Digestive and Liver Disease 51 (2019) 142–148

Contents lists available at ScienceDirect

Digestive and Liver Disease

journal homepage: www.elsevier.com/locate/dld

Liver, Pancreas and Biliary Tract

Diabetes is associated with advanced fibrosis and fibrosis progression in non-genotype 3 chronic hepatitis C patients

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ARTICLE INFO

Article history:
Received 15 February 2018
Received in revised form 1 July 2018
Accepted 4 July 2018
Available online 17 July 2018

Keywords:
Diabetes
Genotype 3
Hepatitis C virus
Liver cirrhosis
Ultrasound

ABSTRACT

Background: Diabetes is a risk factor of fibrosis progression in chronic hepatitis C (CHC). However, only one longitudinal study exploring whether diabetes is associated with progression from non-cirrhotic liver to cirrhosis in CHC patients has been conducted.
Aims: We investigated whether diabetes is associated with progression from non-cirrhotic liver to cirrhosis in non-genotype 3 CHC patients.
Methods: A cohort consisting of 976 non-genotype 3 patients histologically proven to have CHC was studied. After excluding patients with biopsy-proven or ultrasound-identified cirrhosis, there were 684 patients without cirrhosis. All 684 patients underwent hepatocellular carcinoma surveillance using ultrasound every 6 months, with a median duration of follow-up evaluation of 102.4 months. During the follow-up period, 60 patients developed cirrhosis according to ultrasound findings.
Results: For the subgroup of 684 patients without cirrhosis, Kaplan–Meier survival analyses showed no significantly different cumulative incidences of cirrhosis (log-rank test; P = 0.71) among the patients with diabetes as compared to those without. However, after making adjustments for age, gender, fibrosis, steatosis, sustained virological response status, and obesity using Cox's proportional hazard model, diabetes was found to be an independent predictor for cirrhosis (HR = 1.9; 95% CI = 1.05–3.43, P = 0.03).
Conclusions: Diabetes is associated with progression from non-cirrhotic liver to cirrhosis in non-genotype 3 CHC patients.

1. Introduction

Hepatitis C virus (HCV) infection causes a systemic disease that features some extrahepatic manifestations, among which diabetes mellitus (DM) plays a leading role [1,2]. DM is the second most prevalent extrahepatic manifestation, affecting 15% of HCV-infected patients [3]. Evidence indicates that insulin resistance (IR)/DM is more strongly associated with non-3 HCV genotypes [4–6].

HCV steatosis occurs in the setting of multiple metabolic abnormalities collectively referred to as "hepatitis C-associated dysmetabolic syndrome" (HCADS). HCV steatosis has a clinical impact in accelerating fibrogenesis [7].

A meta-analysis revealed that the presence of IR is a significant risk factor for advanced hepatic fibrosis in HCV patients [8]. IR is a precursor for DM and develops before overt DM in patients with HCV [9]. Meanwhile, fibrosis progresses more rapidly to cirrhosis in patients with HCV, IR, and DM [10,11]. Numerous cross-sectional studies have demonstrated an increased risk of advanced fibrosis associated with DM among chronic hepatitis C (CHC) patients [12–18].

Longitudinal cohort studies would provide an ideal means of evaluating the impact of DM in the clinical course of HCV infection. However, there has only been one longitudinal population-based cohort study that followed HCV patients without liver cirrhosis, and that study found a higher risk of progression to cirrhosis in new-onset DM patients than in non-DM patients while adjusting for confounding variables [19]. That study was limited by its use of data from the National Health Insurance Research Database (NHIRD) for Taiwan. One limitation of studies using NHIRD data is the misclas-

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https://doi.org/10.1016/j.dld.2018.07.003
1590-8658/

sification of both the risk and the outcome of interest (in this case, DM was "the risk" and cirrhosis was "the outcome of interest"), such that the association cannot be verified. Furthermore, the validity of using the Ninth Revision of the International Classification of Diseases (ICD-9) codes to define cirrhosis was unknown in that study's cohort.

