Significant improvement of glycemic control in diabetic patients with HCV infection responding to direct-acting antiviral agents

Research Article

SIGNIFICANT IMPROVEMENT OF GLYCAEMIC CONTROL IN DIABETIC PATIENTS WITH HCV INFECTION RESPONDING TO DIRECT-ACTING ANTIVIRAL AGENTS†

Short title: HCV eradication with DAAs improves diabetes

Alessia Ciancio[1], Roberta Bosio[2], Simona Bo[3], Marianna Pellegrini[3], Marco Sacco[1], Edoardo Vogliotti[1], Giulia Fassio[2], Andrea Guido Franz Bianco Mauthe Degerfeld[2], Monica Gallo[2], Chiara Giordanino[2], Lodovico Terzi di Bergamo[1],Davide Ribaldone[1], Elisabetta Bugianesi[1], Antonina Smedile[1], Mario Rizzetto[1],

Giorgio Maria Saracco[1]

[1]Gastroenterology Unit, Department of MedicalSciences, AOU Città della Salute e della Scienza di Torino, University of Turin, Torino, Italy

[2]Gastroenterology Unit, San Luigi Hospital, University of Turin,Torino, Italy

[3]Department of MedicalSciences, AOU Città della Salute e della Scienza di Torino, University of Turin, Torino, Italy

Corresponding author:

Giorgio Maria Saracco, MD, Gastro-hepatology Unit, Department of Medical Sciences,

AOU Città della Salute e della Scienza di Torino, Corso Bramante 8810126 Torino, Italy.

Tel.: +39-011-6336397.FAX: +39-011-6335927.

E-mail: giorgiomaria.saracco@unito.it

Conflicts of interest: none declared.

Source of funding: none declared.

Received 14 August 2017; Revised 19 September 2017; Accepted 19 September 2017

Journal of Medical Virology

DOI 10.1002/jmv.24954

ABSTRACT

*AIM:*Many studies showed insulin resistance amelioration in HCV-patients achieving Sustained Virologic Response (SVR) but results on glycaemic control in diabetic patients are unclear.

This study aimed to assess fasting glucose (FG) and glycated haemoglobin (HbA1c) values before and after therapy with direct-acting antivirals (DAAs) in HCV-patients with type 2 diabetes mellitus (T2DM).

*METHODS:*Of the 122 consecutively recruited patients with chronic hepatitis C and T2DM, 110 patients were treated with DAAs and 12 remained untreated. Clinical, biochemical, virological and metabolic features were collected both at baseline and at 12 weeks after the end of therapy (EOT) or after a comparable period of time in untreated patients.

*RESULTS:*101 patients obtained a SVR (Group 1), while nine were relapsers; Group 2 (21 patients) was composed by the 9 relapsers and the 12 untreated patients. A significant reduction of mean FG (134.3±41.32 mg/dL vs 152.4± 56.40 mg/dL, p= 0.002) and HbA1c values (46.51±16.15 mmol/mol vs 52.15±15.43 mmol/mol,p< 0.001) was found in Group 1 but not in Group 2 (140.6±47.87 mg/dL vs 145.31±30.18 mg/dL, p=0.707, and 55.31±20.58 mmol/mol vs 53.38±9.49 mmol/mol, p= 0.780).

In Group 1, 20.7% of patients could reduce or suspend their antidiabetic therapy compared to none in Group 2 (p= 0.03), despite the significant weight increase observed in Group 1.

*CONCLUSIONS***:** SVR induced a significant amelioration of glycaemic control in diabetic HCV-patients, despite a significant weight increase; larger prospective studies are needed to verify whether these results are maintained over the longterm.

Key words: Anti-hepatitis C virus DAA; Hepatitis C virus; Hepatitis virus

List of abbreviations in the order of appearance:

HCV, hepatitis C virus; T2DM, type 2 diabetes mellitus; CHC, chronic hepatitis C; HCC, hepatocellular carcinoma; IR, insulin resistance; IFN, interferon; SVR, sustained virologic response; DAAs, direct-acting antivirals; FG, fasting glucose; HbA1c, glycated haemoglobin; OLT, orthotopic liver transplant; INHS, Italian national health system; EASL; European Association for the Study of the Liver; AH, arterial hypertension; MS, metabolic syndrome; BMI, body mass index; WC, waist circumference; HC, hip circumference; WHR, Waist-to-Hip Ratio; *IL28B, Interleukin 28B;* AST, aspartate aminotransferase; ALT, alanine aminotransferase; APH, alkaline phosphatase; GGT, gamma glutamyltranspeptidase; TE, transient elastography; US, ultrasound scan; KPa, kilopascal; IQR, interquartile range; CAD, coronary artery disease; ECG, electrocardiograms; OR, odds ratio;CI, confidence interval; RBV, ribavirin; IRS, insulin receptors; SOC, suppressor of cytokines; UKPDS, UK Prospective Diabetes Study.

INTRODUCTION

A burden of data suggests that Hepatitis C Virus (HCV) can impair insulin signalling and boost the onset of type 2 diabetes mellitus (T2DM) beyond and in addition to the histological severity of the associated liver disease[1-3]. The increased metabolic risk associated with HCV infection is supported either by cross-sectional and longitudinal studies: the prevalence of T2DM in chronic hepatitis C (CHC) ranges between 19%-33%[4,5] and its incidence is increased in patients with CHC after liver transplantation[6,7]. In turn, T2DM in CHC seems not only to accelerate the progression of liver disease[8-10] but also to greatly increase the risk of hepatocellular carcinoma (HCC)[11-13], even in patients without cirrhosis and after the eradication of HCV infection[14]. For this reason, reducing the incidence of T2DM and improving the glycaemic control of diabetic patients with CHC are of paramount importance. If HCV is directly involved in the development of insulin resistance (IR) and T2DM, its clearance should result in a parallel decrease in the risk of DM incidence. Accordingly, a successful eradication of HCV could improve IR, glycaemic control and clinical outcomes in patients with established T2DM.

