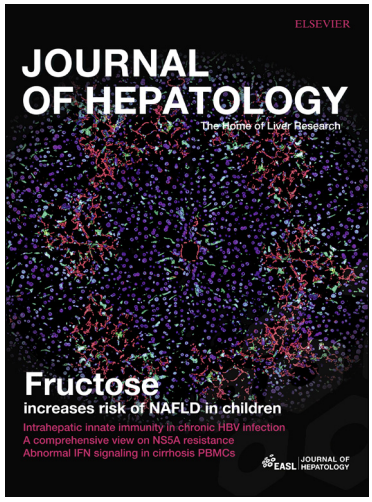

Hepatitis C Virus Eradication by Direct Antiviral Agents Improves Carotid Atherosclerosis in patients with Severe Liver Fibrosis

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ABBREVIATIONS: HCV: hepatitis C virus; G1: genotype 1; CHC: chronic hepatitis C; IR: insulin resistance; HOMA: homeostasis model assessment.

KEY WORDS : ATHEROSCLEROSIS, HCV, DAA, SVR

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ABSTRACT

Background and Aim: Recent studies suggest an association between HCV infection and cardiovascular damage, including carotid atherosclerosis, with a possible effect of HCV clearance on cardiovascular outcomes. We aimed to examine whether HCV eradication by direct antiviral agents (DAA) improves carotid atherosclerosis in HCV-infected patients with advanced fibrosis/compensated cirrhosis.

Materials and Methods: One hundred eighty-two consecutive HCV patients with advanced fibrosis or compensated cirrhosis were evaluated by virological, anthropometric and metabolic measurements. All patients underwent DAA-based antiviral therapy according to AISF/EASL guidelines. Intima-media thickness (IMT), carotid thickening (IMT≥1 mm) and carotid plaques, defined as focal thickening of ≥ 1.5 mm at the level of common carotid, were evaluated by ultrasonography (US) at baseline and 9-12 months after the end of therapy.

Results: Fifty-six percent of patients were males, mean age was 63.1±10.4 years and 65.9% had compensated cirrhosis. One patient out of five had diabetes, 14.3% were obese, 41.8% had arterial hypertension and 35.2% were smokers. Mean IMT was 0.94±0.29 mm, 42.9% had IMT≥1 mm, and 42.9% had carotid plaques. All patients achieved a 12-weeks sustained virological response. IMT significantly decreased from baseline to follow-up (0.94±0.29 mm vs. 0.81±0.27, p<0.001). Consistently, a significant reduction in the prevalence of patients with carotid thickening from baseline to follow-up was observed (42.8% vs. 17%, p<0.001), while no changes were reported for carotid plaques (42.8% vs. 47.8%, p=0.34). These results were confirmed in sub-groups of patients stratified for cardiovascular risk factors and liver disease severity.

Conclusion: HCV eradication by DAA improves carotid atherosclerosis in patients with severe fibrosis without or with additional metabolic risk factors. The impact of this

improvement in the atherosclerotic burden in terms of reduction of major cardiovascular outcomes is worth investigating in the long term.

Lay of Summary

Hepatitis C Virus (HCV) eradication by direct antiviral agents improves carotid atherosclerosis in patients with advanced fibrosis/compensated cirrhosis

The improvement in intima-media thickness and carotid thickening was confirmed after stratification for severity of liver disease and cardiovascular risk factors

HCV eradication by DAA also lead to improvement in glucose homeostasis and increase in cholesterol levels

INTRODUCTION

Hepatitis C virus (HCV) infection affected roughly 71.1 million of individuals in 2015 with an estimated global prevalence of 1.0% even if with major geographical heterogeneity [1].

The clinical burden and the prognosis of HCV infection depends not only on the higher risk of liver-related complications and death, but also of the increase in extrahepatic complications [2]. Consistent with these data, a recent meta-analysis highlighted that HCV infected patients are at higher risk of extrahepatic manifestations related to immune dysregulation (mixed cryoglobulinemia, lymphoma, etc) and metabolic dysfunction (type 2 diabetes, etc) compared to subjects without infection [3]. Notably, these epidemiological data are supported by experimental evidence, and by studies reporting a positive effect of HCV eradication by both interferon-based and direct antiviral agents (DDA)-based therapies on HCV-related extrahepatic manifestations [2].