Therefore, we conducted the present hospital-based cohort study to investigate the relationship of DM and fibrosis progression in non-genotype 3 CHC patients in a cross-sectional and longitudinal manner through careful chart reviews. More specifically, we investigated whether DM was associated with advanced fibrosis in a large cohort of 976 biopsy-proven non-genotype 3 CHC patients via a cross-sectional evaluation, and we examined the differences between DM patients and non-DM patients in terms of the risk of progression from non-cirrhotic liver to ultrasound-identified liver cirrhosis via a longitudinal evaluation.

2. Patients and methods

2.1. Methods

We retrospectively reviewed the medical records of consecutive chronic HCV-infected patients treated with interferon (IFN)-based therapy at a medical center in Taiwan from 1999 to 2011. Patients with Wilson disease, a current or past history of excess alcohol consumption (≥20 g daily), probable autoimmune hepatitis, primary biliary cholangitis (PBC), primary sclerosing cholangitis (PSC), human immunodeficiency virus (HIV) infection, regular use of steatosis-inducing drugs (e.g. amiodarone, methotrexate, tamoxifen and corticosteroids) [20], major uncontrolled depressive illness (which is a contraindication for IFN therapy), previous liver transplantation, evidence of hepatocellular carcinoma (HCC), hepatitis B virus (HBV) co-infection, HCV genotype 3, and unknown HCV genotype were excluded.

Probable autoimmune hepatitis was diagnosed by seropositivity for antinuclear antibodies (ANA) or smooth muscle antibodies (SMA) antibodies at titers >1:80, immunoglobulin G or γ-globulin levels at >1.1x the upper limits, and liver histology showing moderate to severe interface hepatitis [21]. A diagnosis of PBC was made based on elevated alkaline phosphatase (Alk-P) and the presence of antimitochondrial antibodies (AMA) at a titer >1:40 [22]. A diagnosis of PSC was made in patients with elevated Alk-P when a cholangiography showed characteristic bile duct changes with multifocal strictures and segmental dilatations, and when secondary causes of sclerosing cholangitis had been excluded [23,24]. HIV-1 testing was done using an enzyme-linked immunosorbent assay (ELISA) to detect antibodies to HIV-1. Specimens (serum) with a non-reactive result from the ELISA were considered HIV-negative. Specimens with a reactive ELISA result were retested in duplicate. If the result of either duplicate test was reactive, the specimen was reported as reactive and underwent confirmatory testing with western blot analysis.

For each patient, the date of inclusion was the date of the liver biopsy, with the liver biopsy being performed within 6 months before starting IFN-based therapy. DM was identified based on diagnoses documented in medical records, a serum fasting glucose level >126 mg/dL, or the use of antidiabetic drugs [25].

The percentage of steatosis was quantified by determining the average proportion of hepatocytes affected. The degree of liver necroinflammation in each patient was calculated by Histology Activity Index scores [26]. The degree of liver fibrosis in each patient was graded and staged according to the modified Knodell histology index, and advanced fibrosis was defined as a score of 3–4 [27]. All the procedures used in the study were in accordance with the ethical standards of the responsible committees on human experimentation (institutional and national) and with the Helsinki Declaration of 1975, as revised in 2008. This study was approved by the Institutional Review Board of Kaohsiung Chang Gung Memorial Hospital (IRB number: 201601042B0). The requirement for informed consent was waived by the IRB. The data were analyzed anonymously.

2.2. Treatment and follow-up evaluation

All 976 patients were treated with IFN and ribavirin. A sustained virological response (SVR) was defined as undetectable HCV RNA at follow-up week 24 [28].

The patients underwent HCC surveillance using ultrasound and alpha-fetoprotein (AFP) every 6 months [29]. Ultrasound was performed by 30 hepatologists in our department. A diagnosis of liver cirrhosis was made with ultrasound according to the presence of nodular liver surface, small liver size, and coarse liver parenchyma [30]. The gold standard for diagnosing cirrhosis by ultrasound is the presence of nodular liver surface.