In the era of interferon (IFN)-based therapy, this hypothesis was very difficult to be tested due to the very low Sustained Virologic Response (SVR) rates associated to IR and T2DM[15-18] for the negative impact of metabolic abnormalities on therapeutic efficacy. Nevertheless, some clinical trials[19,20] concurred to demonstrate that SVR was associated with improved IR. On the contrary, baseline IR and T2DM do not seem to affect the outcomes of Direct-Acting Antivirals (DAAs)-based therapy[21-23] and the high therapeutic efficacy of novel antivirals will ensure viral eradication in a large number of

diabetic patients. Recent preliminary reports suggest that DAA agents are associated with improvement of fasting glucose (FG) and glycated haemoglobin (HbA1c) values during[24] and after therapy[25-27] in diabetic patients. Although the results of these studies suggest benefits of DAAs on glycemic control, their anecdotal format and/or retrospective design make questionable a generalization of their conclusions.

In order to assess whether SVR has a clinical impact on glycaemic control, we prospectively recruited HCV-positive patients with T2DM receiving DAAs and evaluated their FG andHbA1c levels at baseline and 12 weeks post-treatment; the influence of SVR on antihyperglycaemic medication needs was also assessed. Potentially confounding factors affecting the results (i.e. weight variation) were considered.

METHODS

All the consecutive patients with CHC and T2DM referred to two academic centers of North-Western Italy (Gastrohepatologic Clinic of Molinette Hospital, Turin, Italy and Gastroenterology Division of San Luigi Hospital, Orbassano, Italy) between January and December 2015 were considered. Patients were included into the present study if they fulfilled the following criteria at baseline: age > 18 years; positive results for HCV-RNA by polymerase chain reaction (PCR); CHC at transient elastography; T2DM. Exclusion criteria were: lack of written informed consent, patients in waiting list for orthotopic liver transplant (OLT), post-OLT patients, Child-Pugh C patients, active alcohol intake, presence of ascites, presence of other concomitant liver diseases such as

haemochromatosis, Wilson's disease, drug-related liver disease, autoimmune hepatitis, HbsAg carriership, HIV infection, primary biliary cirrhosis, 1-antitrypsin deficiency.

In 2015, due to Italian National Health System (INHS) rules, only patients with advanced liver fibrosis (F3-F4 according to METAVIR Score[28]) or with severe extrahepatic diseases linked to C virus could be reimbursed for treatment with DAAs; for this reason, a significant minority of diabetic patients with CHC remained untreated: each of them was asked to participate into the study as controls. Out of remaining treatable patients, three refused to be treated but accepted to serve as controls.

Overall, 156 diabetic patients were screened and 122 (78.2%) were included into the study. Patients' flow is reported on Figure 1. One hundred and ten patients were treated with DAAs for 12-24 weeks (Table 1) according to the European Association for the Study of the Liver (EASL) guidelines[29], 101 (91.8%) showed a SVR (HCV-RNA clearance 12 weeks after the end of therapy); 9 of 110 (8.2%) demonstrated a viremic relapse 4 weeks after the end of treatment and remained HCV-RNA positive at week 12**.** Twelve patients were untreated and served as controls for a comparable period of time. Overall, 101 patients who achieved SVR were allocated to Group 1 and 21 who did not obtain SVR or were untreated were attributed to Group 2.

At baseline, a complete medical history and physical examination was undertaken and the following data were obtained: age, gender, smoking status,family history for T2DM, duration of T2DM, type of antidiabetic medication,presence of macro/ microangiopathic complications, arterial hypertension (AH), relevant co-morbidities and the metabolic syndrome (MS),body mass index (BMI), waist circumference (WC), hip circumference (HC),waist/hip ratio (WHR), HCV genotype, *Interleukin-28B*rs12979860 (*IL-28B*)

status. Serum samples were drawn after an overnight (12-h) fasting period in order to assess the viral load (AmpliPrep®/COBAS Taqman® HCV test, Roche Diagnostics, Basel, Switzerland), complete blood count, routine liver biochemistry (alanine aminotransferase [AST], aspartate aminotransferase [AST] levels, total bilirubin, albumin, alkaline phosphatase [APH], gamma glutamyltranspeptidase [GGT]),FG and HbA1c (high-performance liquid chromatography, HPLC G8 Analyzer, Thoso, USA, normal values: 20-38 mmol/mol) values. Components of the MS, including central obesity, hypertriglyceridemia, hypertension and low HDL cholesterol, were recorded.

The severity of liver fibrosis was determined within 3 months from treatment by transient elastography (TE) and patients were stratified according to METAVIR Scale[28]. An abdominal ultrasound (US) was performed in each patient at baseline and at the end of the study (EOS). Baseline and EOS aliquoted plasma samples were stored at -80° C until assayed. Plasma insulin concentrations were assessed in patients who had never received antihyperglycemic agents by a double antibody radioimmunoassay (Diagnostic Products Corporation, Los Angeles, CA; interassay coefficient of variation <13%). IR was calculated on the basis of fasting glucose and insulin levels according to the homeostatic assessment of insulin resistance (HOMA-IR) method[30].

After 12 weeks from the end of therapy (or after a comparable period of time in untreated patients), the anthropometric features and the abovementioned serum measurements were re-assessed in each patient.

The study was performed in accordance with the principles of the Declaration of Helsinki and approved by the local ethics committee (Comitato Etico Interaziendale Città della Salute e della Scienza di Torino, Turin, Italy; Comitato Etico AOU S. Luigi

Gonzaga di Orbassano, Orbassano, Italy); written informed consent was obtained from all patients.