Emerging data also support a link between HCV infection and cardiovascular alterations. HCV infection has been associated with an increased risk of both carotid [4-6] and coronary [7-9] atherosclerosis, myocardial injury [10], peripheral artery disease [11], cerebro- and cardiovascular events [12,13], and finally cardiovascular mortality [14,15]. However, contrasting data have also been reported, and the basis of this association stems on associative data, theoretical speculations, and inconclusive experimental evidence [16]. A recent meta-analysis searched for summarizing available evidence, overall confirming the negative impact of HCV infection on cardiovascular alterations/mortality even if in a context of high heterogeneity, suggesting that a better understanding of this potential association is still required [17].

The availability of safe and effective DAA-based regimens raised the question whether HCV eradication can also improve cardiovascular outcomes. The greater

proportion of available data mostly arise from populations of patients eligible to and underwent interferon (IFN)-based therapies [15, 18-24]. Available data suggest that viral eradication (SVR: sustained virological response) reduces cardiovascular risk, but the design studies do not allow to discriminate whether the beneficial effect of SVR, is due to elimination of the virus, or may be driven by IFN or confounded by patients selection and kind of bias. To contribute resolving this issue, we assessed the impact of SVR on carotid atherosclerosis in a cohort of patients with advanced chronic hepatitis/compensated cirrhosis due to HCV infection who underwent DAA-based therapies, further stratifying the effect according to cardiovascular risk factors and liver disease severity.

MATERIALS & METHODS

Patients

One hundred eighty-two consecutive patients with advanced chronic hepatitis/compensated cirrhosis due to HCV infection were prospectively recruited at three Italian referral centers in Palermo (n=84), Naples (n=72) and Milan (n=26). Patients were included if 1) they had a clinical or histological diagnosis of advanced chronic hepatitis/compensated cirrhosis due to HCV infection at the time of enrollment; 2) they underwent IFN-free, DAA-based therapy. Advanced fibrosis (F3 fibrosis) was diagnosed by histology and/or by liver stiffness measurement (LSM) by FibroScan (≥10 to ≤12 KPa); compensated cirrhosis (F4 fibrosis) was diagnosed by histology and/or by LSM (>12 KPa), and/or by evidence of oesophageal varices.

Exclusion criteria were: 1) advanced cirrhosis (Child-Pugh B and C); 2) hepatocellular carcinoma; 3) liver disease of different or mixed etiology (i.e., excessive

alcohol consumption, hepatitis B, autoimmune liver disease, Wilson's disease, hemochromatosis, α1-antitrypsin deficiency); 4) alcohol intake >20 g/day during the previous year (evaluated by interview of patients on amount, frequency and type, and confirmed by at least one family member); 5) HIV infection; 6) treatment with immunosuppressive drugs and/or regular use of steatosis-inducing drugs (corticosteroids, valproic acid, tamoxifen, amiodarone); 7) previous history of ischemic heart or cerebral disease , and 8) treatment with anti-aggregating drugs or statins; 9) active i.v. illicit drug addiction.

As controls we considered a cohort of 76 consecutive HCV-infected patients with advanced fibrosis/cirrhosis, observed in 2014, studied for atherosclerosis progression, and not underwent antiviral therapy because awaiting for DAA not still available in that year in Italy.

The study was performed in accordance with the principles of the Declaration of Helsinki and its appendices, and with local and national laws. Approval was obtained from the hospital's Institutional Review Board and Ethics Committee, and written informed consent was obtained from all patients.

Clinical and laboratory assessment

Clinical and anthropometric data were collected at the time of the enrollment and 9-12 months after the end of antiviral therapy. BMI was calculated on the basis of weight, in kilograms, and height, in meters. Patients were classified as obese when BMI was ≥30. The diagnosis of arterial hypertension was based on the following criteria: systolic blood pressure ≥135 mm Hg and/or diastolic blood pressure ≥85 mm Hg (measured three times within 30 minutes, in the sitting position and using a brachial sphygmomanometer), or use

of blood-pressure-lowering agents. The diagnosis of type 2 diabetes was based on the revised criteria of the American Diabetes Association, using a value of fasting blood glucose ≥126 mg/dL on at least two occasions [25]. In patients with a previous diagnosis of type 2 diabetes, current therapy with insulin or oral hypoglycemic agents was documented.

