significant increase of CIMT and EpFT in the hepatitis C cirrhotic group compared to the control group and to the hepatitis C non-cirrhotic group. Also, hepatitis C non-cirrhotic patients had statistically significant increase in CIMT and EpFT compared to the control group. These findings suggest increased prevalence of subclinical atherosclerosis in hepatitis C patients especially in the presence of cirrhosis.

In agreement with our findings, Butt et al. have found that HCV infection was associated with a higher risk of atherosclerosis and coronary artery disease after adjustment of traditional risk factors. The study by Ampuero and Romero-Gómez revealed a strong link between HCV infection and the atherogenic process, showing a higher risk of coronary artery stenosis. Cetin et al. found that EpFT of 6.3 mm was determined as a high risk value for subclinical atherosclerosis with a 72.5% sensitivity and 71.7% specificity. Many other previous studies have reported that increased EpFT is associated with CAD and metabolic syndrome.

### Table 5 Carotid intima-media thickness in patients with cirrhosis as related to their Child-Pugh classification.

| Group (n)          | CIMT  | p-value |
|--------------------|-------|---------|
| Controls (20)      | 6.2 ± 2.7 | 0.000  |
| HCV with Cirrhosis (40) | 11.9 ± 1.8 | 0.000  |
| Child class A (10) | 9.2 ± 1.7 | 0.000  |
| Child class B (14) | 10.9 ± 1.1 | 0.002  |
| Child class C (16) | 12.5 ± 1.2 | 0.001  |

P ≤ 0.05, statistically significant; P ≤ 0.01, highly statistically significant.

HCV: hepatitis C virus and CIMT: carotid intima-media thickness.
p < 0.000: cirrhosis group (total) compared to control group;
p = 0.007: child class B group compared to child class A group;
p = 0.001: child class C group compared to child class B group.

### Table 6 Epicardial fat thickness in patients with cirrhosis as related to their Child-Pugh classification.

| Group (n)          | EpFT  | p-value |
|--------------------|-------|---------|
| Controls (20)      | 3.2 ± 1.3 | 0.000  |
| HCV with Cirrhosis (40) | 6.7 ± 2.1 | 0.000  |
| Child class A (10) | 4.6 ± 1.5 | 0.000  |
| Child class B (14) | 5.9 ± 1.2 | 0.028  |
| Child class C (16) | 7.3 ± 1.3 | 0.005  |

P ≤ 0.05, statistically significant; P ≤ 0.01, highly statistically significant.

HCV: hepatitis C virus and EpFT: epicardial fat thickness.
p < 0.000: cirrhosis group (total) compared to control group;
p = 0.028: child class B group compared to child class A group;
p = 0.005: child class C group compared to child class B group.

### Table 7 Correlation of different variables in HCV infected patients with CIMT.

| Parameter          | r-value | p-value |
|--------------------|---------|---------|
| AST, U/L           | 0.385   | 0.002   |
| T bil., mg/dL      | 0.378   | 0.003   |
| Albumin, g/dL      | -0.379  | 0.003   |
| Platelets count 10^9/L | -0.382  | 0.002   |
| INR%               | 0.456   | 0.001   |
| CRP, mg/L          | 0.378   | 0.003   |
| Liver span, cm     | -0.433  | 0.001   |
| Spleen span, cm    | 0.417   | 0.001   |
| PV diameter, mm    | 0.372   | 0.003   |
| EpFT, mm           | 0.465   | 0.001   |

P ≤ 0.05, statistically significant; P ≤ 0.01, highly statistically significant.

AST: aspartate aminotransferase; T bil.: Total bilirubin; INR: international normalized ratio; CRP: C reactive protein; PV: portal vein; EpFT: epicardial fat thickness.

### Table 8 Correlation of different variables in HCV infected patients with EpFT.

| Parameter          | r-value | p-value |
|--------------------|---------|---------|
| AST, U/L           | 0.379   | 0.003   |
| T bil., mg/dL      | 0.384   | 0.002   |
| Albumin, g/dL      | -0.370  | 0.003   |
| Platelets count 10^9/L | -0.378  | 0.003   |
| INR%               | 0.430   | 0.001   |
| CRP, mg/L          | 0.386   | 0.002   |
| Liver span, cm     | -0.424  | 0.001   |
| Spleen span, cm    | 0.437   | 0.001   |
| PV diameter, mm    | 0.379   | 0.003   |
| CIMT, mm           | 0.465   | 0.001   |

P ≤ 0.05, statistically significant; P ≤ 0.01, highly statistically significant.