Definitions

Smokers were defined as persons smoking more than one cigarette per day or known to have smoked within three years before the study recruitment. BMI was calculated as weight in kilograms divided by the square of the height in meters with the subjects wearing light clothing but no shoes. WC was measured at the midpoint between the lower border of the rib cage and the iliac crest. HC was measured at the widest point between hip and buttocks. WHR was defined as WC (cm) divided by HC (cm). Blood pressure was reported as the average of the last three determinations. Hypertension was defined as a systolic or diastolic blood pressure of 130/80 mmHg or higher or a current anti-hypertensive treatment[31].

According to the criteria recommended by the Expert Committee on the Diagnosis and Classification of Diabetes Mellitus, diabetes was defined as fasting plasma glucose ≥ 126 mg/dl (7 mmol/l) in two separate measurements[32].

Diagnosis of metabolic syndrome was made according to the criteria suggested by the International Diabetes Federation[33].

Liver stiffness evaluation

Liver stiffness measurements were performed within 3 months from the start of the study using TE (FibroScan®, Echosens, Paris, France) according to current EASL

guidelines[34]; the validated probe according to BMI was used (M probe if BMI<28 kg/m^2, XL probe if BMI >28 kg/m^2). Results were recorded in kilopascal (KPa); patients were fasting for a minimum of 6 hours prior to TE. Values for each patient were calculated as median of a minimum of 10 consecutive measurements and exams with an interquartile range (IQR) divided by mean value (IQR/M) of >30% or a success rate of 60% were excluded as recommended by current guidelines. Patients were subsequently divided into 4 groups (F0-F1, F2, F3, F4) according to the METAVIR Scale[28]. Patients with cirrhosis underwent upper endoscopy within 3 months from the study inclusion in order to verify the presence of oesophageal/gastric varices.

Diabetic complications

Coronary artery disease (CAD) was assessed by a conventional 12-lead resting electrocardiograms (ECG) interpreted by two trained cardiologists and/or presence of documented events, recorded by a physician (i.e. angina, previous myocardial infarction, coronary artery bypass graft or other invasive procedures to treat CAD); peripheral artery disease was defined when conditions such as ischaemic foot ulcers, gangrene, amputation, vascular surgery, transient ischaemic attacks, strokes, intermittent claudication, absent foot pulses, abnormal brachial and posterior tibial blood pressure using Doppler techniques were reported. Retinopathy was diagnosed by ophthalmoscopy examination performed by an ophthalmologist experienced in diabetic retinopathy. Nephropathy was established by the presence of an albumin excretion rate higher than 20 µg/min at least in two of three urine collections. Distal symmetric polyneuropathy was diagnosed by the presence of neuropathic symptoms, an abnormal

vibration perception threshold, the absence of ≥2 ankle or knee reflexes and/or an abnormal electromyographic test. Autonomic neuropathy was diagnosed by a loss of heart rate variability or postural hypotension.

Statistical analysis

Patients' characteristics at baseline and at the end of follow-up were compared between groups using the Student *t* test for continuous variables and the chi-square (or Fisher's Exact Test), when expected cell frequencies were less than 5 for categorical variables. In particular, weight, waist and hips circumference and biochemical changes were analyzed by paired t-test for the interval between baseline to 12 weeks after the end of therapy (up to 252 days from baseline). Association between predictor variables and glycaemic control improvement were determined by Odds Ratio (OR) and 95% confidence interval (CI) calculated using Cox proportional hazards regression. Variables with $p< 0.1$ in the univariate model were included in the multivariate model.

RESULTS

All patients included in the study protocol were Caucasian; Table 2 summarizes baseline demographic, anthropometric and clinical characteristics of patients who achieved SVR (Group 1) and those who did not or who had not been treated (Group 2). Baseline liver-related and diabetes-related features are reported in Table 3. Age, gender, BMI, abdominal circumferences, hypertension and metabolic syndrome rates,

comorbidities, viral genotypes and viremia, IL-28B status, GGT levels were not significantly different between the two groups. Not surprisingly, Group 2 patients had significantly lower liver stiffness (p= 0.024) as 57% of them could not have access to therapy due to fibrosis lower than F3, according to current INHS rules. No significant differences regarding Child-Pugh status, previous HCC and presence of gastro-oesophageal varices were found between patients with cirrhosis from both groups.

Values of FG and HbA1c levels, antidiabetic medications and prevalence of macro/microangiopathic complications were comparable between the two groups (Table 3).

At the end of the study, Group 1 patients showed a statistically significant decrement of FG (134.3±41.32 mg/dL, p= 0.002) and HbA1c (46.51±16.15 mmol/mol, p < 0.001) levels; among Group 2 patients, no significant variation in FG (140.6±47.87 mg/dL, p = 0.707) and HbA1c (55.31±20.58 mmol/mol, p = 0.780) values was found. Liver function and T2DM parameters of both groups before and at the end of the study are reported in Table 4.

To verify whether ribavirin (RBV) might have influenced post-treatment HbA1c values through its hemolytic effect, we analyzed treated patients who achieved SVR according to the use of the medication: a significant decrease of HbA1c levels was observed both in 74 patients receiving RBV (p= 0.024) and in 27 patients not given RBV (p= 0.007).

Finally, in order to individuate predictive factors clinically relevant for glycaemic amelioration (defined as a ≥20% decrease of HbA1c levels), we used a Cox proportional hazard risk model (Table 5). Based on a multivariable regression analysis, SVR was the

only independent predictive factor associated with the improvement of glycaemic control (OR= 49.75, 95% CI 2.197-112.67, p= 0.014).