Predefined criteria were used to determine the severity of ultrasound-identified steatosis [31]. The diagnosis of HCC was based on the guidelines of the American Association for the Study of Liver Disease (AASLD) or the European Association for the Study of the Liver (EASL) [32,33]. The time at risk of cirrhosis was measured from the date of liver biopsy until the development of ultrasound-identified liver cirrhosis, death or liver transplantation, or at the last recorded visit within the previous 6 months before October 17, 2016.

2.3. Statistical analysis

Baseline characteristics and clinical variables were summarized as mean ± standard deviation, median (interquartile range), or percentage. The chi-square test, Fisher's exact test, or independent two-sample t-test was used, as appropriate, to assess the significance of differences in distributions.

The performance of ultrasound for diagnosing liver cirrhosis was assessed using the area under the receiver operating characteristic curve (AUROC). We calculated the sensitivity, specificity, negative predictive value, positive predictive value, and accuracy by the maximal sum of specificity and sensitivity.

The associations between the clinical factors and the presence of advanced fibrosis were determined by multivariate logistic regression analyses.

We performed a multivariate Cox regression analysis after adjusting for variables associated with increased risk of fibrosis progression in non-cirrhotic CHC patients according to the findings of previous studies (i.e., age, gender, baseline DM, SVR, obesity, steatosis >10%, and the presence of advanced fibrosis) [8,34–39].

Furthermore, to evaluate the impact on the occurrence of liver cirrhosis of developing DM and other metabolic comorbidities during the follow-up period, DM, antilipid treatment, and antihypertensive treatment were introduced as time-dependent categorical covariates in the Cox regression. For example, fixed DM values were used for patients who never had DM (DM, 0) and patients with DM at the time of their inclusion (DM, 1), while patients who did not have DM at inclusion but then developed DM during follow-up evaluation were switched from non-DM to DM status, with their DM values changed from 0 to 1.

A Kaplan–Meier analysis and the log-rank test were performed by comparing the differences in the cumulative incidence of liver cirrhosis between different groups of patients, and by comparing the differences in the cumulative incidence of new onset DM between patients with or without the use of statins. A P-value of

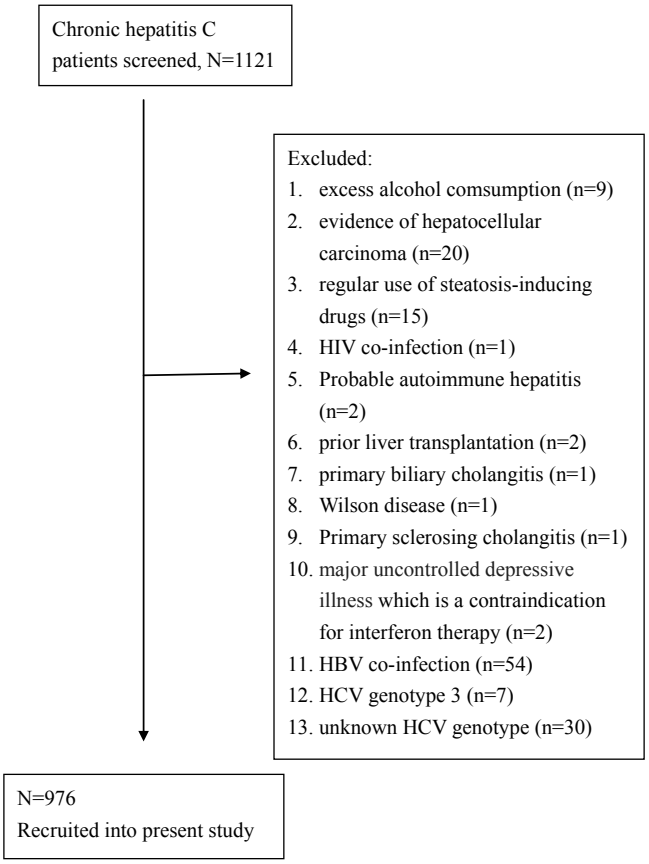

Fig. 1. Patient disposition of the current study. HIV, human immunodeficiency virus; HBV, hepatitis B virus; HCV, hepatitis C virus. [a]Includes amiodarone, methotrexate, tamoxifen and corticosteroids.

less than 0.05 was considered statistically significant. All statistical analyses were performed using STATA (version 11.1).