A 12-hour overnight fasting blood sample was drawn at the time of enrollment and 9-12 months after the end of antiviral therapy to determine serum levels of ALT, total cholesterol, plasma glucose concentration and platelet count.

Carotid artery evaluation

Carotid atherosclerosis was evaluated, at baseline and 9-12 months after the end of antiviral therapy, by an expert physician for centre in a blinded fashion, using a high-resolution B-mode ultrasonography equipped with a multifrequency linear probe. The HCV population considered as control had also US at the enrollment and after 9-12 months. The same operators who performed the baseline exam also performed the follow-up evaluation. The carotid arteries were investigated in longitudinal projections of both the left and right side at the level of the common carotid artery, bulb and internal carotid in each patient. The carotid IMT was measured as the difference between the first (intima lumen) interface and the second (media adventitia) interface on the far wall of the common carotid artery in a section free of plaque beginning 10 mm below their bifurcations and including the bifurcations for 10 mm. For each subject, three measurements on both sides were performed, i.e., the anterior, lateral, and posterior projection of the near and far wall. Maximum (outside the plaque) rather than mean values of IMT were considered, and edge detection was performed manually. Carotid thickening was defined as an IMT≥1 mm. A carotid plaque was defined as focal thickening of ≥ 1.5 mm at the level of common carotid artery.

Measurement of IMT is currently used as intermediate outcome in clinical trials. Studies analysing reproducibility of IMT measurements reported that variability is lowest when determining the mean thickness in the common carotid artery [26]; that the reproducibility of IMT measurements in the common carotid artery is reliable even in patients with increased artery wall thickness [27]; and that [28-30] examinations by different sonographers in multicenter studies is feasible with a good interobserver agreement.

- Regarding the clinical meaning of IMT measurements, an increased IMT is an established predictor of subsequent coronary heart disease and stroke, the two leading causes of cardiovascular death [31], also providing additional prognostic information to that of conventional risk factors [32]. In this clinical cotext, an IMT≥1 mm has been associated with a higher risk of cardiovascular events [33,34].

Antiviral Therapy

All patients underwent DAA-based antiviral therapy. We included only patients with advanced chronic hepatitis or compensated cirrhosis because at the time of the enrolment the Italian Agency of the Drug (AIFA) did not allow treatment of patients with a milder stage of chronic liver disease due to HCV infection.

All patients were treated according to therapeutic schedules suggested by EASL/AISF guidelines available in the time of the enrollment [35,36].

All patients were tested at the baseline for HCV-RNA (real-time PCR COBAS TaqMan HCV Test v2.0 and Roche diagnostics, S.p.A Monza, Italy) and HCV genotype by Versant HCV Genotype 2.0 Assay LIPA, Siemens, Erlangen, Germany. HCVRNA was repeated after 4 weeks of therapy, at the end of therapy, and 12 weeks after stopping

treatment. Sustained virological response was defined as HCVRNA undetectable after 12 weeks from the end of antiviral therapy [35].

Statistics

Continuous variables were summarized as mean ± SD, and categorical variables as frequency and percentage. The Student's t-test and chi-square test were used when appropriate.

Multiple linear regression models were used to assess the relationship between IMT considered as continuous variable, and Λ-IMT (IMT 9-12 months after the end of antiviral therapy – IMT at baseline) with other clinical and biochemical parameters in the entire cohort of HCV-infected patients.

Multiple logistic regression models were used to assess the relationship of IMT≥10 mm and carotid plaques with other clinical and biochemical parameters in the entire cohort of HCV-infected patients. In these models, the dependent variable was the presence of carotid thickening, coded as 1=IMT≥1 mm vs. 0 = IMT<1 mm, and of carotid plaque, coded as 1 vs. 0 = absent. As candidate risk factors, we selected age, gender, BMI, baseline ALT, total cholesterol, blood glucose, diabetes, arterial hypertension, smoking, platelet count, cirrhosis, and Log HCV RNA. Λ BMI, Λ blood glucose, Λ cholesterol, Λ platelet count, and Λ ALT were added in the model predicting ΛIMT.

Variables associated with the dependent variable at univariate analysis (probability threshold, p≤ 0.10) were included in the multivariate regression models. Analyses were performed using SPSS package v.20 (SPSS Inc., Chicago, IL, USA).

RESULTS

Patient features

The baseline features of the 182 patients are shown in Table 1. Fifty-six percent were males, and mean age was 63 years. Fourteen percent of our patients had obesity, and 41.8% were hypertensive. Diabetes was present in about 20% of patients. Mean values for total cholesterol were within the normal range.