Weight and antidiabetic treatment changes

Among Group 1 patients, the mean weight change from baseline to SVR was + 2.61±6.13 kg (p= 0.023); no significant weight variation was observed in Group 2 patients (+ 0.26±1.34 kg, p= 0.56). Baseline to SVR, 63 of 101 (62.3%) sustained responders gained weight (mean gain 4.13±3.12 kg), 9 (8.9%) lost weight (mean loss 3.2±2.43 kg) and 29 (28.8%) maintained a stable weight compared to 6 (28.6%, mean gain 2.34±1.86 kg), 4 (19%, mean loss 1.85±1.34kg) and 11 (52.4%) respectively ofGroup 2 patients (p= 0.02). Group 1 patients with or without significant improvement of glycaemic control (≥20% decrease of HbA1c levels) were then stratified according to the weight variation: 18 (28.5%) out of 63 patients showing weight increase achieved a significant amelioration of HbA1c values compared to 16 (42.1%, p= 0.074) of 38 patients demonstrating either weight decrease or stable weight.

Antihyperglycaemic agents tapering off because of improved glycaemic control was exclusively observed in Group 1, in 21 (20.7%) of the 101 patients who achieved SVR (p= 0.03); in particular, 8 of 19 (42.1%) patients treated by oral hypoglicaemic drugs reduced or suspended their therapy and 13 of 46 (28.2%) patients on insulin therapy decreased the dosage or withdrew the treatment, despite their mean weight gain of 2.85±4.33 kg.

Nine (8.9%) patients had to escalate their antihyperglicaemic medications compared to 1 (4.8%) of Group 2 (p= 0.34); this subset of patients showed a mean weight increase of 5.8±3.12 kg.

DISCUSSION

In our study, clearance of HCV induced a significant improvement in glycaemic control among 101 diabetic patients, as demonstrated by the reduction of FG and HbA1c levels 12 weeks after the end of treatment with DAAs. This result was obtained despite the weight gain observed in more than 60% of cured patients, suggesting that improved glycaemic control was not due to lifestyle change. The weight increase found in our patients was surprising but not unexpected as it was already described by Azad and coll.[35] who reported a significantly higher mean weight gain in diabetic patients compared to non-diabetics treated with sofosbuvir + ledipasvir (+5.8±7.2 pounds versus 1.9±7.8 pounds, p= 0.03). The reason for the weight increase observed among diabetic patients with SVR is still unexplained; we can speculate that the awareness of being definetively cured for HCV might have induced a less stringent nutritional compliance in the responders. Our multivariate analysis did not reveal confounders in the association between SVR and glycaemic control improvement, providing indirect support for a causal role of HCV in the induction of T2DM. The way whereby HCV induces T2DM is IR[3]; HCV was shown to impair the hepatocyte insulin signalling pathway by several ways[36], including the stimulus to the production of tumour necrosis factor-α, the serine phosphorylation of the insulin receptors (IRS), the over-expression

of the suppressor of cytokines (SOC-3)[37,38] and the induction of SOC-7[39]. The mechanism through which DAAs therapy ± RBV ameliorates IR has not been fully established but it is most likely mediated via viral clearance, rather than a direct pharmacological effect. To date, many reports[15,19,20,40] showed that HCV clearance is associated with a significant improvement of IR but our study has demonstrated that this amelioration translates into a definite clinical benefit for diabetic patients. In fact, the improved glycaemic control had a relevant impact on antihyperglycemic medication needs in patients who achieved SVR; if we exclude patients on diet only, more than 30% of diabetics decreased the dosage or withdrew the treatment. On the other hand, 9 (8.9%) patients showing a particularly relevant weight gain (+ 5.8±3.12 kg) had to escalate their anti-diabetic therapy despite the viral clearance; for this reason, the eradication of HCV in diabetic patients should not preclude proper counselling on diet and physical activity continuation.

If confirmed over the longterm, improvement of glycaemic control might add other benefits to diabetics with advanced liver disease who achieved SVR. First, good glycaemic control prevents the onset and progression of acute and long-term diabetes-related complications and, conversely, poor metabolic control could enhance or accelerate diabetes-related events. This is what usually occurs in ordinary HCV-negative diabetic patients where long-term tight glycemic control is difficult to achieve, as demonstrated in the UK Prospective Diabetes Study (UKPDS), showing an inexorable decline in glucose control over time[41].

It is reasonable to hypothesize that a successful eradication of HCV would improve clinical outcomes in patients with T2DM as reported by a recent study[42] showing that in diabetic patients with CHC treated with IFN/RBV and followed up for more than 7

years, none of those who obtained SVR developed retinopathy and/or neuropathic symptoms.

Secondly, T2DM remains a strong risk factor for HCC development in cirrhotics after SVR has been obtained[43,44] but a good glycaemic control seems to reduce the incidence of HCC[45,46]. Third, pre-OLT diabetes is associated with increased incidence of post-OLT mortality[47]; a recent study[48] showed that good glycaemic control pre-operatively may protect against the harmful effect of T2DM on infection and is associated with a reduced risk of readmission to intensive care unit and a reduced length of stay in hospital.

Our findings have some limitations: although statistically significant, our results are issued from a study including a relatively small and selected population of patients with HCV infection and mild T2DM and need to be confirmed by larger studies, including patients with severe T2DM of long duration. Moreover, before drawing definite conclusions, our study needs a longer follow up period in order to assess whether the results obtained by SVR regarding glycaemic control are maintained over the longterm and – more important – if this improvement will have an impact on T2DM outcome. It is conceivable that among CHC patients, T2DM natural history may be influenced not only by viral clearance but also by genetic and lifestyle-related aspects, such as dietary habits, physical activity and adherence to therapy. We still need to assess if viral eradication is "per se" a protection from T2DM complications or whether the abovementioned factors play anyway a pivotal role in the outcome of the disease.

Finally, the impact of baseline liver steatosis on post-SVR glycemic control was not studied and we were not able to report details regarding oral hypoglycemic agents.