3. Results

3.1. Demographic and clinical characteristics of all patients (N = 976)

The inclusion and exclusion of potential subjects for this study are depicted in Fig. 1. Among 1121 screened CHC patients, 976 (87.1%) were subsequently included. The demographic and clinical characteristics of all the patients (N = 976) are shown in Table 1. The SVR rate for these patients was 71.6%. Around 50% of the patients were male, and the mean age of the patients was 53.5 years. Steatosis >10% was found in 27.3% of the patients, advanced fibrosis was found in 46%, and 37.3% had moderate or severe necroinflammation. Fifty percent of the patients were genotype 1, and 47.3% of the patients were genotype 2. Nineteen percent of the patients had DM, and 24% of the patients were obese (body mass index (BMI) > 27 kg/m^2) [40].

3.2. Factors associated with advanced fibrosis

A multivariate logistic regression analysis showed that age ≥60 years, the presence of moderate to severe necroinflammation, and DM were independent factors associated with the presence of advanced fibrosis (Table 2).

Table 1
Demographic and clinical characteristics of all patients (N = 976).

Variables	
Age (year)	53.5 ± 10.7
Male	494 (50.6%)
DM	187 (19.2%)
BMI (kg/m^2)	
<24	416 (42.6%)
24–27	323 (33.1%)
>27	235 (24.1%)
AST (IU/L)	93 (63–136)
ALT (IU/L)	131 (95–195)
Bilirubin (mg/dL)	1.0 ± 0.4
Viral load >600,000 IU/mL	169 (17.3%)
Fibrosis score	
0–2	525 (53.8%)
3–4	451 (46.2%)
Steatosis	
≤10%	710 (72.75%).
>10%	266 (27.25%),
Necroinflammation score	
<9	612 (62.7%)
≥9	364 (37.3%)
Genotype	
1	478 (50.0%)
2	462 (47.3%)
Mixed	24 (2.5%)
6	12 (1.2%)

Data were expressed as mean ± SD or median (interquantile). DM, diabetes mellitus; BMI, body mass index; AST, aspartate aminotransferase; ALT, alanine aminotransferase; patients were categorized as normal weight or underweight (<24 kg/m^2), overweight (24–27 kg/m^2), or obese (>27 kg/m^2) according to the definition of the Health Promotion Administration of the Ministry of Health and Welfare in Taiwan [40].

3.3. The diagnosis of cirrhosis by ultrasound

There were 254 patients with liver cirrhosis diagnosed by liver biopsy, meaning that the prevalence of liver cirrhosis for the full cohort was 26.0%. The sensitivity of ultrasound for diagnosing cirrhosis was 60.1%, its specificity was 90.7%, its positive predictive value was 69.7%, its negative predictive value was 86.7%, its AUROC was 0.76, and its diagnostic accuracy was 82.89%.

3.4. Baseline factors associated with the progression from non-cirrhotic to cirrhotic liver

We excluded the patients with liver biopsy-proven cirrhosis (N = 254) at baseline. In the remaining 722 patients, there were 38 patients with ultrasound-identified liver cirrhosis. After excluding these 38 patients, there were 684 patients without liver biopsy-proven or ultrasound-identified liver cirrhosis at baseline. In this subgroup cohort, the median duration of follow-up evaluation was 102.4 months. During the follow-up period, 60 patients developed liver cirrhosis according to ultrasound findings. The demographic and clinical characteristics of the non-cirrhotic patients according to the presence or absence of DM are shown in Table 3. One hundred and three of these patients had DM, while 581 of the patients did not have DM. The mean age was older, the median alanine aminotransferase (ALT) level was higher, and the incidences of HCC and cirrhosis were higher in the DM patients compared with the non-DM patients. According to multivariate stepwise Cox analyses, a fibrosis score of 3, the presence of DM, and non-SVR were identified as independent factors that were significantly associated with the development of liver cirrhosis (Table 4). The cumulative incidence of liver cirrhosis development in the DM patients was not different than that in the non-DM patients (log rank test, P = 0.71; Fig. 2).