Most patients (83.4%) were infected by HCV genotype 1b. Regarding disease severity, 65.9% had compensated cirrhosis and one patient in five had platelet count <100,000 mmc.

Carotid atherosclerosis and risk factors at baseline

Mean IMT was 0.94±0.29 mm, and both carotid thickening and carotid plaques were found in 42.9% of patients. None of the study participants had clinically relevant (i.e., ≥60%) carotid stenosis.

Older age and lower platelet count were associated with higher IMT ($p<0.10$ for both), though only older age ($p<0.001$) was maintained as independent predictor at multiple linear regression analysis. Similarly, older age and platelet ($p<0.10$ for both) were associated with carotid thickening, and again only older age (OR 1.07, 95%CI 1.03–1.10, $p<0.001$) was the factor independently linked with this outcome at multivariate logistic regression analysis. Finally, when looking at carotid plaques as main outcome, older age, diabetes and lower platelet count ($p<0.10$ for all) were linked to their presence, older age (OR 1.04, 95%CI 1.01–1.08, $p=0.004$) and lower platelets (OR 1.00, 95%CI 1.00–1.01, $p=0.003$) being confirmed as risk factors at multivariate logistic regression analysis

Antiviral Therapy

One hundred one patients had received ombitasvir/paritaprevir/ritonavir and dasabuvir ± ribavirin for 12-24 weeks, 36 sofosbuvir and simeprevir ± ribavirin for 12 weeks, 33 sofosbuvir/ledipasvir ± ribavirin for 12-24 weeks, 9 sofosbuvir and ribavirin for 24 weeks, and 4 sofosbuvir and daclatasvir ± ribavirin for 12 weeks.

Carotid atherosclerosis changes after antiviral therapy

Mean IMT significantly decreased from baseline to 9-12 months after the end of antiviral therapy (0.94±0.29 mm vs 0.81±0.27, p<0.001) (Figure 1A). Consistently, we observed a significant reduction in the proportion of patients with carotid thickening (42.8% vs 17%, p<0.001) (Figure 1B), while no differences were reported for carotid plaques (42.8% vs 47.8%, p=0.34) (Figure 1C). We also observed a significant reduction in blood glucose (103.5±23.2 vs 93.6±20.1, p<0.001) and ALT (81.9±49.5 vs 22.6±11.3, p<0.001) levels, and an increase in cholesterol levels (160.0±29.5 vs 173.0±30.5, p<0.001) and PLT values (153.3±64.3 vs 169.4±65.8, p=0.01), while BMI (25.6±3.7 mm vs 25.9±3.9, p=0.45) did not change. The increase in cholesterol levels was present in both cirrhotic (157.0±28.7 vs 168.7±30.5, p=0.005) and no cirrhotic (165.6±30.3 vs 180.2±29.7, p=0.01) patients. The improvement in blood glucose levels was observed in patients without (95.7±13.0 vs 88.6±27.2, p<0.001) and with (134.7±29.0 vs 90.7±32.4, p<0.001) diabetes; some diabetic patients reduced the need of insulin and/or oral antidiabetic drugs.

Notably, when looking at factors associated with Λ IMT change neither baseline parameters, neither changes in BMI, blood glucose, cholesterol, ALT and PLT levels were found to be significantly associated (p>0.10 for all). Consistent with these results, the improvement in IMT and carotid thickening was largely confirmed across sub-groups of

patients discriminated according to cardiovascular risk factors and liver disease severity (Table 2). Specifically, IMT and carotid thickening significantly improved in patients younger or older than 65 years, nonobese, smokers and not smokers, with and without diabetes, with and without arterial hypertension, and with and without hypercholesterolemia (Table 2). Similar results were obtained according to liver disease severity in patients with advanced fibrosis and cirrhosis, and in those with PLT values <100,000 mmc and ≥100,000 mmc (Table 2). The only subgroup in which we observed a non significant trend for IMT and carotid thickening improvement was the cohort of obese patients (IMT 0.94±0.37 vs 0.87±0.31, p=0.44; IMT≥10 mm 30.7% vs 23%, p=0.53). However the strength of this analysis is limited by the small number of obese patients (n=26).