In conclusion, a significant improvement of glycaemic control was observed in diabetic patients with CHC who obtained SVR with a clinically meaningful de-escalation of antihyperglycaemic therapy; these findings raise the question as to whether the HCV eradication may also impacts the future morbidity and mortality due to T2DM. For this reason, close T2DM follow up post-HCV treatment is warrantedand large prospective studies are needed to validate these results.

ACKNOWLEDGEMENTS

No specific funding was obtained for this study.

REFERENCES

1. White DL, Ratziu V, El-Serag HB. Hepatitis C infection and risk of diabetes: a systematic review and meta-analysis. J Hepatol 2008; 49: 831-844
2. Naing C, Mak JW, Ahmed SI, Maung M. Relatioship between hepatitis C virus infection and type 2 diabetes mellitus: a meta-analysis. World J Gastroenterol 2012 18: 1642-51
3. Negro F. Facts and fictions of HCV and comorbidities: steatosis, diabetes mellitus, and cardiovascular diseases. J Hepatol 2014; 61: S69-S78
4. Mason AL, Lau JY, Hoang N et al. Association of diabetes mellitus and chronic hepatitis C virus infection. Hepatology 1999 29: 328-333
5. Lecube A, Hernández C, Genescà J, Esteban JI, Jardí R, Simó R. High prevalence of glucose abnormalities in patients with hepatitis C virus infection: a multivariate analysis considering the liver injury. Diabetes Care2004; 27: 1171-1175
6. Knobler H, Stagnaro-Green A, Wallenstein S, Schwartz M, Roman SH. Higher incidence of diabetes in liver transplant recipients with hepatitis C. J ClinGastroenterol 1998 26: 30-33
7. Delgado-Borrego A, Liu YS, Jordan SH et al. Prospective study of liver transplant recipients with HCV infection: evidence for a causal relationship between HCV and insulin resistance. Liver Transpl 2008 14: 193-201
8. Hui JM, Sud A, Farrell GC et al. Insulin resistance is associated with chronic hepatitis C virus infection and fibrosis progression. *Gastroenterology* 2003; 125: 1695-1704

9. Bugianesi E, Marchesini G, Gentilcore E et al. Fibrosis in genotype 3 chronic hepatitis C and nonalcoholic fatty liver disease: Role of insulin resistance and hepatic steatosis. Hepatology 2006; 44: 1648-1655
10. Cua IH, Hui JM, Kench JG, George J. Genotype-specific interactions of insulin resistance, steatosis, and fibrosis in chronic hepatitis C. Hepatology 2008; 48: 723-731
11. Lai MS, Hsieh MS, Chiu YH, Chen TH. Type 2 diabetes and hepatocellular carcinoma: a cohort study in high prevalence area of hepatitis virus infection. Hepatology 2006; 43: 1295-1302
12. Veldt BJ, Chen W, Heathcote EJ et al. Increased risk of hepatocellular carcinoma among patients with hepatitis C cirrhosis and diabetes mellitus. Hepatology 2008; 47: 1856-1862
13. Giordanino C, Ceretto S, Bo S et al. Type 2 diabetes mellitus and chronic hepatitis C: which is worse? Results of a long-term retrospective cohort study. Dig Liver Dis 2012; 44: 406-412
14. Hung CH, Lee CM, Wang JH et al. Impact of diabetes mellitus on incidence of hepatocellular carcinoma in chronic hepatitis C patients treated with interferon-based antiviral therapy. Int J Cancer 2011; 128: 2344-2352
15. Romero-Gómez M, Del Mar Viloria M, Andrade RJet al.Insulin resistance impairs sustained response rate to peginterferon plus ribavirin in chronic hepatitis C patients. Gastroenterology 2005; 128: 636-641
16. D'Souza R, Sabin CA, Foster GR. Insulin resistance plays a significant role in liver fibrosis in chronic hepatitis C and in the response to antiviral therapy. Am J Gastroenterol 2005; 100: 1509-1515

17. Persico M, Capasso M, Persico E et al. Suppressor of cytokine signaling3 (SOCS3) expression and hepatitis C virus-related chronic hepatitis: Insulin resistance and response to antiviral therapy. Hepatology 2007; 46: 1009-1015
18. Poustchi H, Negro F, Hui J et al. Insulin resistance and response to therapy in patients infected with chronic hepatitis C virus genotypes 2 and 3. J Hepatol 2008; 48: 28-34
19. Delgado-Borrego A, Jordan SH, Negre B et al. Reduction of insulinresistancewitheffectiveclearance of Hepatitis C infection: resultsfromthe HALT-C trial. ClinGastroenterol Hepatol 2010 8: 458-462
20. Aghemo A, Prati GM, Rumi MG et al. A sustained virological response preventsdevelopment of insulinresistance in chronic hepatitis C patients. Hepatology 2012; 56: 1681-1687
21. YounossiZ, Negro F, Serfaty L et al. Homeostasis model assessment of insulin resistance does not seem to predict response to telaprevir in chronic hepatitis C in the REALIZE trial. Hepatology 2013 58: 1897-1906
22. Serfaty L, Forns X, Goeser T et al. Insulin resistance and response to telaprevir plus peginterferon alpha and ribavirin in treatment-naïve patients infected with HCV genotype 1. Gut 2012 61: 1473-1480
23. Willemse SB, Baak LC, Kuiken SD et al. Sofosbuvir plus simeprevir for the treatment of HCV genotype 4 patients with advanced fibrosis or compensated cirrhosis is highly efficacious in real life. J Viral Hepat 2016 23:950-954
24. Pavone P, Tieghi T, D'EttorreGet al. Rapid decline of fasting glucose in HCV diabetic patients treated with direct-acting antiviral agents. ClinMicrobiol Infect 2016 22: 462.e1-462.e3