In the subgroup analysis, DM was not independently associated with the development of liver cirrhosis (Supplementary Table 1) in

Table 2
Univariate and multivariate logistic regression analysis on factors associated with presence of advanced fibrosis (fibrosis score 3–4 modified Knodell histology index) (N = 976).

Variables	Comparison	Univariate			Multivariate		
		OR	95%CI	*P*	OR	95%CI	*P*
Age (year)	≥60 vs <60	1.81	1.36–2.40	<0.001	1.59	1.18–2.14	0.002
DM	Yes vs no	1.93	1.39–2.67	<0.001	1.82	1.30–2.56	<0.001
Necroinflammation	≥9 vs <9	3.15	2.41–4.13	<0.001	2.94	2.23–3.87	<0.001
Sex	Male vs female	0.76	0.59–0.98	0.04	0.80	0.61–1.04	0.10
Obesity	BMI > 27 vs ≤27 (kg/m^2)	1.01	0.75–1.36	0.95	1.02	0.74–1.4	0.90
Steatosis	>10 vs ≤10%	1.02	0.77–1.36	0.88	0.98	0.73–1.33	0.90

DM, diabetes mellitus; BMI, body mass index.

Table 3
Comparison of demographic and clinical characteristics between chronic hepatitis C patients with diabetes and those without diabetes (N = 684).

Variables	Diabetes, N = 103	Non-diabetes, N = 581	*P*
Age (year)	56.67 ± 7.95	51.49 ± 11.37	<0.001
Male	62 (60.19%)	297 (51.12%)	0.09
BMI (kg/m^2)			0.24
<24	43 (41.75%)	265 (45.69%)	
24–27	29 (28.16%)	185 (31.9%)	
>27	31 (30.10%)	130 (22.41%)	
AST (IU/L)	81 (58–137)	84 (57–124)	0.44
ALT (IU/L)	138.5 (96–211)	128 (90–191)	0.04
Bilirubin (mg/dL)	0.85 ± 0.28	0.89 ± 0.34	0.25
Viral load >600,000 IU/mL	19 (22.89%)	95 (20.04%)	0.55
Fibrosis score			0.50
0–2	75 (72.82%)	441 (75.9%)	
3	28 (27.18%)	140 (24.1%)	
Steatosis			0.30
≤10%	70 (67.96%)	424 (72.98%)	
>10%	33 (32.04%)	157 (27.02%)	
Necroinflammation score			0.40
<9	66 (64.08%)	397 (68.33%)	
≥9	37 (35.92%)	184 (31.67%)	
Genotype 1	41 (39.81%)	273 (46.99%)	0.18
SVR	84 (81.55%)	458 (78.83%)	0.53
HCC incidence	17 (16.5%)	37 (6.37%)	<0.001
Follow-up years for cirrhosis	8.22 ± 3.37	8.13 ± 3.67	0.83
Cirrhosis incidence	15 (14.56%)	45 (7.75%)	0.02

Data were expressed as mean ± SD or median (interquantile). BMI, body mass index; AST, aspartate aminotransferase; ALT, alanine aminotransferase; patients were categorized as normal weight or underweight (<24 kg/m^2), overweight (24–27 kg/m^2), or obese (>27 kg/m^2) according to the definition of the Health Promotion Administration of the Ministry of Health and Welfare in Taiwan [40].