Notably, in the cohort of 76 HCV-infected patients with advanced fibrosis/compensated cirrhosis (mean age 64 years, males 48%, mean BMI 25.6 Kg/m^2, mean cholesterol 160mg/dL, 9.2% with diabetes, 43% with arterial hypertension, 60% smokers) not underwent antiviral therapy, we did not observe any significant change in IMT (0.88±0.30 vs 0.94±0.40, p=0.29)

DISCUSSION

Available evidence, even if sometimes contrasting, suggest that HCV infection is associated with cardiovascular alterations including carotid atherosclerosis [4-15], and that virological clearance, either spontaneous or mostly obtained with IFN-based regimens, may improve cardiovascular outcomes [18-24]. In our cohort of HCV patients with severe fibrosis and, in two-thirds of cases, compensated cirrhosis, HCV eradication by DAA improved carotid atherosclerosis in terms of reduction of both IMT and carotid thickening.

Notably, when patients were stratified for cardiovascular risk factors and liver disease severity, this improvement was confirmed in all subgroups except in obese patients.

Specifically, we found that SVR by DAA leads to reduction in IMT and in the prevalence of patients with carotid thickening at risk for cerebro-cardiovascular events [33,34], while no changes were observed in untreated patients. To the best of our knowledge, this is one of the first evidences demonstrating a beneficial impact of virological eradication by DAA on cardiovascular alterations. Cohort studies reporting and improvement in myocardial damage, cardiovascular events and cardiovascular mortality were mostly done in high selected populations eligible to and undergone IFN-based therapies [18-21]. Consistently, it is not clear whether the observed positive effect was due to virological clearance, or it is driven by IFN or by selection bias. Only two recent studies assessed the impact of DAA on cardiovascular outcomes [22-24]. The first, is a large prospective study on a cohort of patients with compensated HCV-related cirrhosis who underwent IFN or DAA-based antiviral therapies, and where SVR was associated with a significant reduction on cardiovascular events: however this study did not split the results for type of therapeutic regimen (IFN or DAA) [22,23]. The second is an unpublished retrospective analysis on a large cohort of US patients, where treatment with DAA compared to no treatment was associated with reduction in the incidence of both cardiovascular and cerebrovascular events [24].

The present study included a cohort of HCV-infected patients, about half of them older 65 years, with a significant prevalence of cardiovascular risk factors, and where age and PLT values –as already published [6]- where major risk factors for atherosclerosis. This picture allowed us to assess the impact of SVR on carotid atherosclerosis in patients with or without cardiometabolic alterations. Notably, we did not identify any baseline and/or

dynamic predictor of IMT improvement. Consistent with these data we found that the beneficial impact of HCV eradication on carotid atherosclerosis was largely maintained in patients with or without cardiovascular risk factors like, diabetes, arterial hypertension, hypercholesterolemia and smoking, the strength of these analyses being sometimes affected by the small sample of some subgroups. Along this line, in the small group of obese patients, we only found a non nonsignifcant trend for carotid atherosclerosis improvement. While a recent meta-analysis suggests that the impact of HCV on cardiovascular damage could be more pronounced in patients at higher cardiometabolic risk [17], the present study is the first that stratified the impact of HCV eradication on carotid atherosclerosis according to cardiometabolic risk factors,.

In our study, we also found that the effect of SVR on carotid atherosclerosis was maintained independently of the severity of liver damage –cirrhosis vs advanced fibrosis and lower vs higher platelet values- Concerning this topic, available results on cohorts of patients who underwent IFN-based therapies are very controversial, ranging from no impact of liver fibrosis to an improvement in cardiometabolic prognosis [37], to a more pronounced effect in patients with mild disease [20] or in patients with advanced fibrosis/cirrhosis [22,38,39]. Notably, the before quoted retrospective analyses of a large cohort of US patients, reported that DAA therapy reduced incidence of both cardio- and cerebrovascular events in patients with cirrhosis, while reducing the risk of only cardiovascular events in patients without cirrhosis [24]. Differences in baseline characteristics of patients, in the prevalence of cardiovascular risk factors, in the evaluated cardiovascular outcomes and in the type of antiviral regimens could explain the contrasting results.

The present study also observed that HCV eradication by DAA leaded to an improvement in blood glucose levels and to an increase on serum cholesterol levels. These data agree with results already reported on cohorts of HCV patients who underwent IFN-based or DAA-based regimens [40-43], and find explanation on the ability of the virus to interfere with cellular glucose and lipid metabolism [44].