25. Pashun RA. Shen NT, Jesudian A. Markedly improved glycemic control in poorly controlled type 2 diabetes following Direct Acting Antiviral treatment of genotype 1 Hepatitis C. Case Rep Hepatol 2016 http://dx.doi.org/10.1155/2016/7807921
26. Fabrizio C, Procopio A, Scudeller L et al. HCV and diabetes: towards a "Sustained" glycaemic improvement after treatment with DAAs ?ClinMicrobiol Infect 2016 doi: 10.1016/j.cmi.2016.09.021
27. LeClerc SB, Falck-Ytter Y, Falck-Ytter C, Pascuzzi K, Burant C, Rife K. Viral response to hepatitis C direct-antiviral significantly improves diabetes control. Hepatology 2016 64 (suppl. 1): 488A
28. METAVIR Cooperative Study Group. Intraobserver and interobserver variations in liver biopsy interpretation in patients with chronic hepatitis C. Hepatology 1994 20: 15-20
29. Pawlotsky JM, Aghemo A, Back D et al. EASL recommendations on treatment of Hepatitis C 2015. J Hepatol 2015 63: 199-236
30. Wallace TM, Levy JC, Matthews DR. Use and abuse of HOMA modeling. Diabetes Care 2004 27: 1487-1495
31. ESH/ESC Task Force for the Management of Arterial Hypertension. 2013 Practice Guidelines for the management of arterial hypertension of the European Society of Hypertension (ESH) and the European Society of Cardiology (ESC): ESH/ESC Task Force for the Management of Arterial Hypertension. J Hypertens 2013 31: 1925-1938
32. American Diabetes Association. Diagnosis and classification of diabetes mellitus. Diabetes care 2011 34 (suppl. 1): 62-69

33. George K, Alberti MM, Zimmet P, Shaw J. The metabolic syndrome – a new worldwide definition. IDF Epidemiology Task Force Consensus Group. Lancet 2005 366: 1059-1062
34. European Association for the Study of the Liver, AsociacionLatinoamericana para el Studio del H., EASL-ALEH. Clinical Practice Guidelines: Non-invasive tests for evaluation of liver disease severity and prognosis. J Hepatol 2015 63: 237-264
35. Azad A, Bichoupan K, Chekuri S, Schiano T, Branch AD. Diabetes is a risk factor for weight gain among patients cured of HCV with Sofosbuvir/ledipasvir. Hepatology 2016 64 (suppl. 1): 377A
36. Kaddai V, Negro F. Current understanding of insulin resistance in hepatitis C. *Expert Rev Gastroenterol Hepatol* 2011; 5: 503-516
37. Kawaguchi T, Yoshida T, Harada M et al. Hepatitis C virus down-regulates insulin receptor substrates 1 and 2 through up-regulation of suppressor of cytokine signalling 3. Am J Pathol 2004 165: 1499-1508
38. Persico M, Russo R, Persico E et al. SOCS3 and IRS-1 gene expression differs between genotype 1 and genotype 2 hepatitis C virus-infected HepG2 cells. ClinChem Lab Med 2009 47: 1217-1225
39. Pazienza V, Vinciguerra M, Andriulli A, Mangia A. Hepatitis C virus core protein genotype 3° increases SOCS-7 expression through PPAR-γ in Huh-7 cells. J Gen Virol 2010 91: 1678-1686
40. Kawaguchi Y, Mizuta T, Oza N et al. Eradication of hepatitis C virus by interferon improves whole-body insulin resistance and hyperinsulinemia in patients with chronic hepatitis C. Liver Int 2009; 29: 871-877

41. UK Prospective Diabetes Study (UKPDS) Group. Intensive blood-glucose control with sulphonylureas or insulin compared with conventional treatment and risk of complications in patients with type 2 diabetes (UKPDS 33). Lancet 1998; 352: 837-53.

42. Coppo C, Bonfanti D, Bo S et al. Risk of microangiopathy in type 2 diabetes mellitus patients with or without chronic hepatitis C. Results of a retrospective long-term controlled cohort study. Dig Liv Dis 2015 47: 405-410

43. Hedenstierna M, Nangarhari A, Weiland O, Aleman S. Diabetes and cirrhosis are risk factors for hepatocellular carcinoma after successful treatment of chronic hepatitis C. Clin Infect Dis 2016 63: 723-729

44. Van der Meer AJ, Feld JJ, Hofer H et al. Risk of cirrhosis-related complications in patients with advanced fibrosis following hepatitis C virus eradication. J Hepatol 2016 http://dx.doi.org/10.1016/j.jhep.2016.10.017

45. Donadon V, Balbi M, Valent F, Avogaro A. Glycated hemoglobin and antidiabetic strategies as risk factors for hepatocellular carcinoma. World J Gastroenterol 2010 16: 3025-3032

46. Arase Y, Kobayashi M, Suzuki F et al. Effect of type 2 diabetes on risk for malignancies includes hepatocellular carcinoma in chronic hepatitis C. Hepatology 2013 57: 964-973

47. Aguilar M, Liu B, Holt EW, Bhuket T, Wong RJ. Impact of obesity and diabetes on waitlist survival, probability of liver transplantation and post-transplant survival among chronic hepatitis C virus patients. Liver Int 2016 36: 1167-1175

48. Boughton C, Mustafa O, Suddle A et al. Good glycaemic control pre-operatively protects against adverse outcomes associated with diabetes in liver transplant recipients. Hepatology 2016 64 (suppl. 1): 493A

FIGURE LEGENDS

Figure 1. Flow of the recruited patients.

TABLES

Table 1. Antiviral treatment of diabetic patients.