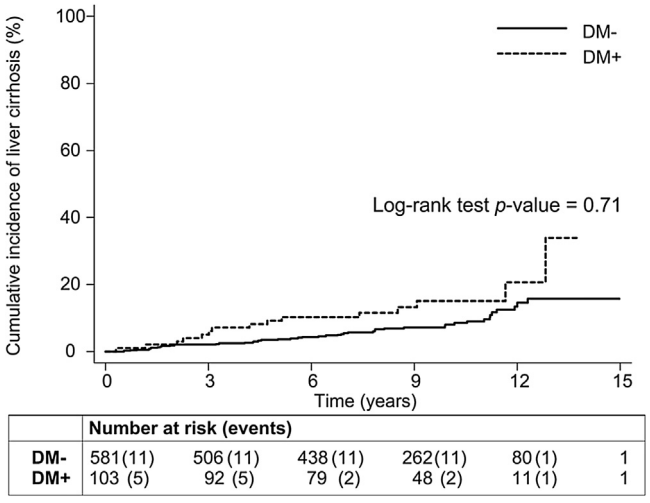

Fig. 2. Cumulative incidence of cirrhosis in chronic hepatitis C patients with diabetes and those without diabetes.

the SVR patients (N = 542). Also, the cumulative incidence of liver cirrhosis development in the DM patients was not different than that in the non-DM patients in the SVR group (log rank test, P = 0.45; Supplementary Fig. 1), and DM was not independently associated with the development of liver cirrhosis (Supplementary Table 2) in the non-SVR patients (N = 142). Furthermore, the cumulative incidence of liver cirrhosis development in the DM patients was not different than that in the non-DM patients in the non-SVR group (log rank test, P = 0.79; Supplementary Fig. 2).

3.5. Impact of developing DM and other metabolic comorbidities during the follow-up on the progression from non-cirrhotic to cirrhotic liver

Sixty-two patients developed DM during the follow-up period. Antilipid treatment was noted in 112 patients and antihypertensive treatment was noted in 196 patients when considering both the baseline and the follow-up period. DM, antilipid treatment, and antihypertensive treatment were introduced as time-dependent categorical covariates in the Cox regression. Predictive analysis of the features associated with the risk of liver cirrhosis was conducted using univariate and multivariate Cox models, and the results are shown in Supplementary Table 3. Those results showed that DM, antilipid treatment, and antihypertensive treatment were not associated with the development of liver cirrhosis.

Table 4
Univariate and multivariate Cox's proportional hazard analysis of factors associated with liver cirrhosis development in non-cirrhotic patients (N = 684).

Variables	Comparison	Univariate			Multivariate		
		HR	95%CI	*P*	HR	95%CI	*P*
DM	Yes vs no	1.90	1.06–3.4	0.03	1.9	1.05–3.43	0.03
Fibrosis score	3 vs 0–2	2.65	1.59–4.39	<0.001	2.06	1.23–3.45	0.006
SVR	No vs yes	6.49	3.87–10.88	<0.001	6.05	3.58–10.24	<0.001
Age (year)	≥60 vs <60	1.78	1.03–3.07	0.04	1.52	0.88–2.64	0.14
Sex	Male vs female	0.86	0.52–1.42	0.55	0.84	0.5–1.4	0.50
Obesity	BMI> vs ≤27 (kg/m^2)	1.47	0.85–2.56	0.17	1.17	0.66–2.08	0.58
Steatosis	>10 vs ≤10%	1.65	0.98–2.76	0.06	1.58	0.93–2.69	0.09

DM, diabetes mellitus; SVR, sustained virological response; BMI, body mass index.

3.6. The role of antidiabetics and glycemic control in the development of cirrhosis

A Kaplan–Meier analysis and the log-rank test were performed to compare the differences in the cumulative incidence of liver cirrhosis for DM patients with diet control (N = 14), DM patients receiving oral antidiabetic (OAD) treatment (N = 142), and DM patients receiving insulin treatment (n = 9). The results showed that there was no significant difference in the cumulative incidence of liver cirrhosis among these three groups of DM patients (log-rank test, P = 0.68, Supplementary Fig. 3). A Kaplan–Meier analysis and the log-rank test were also performed to compare the differences in the cumulative incidence of liver cirrhosis for DM patients with a mean glycated hemoglobin (HbA1c) level of <7.0% (N = 73) to those for DM patients with a mean HbA1c level of ≥7.0% (N = 71). The results showed that there was no significant difference in the cumulative incidence of liver cirrhosis between these two groups of patients (log-rank test, P = 0.87, Supplementary Fig. 4).