While reporting an improvement in IMT, we did not observe any effect of virological eradication on carotid plaques. This issue could stem by the fact that, in a short-medium term scenario, IMT could be more sensitive that a stable plaque to changes in inflammatory and fibrogenic mediators related to HCV infection. HCV in fact can lead to increased cardiovascular risk by leading to insulin resistance [45], by inducing a systemic inflammatory status via NK and TH1-mediated responses [46], via increase in TNFalpha and IL-6 levels [47,48] and reduction in adiponectin levels [49], and via endothelial damage directly related to the HCV infection [50,51]. However, we cannot exclude a potential effect of virological eradication on carotid plaques in a longer follow-up.

The present study has some limitations. First, inter-observer concordance of carotid atherosclerosis examinations was not assessed, this issue potentially affecting the interpretation of our results. However, first, all tests were performed by expert operators following the same protocol; second, different relevant studies assessing carotid atherosclerosis were based on multicenter cohorts and/or on multiple operators [29,30], third, different studies reported good interobserver concordance for carotid atherosclerosis assessment by ultrasound [29,30]; fourth, our group [52] recently reported that interobserver agreement for carotid atherosclerosis evaluation is overall good among the groups involved in this study. Another limitation is the lack of data on patients with mild liver fibrosis limiting the application of our data to HCV-infected individuals with advanced

fibrosis/compensated cirrhosis. This is due to the availability in Italy, when we started the study, of treatment with DAA only for patients with advanced fibrosis/cirrhosis. However, we are confident to observe a similar beneficial cardiovascular effect of SVR in patients with mild liver fibrosis, as also suggested by recently presented unpublished retrospective data [24]. Finally, the absence of data about main cardiovascular outcomes and long-term follow-up further limit the clinical interpretation of our results.

In conclusion, in patients with advanced fibrosis/compensated cirrhosis due to HCV infection, SVR by DAA improves carotid atherosclerosis, this effect being largely confirmed after stratification for cardiovascular risk factors and for liver disease severity. Further studies are needed to evaluate the long-term clinical meaning of these data.

Figure Legends

Figure 1. Differences in Intima-Media Thickness (Student's t-test) (A) and in the prevalence of Carotid Thickening (chi-square test) (B) and Carotid Plaques (chi-square test) (C) in patients with advanced fibrosis/compensated cirrhosis due to HCV infection at baseline and 9-12 months after the end of direct antiviral agent-based therapy.

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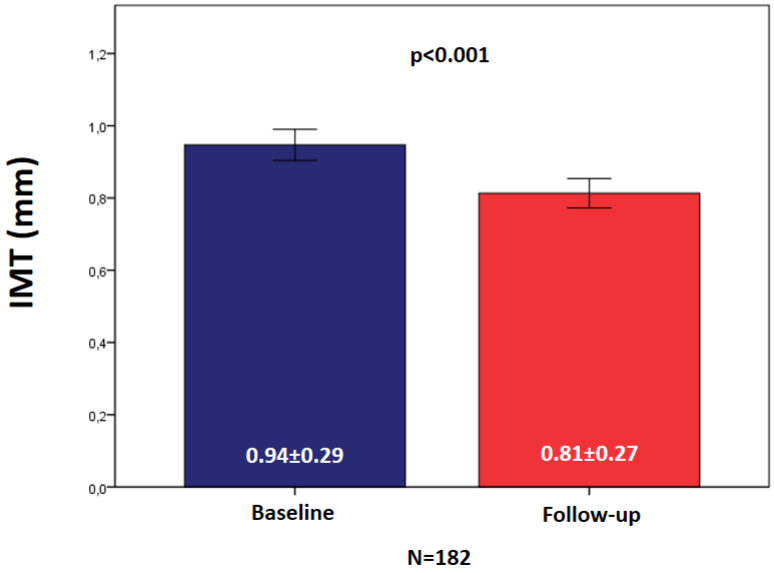