	Patients, n (%)	SVR, n (%)
Sofosbuvir + ribavirin	13 (11.8%)	12 (92.3%)
Sofosbuvir + Daclatasvir with or without ribavirin	11 (10%)	11 (100%)
Sofosbuvir + Ledipasvir with or without ribavirin	46 (41.8%)	44 (95.6%)
Sofosbuvir + Simeprevir with or without ribavirin	22 (20%)	18 (81.8%)
Ombitasvir-Paritaprevir-Ritonavir with or without ribavirin	6 (5.5%)	5 (83.3%)
Ombitasvir-Paritaprevir-Ritonavir + Dasabuvir with or without ribavirin	12 (10.9%)	11 (91.7%)

Table 2. Baseline demographic, anthropometric and clinical features of the patients.

	Group 1 (101 pts)	Group 2 (21 pts)	p
Male, n (%)	71 (70.3%)	14 (66.7%)	0.945
Age† (years)	61.39 ± 10.92	65.06± 13.12	0.276
Weight† (kg)	75.29 ± 13.70	76.62 ± 19.33	0.771
Height† (cm)	169 ± 8	168 ± 12	0.722
BMI† (kg/m²)	26.58 ± 4.70	27.63 ± 4.81	0.401
Smokers, n (%)	22 (21.8%)	5 (23.8%)	0.411
Waist Circumference†(cm)	107.94 ±12.65	101.00 ±8.97	0.067
Hip Circumference†(cm)	100.84 ±13.09	103.75 ±13.49	0.509
Waist/Hip ratio	1.07	0.97	0.090
Arterial Hypertension, n (%)	40 (39.6%)	7 (33.3%)	0.632
Metabolic Syndrome, n (%)	37 (36.6%)	5 (45.5%)	0.858
Co-morbidities, n (%)			0.267
None	64 (63.4%)	14 (66.7%)	
Cardiovascular disease	8 (7.9%)	1 (4.8%)	
Kidney disease	3 (3%)	2 (9.5%)	
Thyroid disease	6 (5.9%)	0 (0%)	
Mental depression	4 (4%)	2 (9.5%)	
Biliary stones	4 (4%)	0 (0%)	
Bronchopathy	3 (3%)	0 (0%)	
Vasculitis	2 (2%)	2 (9.5%)	
Dermatologic disease	7 (6.9%)	0 (0%)	

† mean ± SD

Table 3. Baseline liver-related clinical, biochemical, virological and metabolic features of the patients.

	Group 1 (101 patients)	Group 2 (21 patients)	p
HCV-RNA (log_{10} IU/mL)*	6.50 (6.11 – 6.76)	6.71 (6.26 – 6.92)	0.391
Genotype, n (%) **1a** **1b** **2** **3** **4**	 4 (4%) 55 (54.5%) 13 (12.9%) 19 (18.8%) 10 (9.9%)	 1 (5%) 13 (65%) 1 (5%) 3 (15%) 2 (10%)	0.85
***IL28B*, n (%)** **CC** **CT** **TT**	 22 (22%) 59 (58%) 20 (10%)	 12 (57%) 8 (38%) 1 (5%)	0.06
Liver stiffness (kPa)*	24.31± 14.79	18.62 12.52	0.11
Metavir, n (%) **0-1** **2** **3** **4**	 6 (5.9%) 3 (3%) 19 (18.8%) 73 (72.3%)	 4 (19.1%) 5(23.8%) 2 (9.5%) 10 (47.6%)	0.024
Cirrhosis, n (%) **Child A** **Child B**	 63 (86%) 10 (14%)	 7 (70%) 3 (30%)	0.365
Previous HCC, n (%)	6 (6%)	1 (5%)	1
Oesophageal/gastric varices, n (%)	46 (45.5%)	6 (28.6%)	0.235
Diet only **Diet + OHA** **Diet + insulin** **Diet + OHA+ insulin**	36 (35.6%) 19 (18.8%) 38 (37.6%) 8 (7.9%)	7 (33.3%) 8 (38.1%) 4 (19%) 2 (9.5%)	0.081
Retinopathy, n (%)	7 (6.9)	3 (14.3%)	0.374
Nephropathy, n (%)	2 (2%)	0 (0%)	0.436
Coronary artery disease, n (%)	2 (2%)	0 (0%)	1
Peripheral artery disease, n (%)	5 (5%)	0 (0%)	0.586
Neuropathy, n (%)	4 (4%)	0 (0%)	1

* mean ± SD

Abbreviations: IL28B, Interleukin 28B; GGT, Gamma-glutamyltranspeptidase**;** HCC, epatocellular carcinoma; HbA1c, Glycated haemoglobin**;** OHA, oral hypoglycemic agents.

Table 4. Anthropometric, biochemical and metabolic variations in sustained responders and relapsing/untreated patients.

	Group 1 (101 patient)			Group 2 (21 patients)		
	Baseline	**End of study**	**p**	**Baseline**	**End of study**	**p**
AST (IU/mL) *	42.3 ± 37.6	28.2 ± 11.0	0.02	40.3 ± 38.4	42.2 ± 39.8	0.85
ALT (IU/mL) *	81.2 ± 77.2	36.0 ± 12.0	<0.001	78.7 ± 67.3	82.2 ± 71.6	0.74
GGT (IU/mL) *	87.8 ± 81.0	62.5 ± 73.2	0.02	65.2 ± 64.0	67.0 ± 68.1	0.43
Bilirubin (mg/dL) §	1.0 (0.6 – 1.8)	0.8 (0.7 – 1.1)	0.12	0.9 (0.6 – 1.3)	1.0 (0.7 - 1.4)	0.52
Albumin (g/L) §	42 (31 – 48)	44 (33 – 49)	0.09	43 (34 – 47)	42 (33 – 48)	0.78
INR§	1.3 (1.0 – 1.8)	1.0 (1.0 – 1.5)	0.08	1.2 (1.0 – 1.7)	1.3 (1.0 – 2.1)	0.33
Leukocytes (x10^3/µL cells) §	4.2 (2.2 – 7.8)	5.4 (3.3 – 8.2)	0.07	4.4(2.8 – 6.2)	4.1 (2.3 – 7.3)	0.64
Platelets (x10^3/µL cells) §	155 (62 – 287)	173 (52 – 274)	0.08	164 (73 – 245)	162 (68 – 274)	0.35
Glucose (mg/dL) *	152.4 ± 56.4	134.3 ± 41.3	0.002	145.3 ± 30.2	140.0 ± 47.9	0.71
HbA1c (mmol/mol) *	52.2 ± 15.4	46.5 ± 16.2	<0.001	53.4 ± 9.5	55.3 ± 20.6	0.78
HOMA-IR*	5.2 ± 2.5	3.1 ± 1.6	<0.001	4.9 ± 2.6	4.6 ± 2.3	0.29
Body weight (kg) *	75.3 ± 13.7	77.9 ± 19.8	0.02	76.6 ± 19.3	76.9 ± 20.7	0.56