3.7. The role of ultrasonographic detection of hepatic steatosis during the follow-up in the development of cirrhosis

Instances of ultrasonographic detection of hepatic steatosis at 6 months after the end of treatment were recorded. As was mentioned in the EASL guidelines, "despite observer dependency, ultrasound robustly diagnoses moderate and severe steatosis" [41]. A Kaplan–Meier analysis and the log-rank test were performed to compare the differences in the cumulative incidence of liver cirrhosis for patients with moderate or severe steatosis and patients with no steatosis or mild steatosis. The results showed that there was no significant difference in the cumulative incidence of liver cirrhosis between patients with moderate or severe steatosis and patients with no steatosis or mild steatosis (log-rank test, P = 0.36, Supplementary Fig. 5).

3.8. Does the use of statins further increase the diabetogenic risk of HCV alone?

We excluded patients with DM at baseline and patients who developed DM before the use of statins during the follow-up period (N = 139). In the remaining 545 patients, there were 65 patients who received statin treatment including at the baseline or at some point during the follow-up period. New onset DM was noted in 94 patients (71 patients without statin treatment and 23 patients with statin treatment). There was no significant difference in the cumulative incidence of new onset DM in those who received statin treatment compared to those who did not receive statin treatment (Supplementary Fig. 6, log-rank test P-value = 0.34).

4. Discussion

Although the steatosis–IR–DM–fibrosis relationship in CHC patients is well established in the literature [7–19], there has only been one longitudinal population-based cohort study that evaluated whether DM is associated with progression from non-cirrhotic liver to cirrhosis in CHC patients [19]. In the present hospital-based cohort study relying on careful chart reviews, the results showed via a longitudinal evaluation with a median duration of follow-up of 102.4 months, that baseline DM was associated with progression from non-cirrhotic liver to cirrhosis in non-genotype 3 CHC patients.

A previous study enrolled a total of 4302 HCV-positive patients treated with IFN.

In the patients with DM, the incidence of HCC was found to be decreased when the patients had a mean HbA1c level of <7.0% during follow-up [42]. In the present study, a Kaplan–Meier analysis showed that there was no significant difference in the cumulative incidence of liver cirrhosis in DM patients with a mean HbA1c level of <7.0% compared to those with a mean HbA1c level of ≥7.0%. However, the number of such cases in this study was too small to definitively conclude whether or not improved DM control slowed down the rate of fibrosis progression.

In this study, in order to analyze the effect of antidiabetic drugs on liver outcome, DM patients were further divided into those with diet control, those receiving OAD therapy, and those receiving insulin therapy. A Kaplan–Meier analysis showed that there was no significant difference in the cumulative incidence of liver cirrhosis among these three groups of DM patients. This phenomenon may have been due, however, to the non-compliance of patients. Many patients with DM do not receive the recommended levels of health care due to the inconvenience of insulin therapy.

HCV steatosis occurs in the setting of multiple metabolic abnormalities collectively referred to as HCADS [7]. Of the patients included in this study, 62 developed DM during the follow-up period. DM, antilipid treatment, and antihypertensive treatment were introduced as time-dependent categorical covariates in the Cox regression. The results showed that DM, antilipid treatment, antihypertensive treatment, and baseline obesity were not associated with the risk of liver cirrhosis during the follow-up. A previous study reported that baseline DM was an independent predictor for cirrhosis in CHC patients. In contrast, hypertension, hyperlipidemia, and obesity were not associated with cirrhosis in CHC patients [19]. The results of our study are thus partially consistent with those of the previous study [19].