Figure 1A

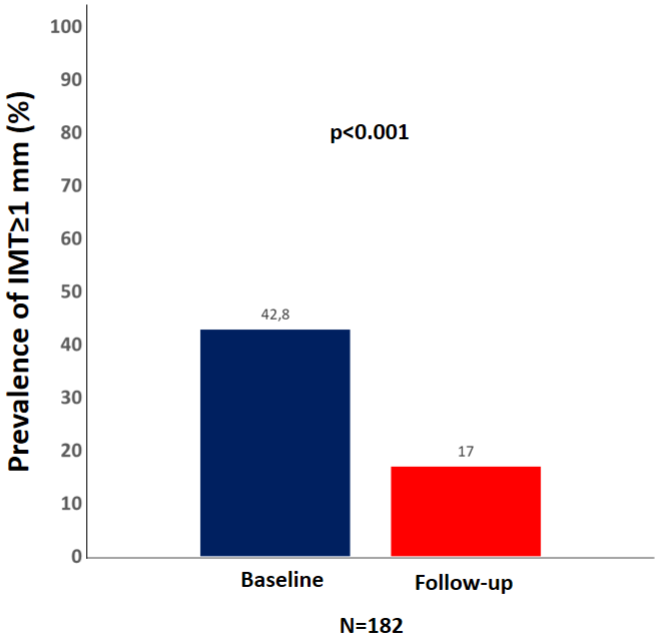


Figure 1B

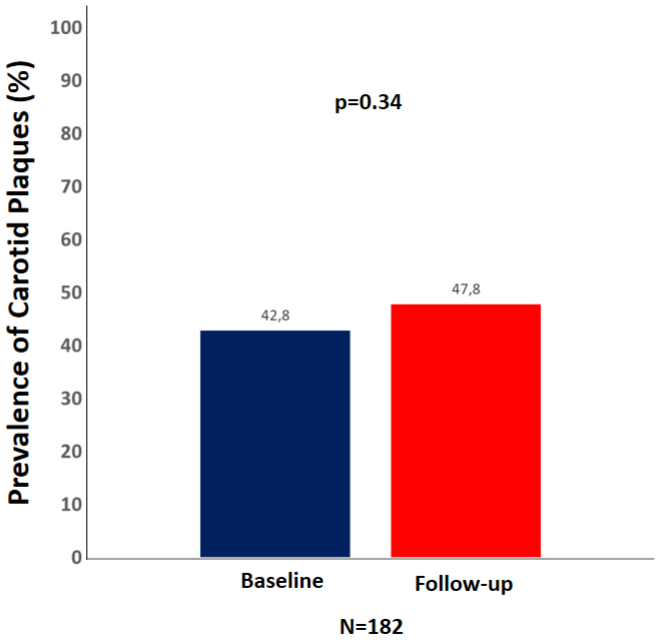


Figure 1C

Table 1. Characteristics of 182 Patients with Advanced Fibrosis/Compensated Cirrhosis due to HCV Infection.

Variable	Chronic Hepatitis C with Advanced Fibrosis/Cirrhosis (N=182)
Male Gender - %	56
Age – years	63.1±10.4
Age > 65 years	47.3
Body Mass Index – Kg/m^2	25.6±3.7
BMI≥30 Kg/m2	14.3
Blood Glucose – mg/dL	103.5±23.2
Type 2 Diabetes - %	19.8
Arterial Hypertension - %	41.8
Total Cholesterol – mg/dL	159.9±29.5
Smoking - %	35.2
IMT – mm	0.94±0.29
IMT≥1 mm - %	42.9
Carotid Plaques - %	42.9
ALT – IU	81.9±49.5
PLT – 10^3 mmc	153.3±64.3
HCV Genotype 1/1a/1b/2/3/4	0.5/8.2/83.4/1.1/2.2/1.6
HCVRNA –Log	5.8±0.6
Cirrhosis - %	65.9

Abbreviations: BMI, body mass index; IMT, intima-media thickness, ALT, alanineaminotransferase; PLT, platelet; HCV, hepatitis C virus; HCVRNA, hepatiti C virus ribonucleid acid.

Table. Changes from baseline to follow-up in intima-media thickness and in the prevalence of carotid thickening (IMT≥1 mm) and carotid plaques in subgroups of patients discriminated according to cardiovascular risk factors and liver disease severity.