* mean ± SD
§ median (range)

Abbreviations: AST, Aspartate transaminase; ALT, alanine transaminase; GGT, Gamma-glutamyltranspeptidase; INR, International Normalized Ratio; HbA1c, Glycated haemoglobin; HOMA-IR, Homeostasis Model Assessment – Insulin Resistance

Table 5.Results of the multivariable Cox regression analysis of the factors potentially associated with significant improvement of glycaemic control.

	OR	C.I.	p
HCV RNA levels	0.999	0.999-1.000	0.154
Liver stiffness	1.014	0.970-1.059	0.541
GGT levels	1.002	0.996-1.009	0.496
Albumine levels	0.713	0.224-2.275	0.568
Platelets	0.999	0.999-1.000	0.256
BMI	1.115	0.976-1.274	0.108
SVR	49.75	2.197-112.67	0.014
Insulin therapy	1.379	0.408-4.659	0.605
Arterial hypertension	1.083	0.339-3.465	0.893

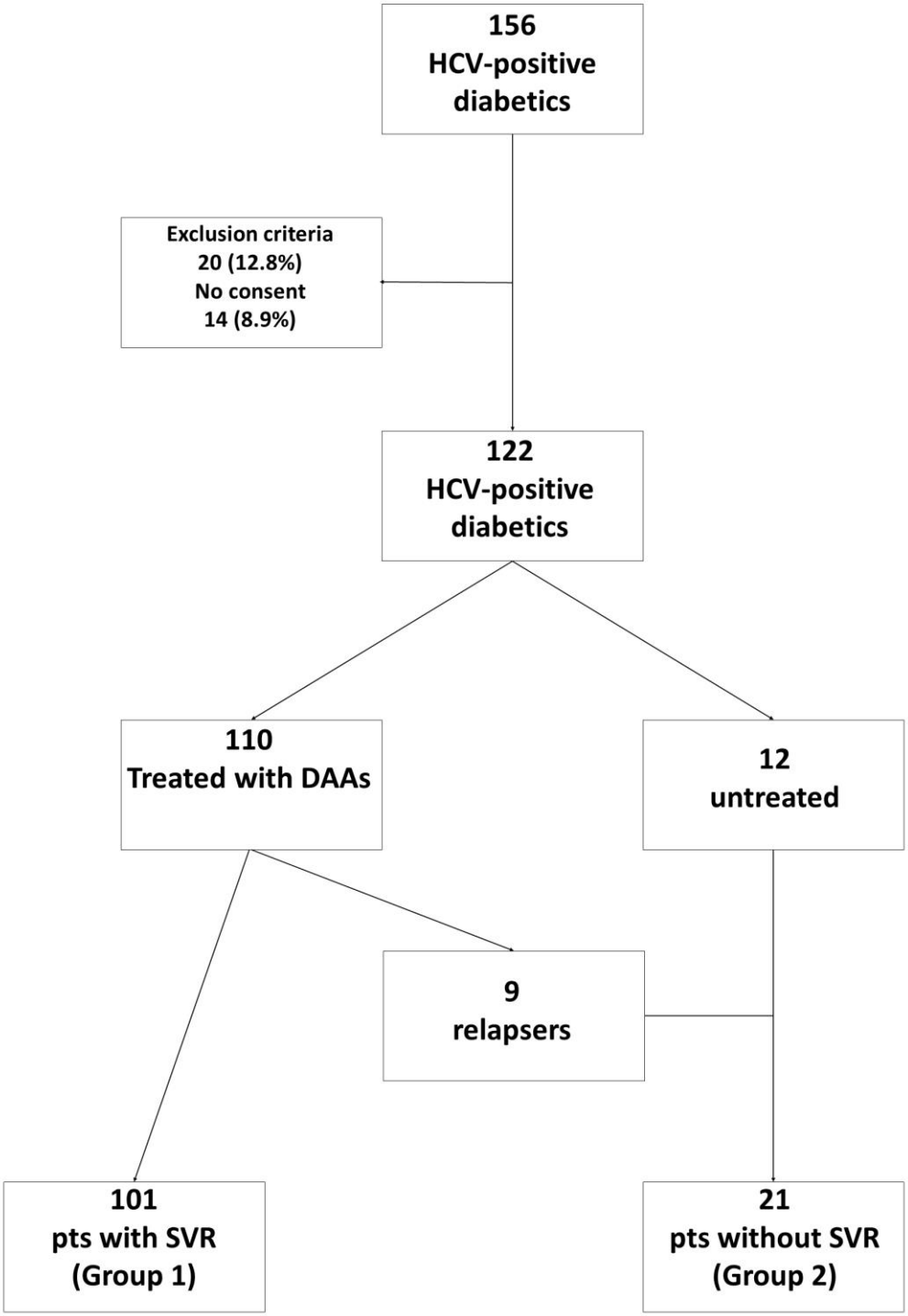

Figure 1