There is ample evidence that those with HCV infection are exposed to a greater risk of developing type 2 DM [43,44]. Also, the use of statins has been associated with a greater risk of developing type 2 DM [45]. In our cohort, there was no significant difference in the cumulative incidence of new onset DM in those who received statin treatment compared to those who did not receive statin treatment. However, the number of such cases in this study was too small to conclude whether or not the use of statins further increases the diabetogenic risk of HCV alone.

HCV steatosis has a clinical impact in accelerating fibrogenesis. However, the results of this study showed that there was no significant difference in the cumulative incidence of liver cirrhosis between patients with moderate or severe steatosis and patients with no steatosis or mild steatosis as determined by ultrasound during the follow-up. Furthermore, according to the cross-sectional evaluation, the presence of steatosis (>10%) as determined by histology was not associated with advanced fibrosis (Table 2), while it was also not associated with the development of liver cirrhosis according to the longitudinal evaluation (Table 4). In contrast, a previous review paper reported that cross-sectional and prospective studies have shown that fibrosis progression risk is increased in the presence of HCV–steatosis [46]. This discrepancy between the present study and previous studies [46] may be due to different patient characteristics and the inclusion of different covariates in the multivariate analyses.

According to recent evidence, the ultrasonographic fatty liver indicator (US-FLI), correctly identified steatosis rates of as low as 10% (with sensitivity of 90.1% and specificity of 90%) in 352 biopsied patients with various chronic liver diseases [47]. However, the sensitivity of US-FLI in detecting mild steatosis (≥10% steatosis on histology) was only 76.7% in patients with HCV [47]. In summary, ultrasound is not accurate in detecting mild steatosis [48–51]. The fundamental limitations of ultrasound are that individual sonographic features are affected by factors other than steatosis, including patient (coexistent renal disease and obesity) and acquisition (operator, scanner, transducer, and instrument settings) factors [52].

In this study, ultrasound was found to be highly specific in diagnosing cirrhosis, with a specificity of 90.7%. However, the sensitivity was low (60.1%), results which are compatible with those of previous studies [53,54]. A previous study reported that transient elestography (TE) and ultrasound gave complementary information, finding that TE was more accurate than ultrasound for ruling out cirrhosis, while it was less accurate for ruling it in. The combination of TE and ultrasound thus offered the best diagnostic performance [55].

The strengths of the present study are its uses of a baseline histological evaluation of liver damage and a long follow-up period. The main limitation of the study is that it used ultrasound to diagnose liver cirrhosis in the follow-up evaluations. The sensitivity of ultrasound for diagnosing liver cirrhosis is low; thus, the incidence of liver cirrhosis in the study cohort may have been underestimated. However, the specificity of ultrasound for diagnosing liver cirrhosis is high and can be considered sufficient to confirm the diagnosis of liver cirrhosis. That said, further research using the combination of ultrasound and TE for follow-up evaluations would allow for better diagnostic performance in terms of diagnosing liver cirrhosis and so could be helpful for clarifying this issue. Second, the present study was a retrospective study. Height, weight, and waist circumstance were not recorded regularly during the follow-up for the study cohort. Therefore, we could not evaluate the impact of obesity and truncal obesity fluctuations over time on the development of cirrhosis, in spite of the fact that these factors are known to be important factors for fibrosis progression in CHC patients [56–58].

In summary, in this hospital-based cohort study that enrolled non-genotype 3 CHC patients, DM was found to be an independent predictor for advanced fibrosis in the cross-sectional evaluation. Furthermore, baseline DM was still found to be an independent predictor for liver cirrhosis development in the longitudinal evaluation. It can thus be concluded that non-genotype 3 CHC patients with DM are at an increased risk of liver cirrhosis over time during follow-up.

Conflict of interest

None declared.

Funding and Acknowledgements

This study was supported by Grant CMRPG8E1301 from the Chang Gung Memorial Hospital-Kaohsiung Medical Center, Taiwan. The funders had no role in the study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.dld.2018.07.003.

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