AST: aspartate aminotransferase; T bil.: Total bilirubin; INR: international normalized ratio; CRP: C reactive protein; PV: portal vein; CIMT: carotid intima-media thickness.
heart disease, carotid atherosclerosis, peripheral artery disease and, ultimately, CVD-related mortality.\textsuperscript{55} Lin et al. reported that HCV-infected subjects had 1.76-fold increased risk of an ischemic electrocardiogram when compared with HCV non-infected subjects.\textsuperscript{56} Zakaria et al. found that mean CIMT of HCV infected Egyptians of a low cardiovascular risk was significantly greater than that of healthy controls.\textsuperscript{29} Although the results of some studies suggest that hepatitis C infection is an independent risk factor for carotid atherosclerosis,\textsuperscript{57,58} these findings have not been confirmed by others who have shown that chronic viral hepatitis may protect from atherosclerosis in the carotid arteries.\textsuperscript{59} In another report, no independent association could be found between serological markers for hepatitis C virus infection and cardiovascular morbidity or atherosclerosis in the carotid arteries.\textsuperscript{60}

Assessing cirrhotic candidates for LT has revealed a high prevalence of asymptomatic CAD in these patients. Sixty-five LT candidates without known CAD underwent multidetector computed tomography coronary angiography: 58% had mild CAD and 34% had moderate to severe CAD.\textsuperscript{61} Other studies have revealed a significant burden of unrecognized, asymptomatic CAD.\textsuperscript{62,22} Carey et al.\textsuperscript{63} reported that 13.3% of liver transplanted patients with moderate or severe coronary stenosis were asymptomatic, presumably due to the masking effect of poor functional status.

Our study revealed that CRP was significantly increased in hepatitis C patients with cirrhosis compared to the control group and hepatitis C patients without cirrhosis. Our study agrees with many studies which concluded that the majority of endothelial biomarkers were elevated in the chronic HCV patients. Although varied, the biomarker pattern points to an increased risk of cardiovascular mortality in the chronic HCV patients: CRP, sVCAM-1, sICAM-1, and s-E-selectin, were all elevated in these patients.\textsuperscript{64–66} Shimada et al. found that elevated serum level of CRP is an independent predictor of cardiovascular events in patients with CAD.\textsuperscript{67}

In our study, there were positive correlations between CIMT and AST, total bilirubin, INR%, CRP, spleen span, portal vein diameter and EpFT in addition to negative correlations between CIMT and albumin, platelets count and liver span in all HCV infected patients. There were also positive correlations between EpFT and AST, total bilirubin, INR%, CRP, spleen span, portal vein diameter and CIMT in addition to negative correlations between EpFT and albumin, platelets count and liver span in all HCV infected patients. These findings in addition to the statistically significant increase of EpFT and CIMT in correlation with the Child-Pugh functional scoring of cirrhosis indicate that the subclinical atherosclerosis detected in our patients with hepatitis C is correlated with the functional scoring of cirrhosis and the severity of liver disease.

In agreement with our findings, Petta and his colleagues have found that severe hepatic fibrosis is associated with a higher risk of carotid atherosclerosis in chronic hepatitis C patients.\textsuperscript{68} In another study carried out on fatty liver patients, Petta et al. found that higher epicardial fat thickness is associated with the severity of liver fibrosis; in addition to that they concluded that the morphological and functional cardiac alterations are more pronounced in accordance with the severity of fibrosis.\textsuperscript{59} Dogan and his colleagues found that the Framingham risk score (FRS) which provides an estimate of coronary heart disease (CHD) risk is associated with the noninvasive scoring indexes like fibrosis scan and they also found that assessment of liver fibrosis in patients with fatty liver may be useful for the risk stratification of CHD in the absence of liver biopsy in clinical practice.\textsuperscript{70}

8. The novelty of the study

To our knowledge the present study is unique in assessing EpFT in post hepatitis C liver cirrhosis and in correlating the degree of EpFT with Child-Pugh functional scoring of cirrhosis and with ultrasound and laboratory parameters that define severity of liver disease. Previous studies have used CIMT (not EpFT) for screening of atherosclerosis in hepatitis C patients while EpFT have been used only in assessment of atherosclerosis in fatty liver disease patients.

9. Limitation

For single-center study with small sample size, we suggest that multicenter approaches may be necessary to attain larger sample sizes.

10. Conclusions

Our study revealed increased EpFT and CIMT in chronic hepatitis C virus patients especially in those with cirrhosis, and the degree of EpFT and CIMT was shown to be correlated with the Child-Pugh functional scoring of cirrhosis and with the ultrasound and laboratory parameters that define severity of liver disease. Our study also showed close correlation between EpFT and CIMT in these patients.

The echocardiographic assessment of EpFT and the carotid Doppler assessment of CIMT may provide appropriate and simple screening markers for subclinical atherosclerosis and cardiovascular risk in patients with chronic hepatitis C virus with and without cirrhosis. Patients undergoing LT would likely benefit from echocardiographic assessment of EpFT and carotid Doppler for evaluation of CIMT in addition to stress testing to help identify those patients who are at high or low risk for cardiac outcomes in the LT.

Conflict of interest

The authors declare that they have no conflict of interest.

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