	Age ≤65 (n=96)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.85±0.26	0.73±0.23	0.001
IMT ≥1 mm - %	30.2%	11.4%	0.001
Carotid Plaques - %	37.5%	43.7%	0.38
	Age >65 (n=86)		
	Baseline	**Follow-up**	**P value**
IMT - mm	1.04±0.29	0.89±0.29	0.001
IMT ≥1 mm - %	56.9%	23.2%	<0.001
Carotid Plaques - %	48.8%	52.3%	0.64
	BMI ≤30 (n=156)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.94±0.27	0.80±0.27	<0.001
IMT ≥1 mm - %	44.8%	16%	<0.001
Carotid Plaques - %	41.6%	46.7%	0.36
	BMI >30 (n=26)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.94±0.37	0.87±0.31	0.44
IMT ≥1 mm - %	30.7%	23.0%	0.53
Carotid Plaques - %	50.0%	53.8%	0.78
	Smoking (n=64)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.96±0.33	0.84±0.33	0.03
IMT ≥1 mm - %	46.8%	21.8%	0.003
Carotid Plaques - %	48.4%	54.6%	0.47
	No Smoking (n=118)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.93±0.26	0.79±0.24	<0.001
IMT ≥1 mm - %	40.6%	14.4%	<0.001
Carotid Plaques - %	39.8%	44.1%	0.51

	Diabetes (n=36)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.97±0.27	0.87±0.29	0.11
IMT ≥1 mm - %	52.7%	22.2%	0.007
Carotid Plaques - %	58.3%	58.3%	1.00
	No Diabetes (n=146)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.93±0.29	0.83±0.30	0.01
IMT ≥1 mm - %	40.4%	15.7%	<0.001
Carotid Plaques - %	39.0%	45.2%	0.28
	Arterial Hypertension (n=76)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.95±0.28	0.80±0.20	0.24
IMT ≥1 mm - %	44.7%	21.1%	0.002
Carotid Plaques - %	48.6%	55.2%	0.41
	No Arterial Hypertension (n=106)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.94±0.30	0.79±0.25	<0.001
IMT ≥1 mm - %	41.5%	14.1%	<0.001
Carotid Plaques - %	38.7%	42.4%	0.57
	Hypercholesterolemia (n=18)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.86±0.21	0.74±0.17	0.06
IMT ≥1 mm - %	33.3%	5.5%	0.03
Carotid Plaques - %	50%	61.1%	0.50
	No Hypercholesterolemia (n=152)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.97±0.30	0.82±0.29	<0.001
IMT ≥1 mm - %	46.7%	18.4%	<0.001
Carotid Plaques - %	41.4%	46%	0.41
	PLT <100,000 (n=35)		
	Baseline	**Follow-up**	**P value**
IMT - mm	1.01±0.41	0.85±0.40	0.10
IMT ≥1 mm - %	48.5%	20%	0.01
Carotid Plaques - %	28.5%	40%	0.31
	PLT ≥100,000 (n=147)		

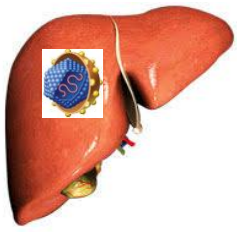
	Baseline	Follow-up	P value
IMT - mm	0.93±0.25	0.80±0.23	<0.001
IMT ≥1 mm - %	41.4%	16.3%	<0.001
Carotid Plaques - %	46.2%	51.4%	0.55
	No Cirrhosis (n=62)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.90±0.24	0.78±0.22	0.006
IMT ≥1 mm - %	41.9%	9.6%	<0.001
Carotid Plaques - %	43.5%	48.3%	0.59
	Cirrhosis (n=120)		
	Baseline	**Follow-up**	**P value**
IMT - mm	0.97±0.31	0.82±0.30	<0.001
IMT ≥1 mm - %	43.3%	20.8%	<0.001
Carotid Plaques - %	42.5%	47.5%	0.43

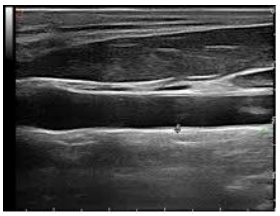
Abbreviations: IMT, intima-media thickness

P value by Student's t-test for IMT, and by chi-square test for IMT≥1 mm and carotid plaques.

Highlights

- *HCV eradication by direct antiviral agents improves carotid atherosclerosis*
- *Atherosclerosis improvement is confirmed after stratification for cardiovascular risk factors*
- *Atherosclerosis improvement is observed in patients with and without cirrhosis*





Ultrasonographic Assesment of Intima-media Thickness and Carotid Tickening in Patients with Advanced Fibrosis/Compensated Cirrhossi due to HCV Infection:

Impact of SVR by DAA