Microalbuminuria in hepatitis C-genotype 4: Effect of pegylated interferon and ribavirin

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METHODS: Three hundred subjects, including 233 HCV genotype-4 infected patients, were tested for cryoglobulinemia, microalbuminuria, albumin creatinine ratio (ACR), urea, creatinine, and estimated glomerular filtration rate (eGFR). The parameters were measured again in the HCV patients after 48 wk of treatment with pegylated interferon and ribavirin.

RESULTS: Significantly higher levels of microalbuminuria were detected in HCV-positive patients compared to HCV-negative controls (median 9.5 vs 5.9, respectively, Kruskal-Wallis $P = 0.017$). Log microalbuminuria was significantly correlated with hepatic inflammation ($r = 0.13$, $P = 0.036$) and fibrosis ($r = 0.12$, $P = 0.061$), but not with viral load ($r = -0.03$, $P = 0.610$), or alanine transaminase ($r = -0.03$, $P = 0.617$). Diabetes mellitus neither significantly moderated ($\chi^2 = 0.13$, $P = 0.720$), nor mediated (Sobel test $P = 0.49$) the HCV effect. HCV status was significantly associated with log microalbuminuria ($\chi^2 = 4.97$, $P = 0.026$), adjusting for age, gender, diabetes, cryoglobulinemia, urea and creatinine. A positive HCV status was not significantly associated with low eGFR (< 60 mL/min every 1.73 m$^2$) [odds ratio (OR): 0.5, 95% confidence interval (CI): 0.2-1.4], nor with high ACR (OR: 1.7, 95% CI: 0.7-4.1). End-of-treatment response (ETR) was achieved in 51.9% of patients. Individuals with ETR had significantly lower microalbuminuria post-treatment ($\chi^2 = 8.19$, $P = 0.004$).

CONCLUSION: HCV affected the development of microalbuminuria independent of diabetes or cryoglobulinemia. Combination therapy of pegylated interferon-ribavirin had a positive effect in reducing microalbuminuria.

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Key words: Hepatitis C virus; Genotype; Kidney diseases; Albuminuria; Proteinuria; Peginterferon α-2a; Ribavirin

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INTRODUCTION

Hepatitis C virus genotype 4 (HCV-G4) is prevalent in the Middle East and Africa and has spread to several regions in Europe\(^3\). HCV infection is associated with several renal diseases including mixed essential cryoglobulinemia, membranoproliferative glomerulonephritis and, less frequently, membranous nephropathy and crescentic glomerulonephritis\(^2\). HCV is a significant cause of glomerulopathy in countries with a high prevalence of HCV infection\(^\circ\). Several studies have postulated a causal link between HCV infection and renal diseases through the induction of cryoglobulinemia\(^1\). The affinity for the kidney mesangium appears to be a major factor responsible for the precipitation of type-II mixed cryoglobulins in glomerular structures and the ensuing damage\(^2\). The principal clinical manifestations of glomerular disease in HCV-infected patients are the presence of proteinuria and microscopic hematuria with or without impaired kidney function\(^8\). Diabetes mellitus-mediated glomerulonephropathy in HCV patients may be considered, particularly with epidemiological studies showing a high occurrence of type 2 diabetes in patients with chronic HCV-G4\(^1\). Various approaches have been tried for the treatment of HCV-associated glomerulonephritis, including immunosuppressive therapy (corticosteroids and cytotoxic agents), plasma exchange and antiviral agents. Limited data exist regarding antiviral treatment of HCV-associated glomerulonephritis.

We present a novel study reporting the prevalence of microalbuminuria, with other measures of renal insufficiency [albumin creatinine ratio (ACR), estimated glomerular filtration rate (GFR)] in HCV-G4 patients compared to HCV-negative controls, and the effect of viral load on liver histology, microalbuminuria, and renal insufficiency. Also, we investigated the effect of antiviral therapy with pegylated interferon and ribavirin on microalbuminuria, with other measures of renal insufficiency.

MATERIALS AND METHODS

In this prospective study, 300 consecutive eligible subjects who attended the gastroenterology clinic in Hamad Hospital were recruited in the period between January 2006 and 2009. Of these, 233 were chronic HCV-G4 patients who fulfilled the inclusion criteria, and 77 were control subjects who were either non hepatic patients followed in the clinic during the same period or healthy volunteers who were referred to Hamad hospital when they were ill. Controls were eligible if they did not have liver disease, evidenced by: persistently normal transaminases, negative serology for hepatitis serology, negative screening for auto-immune disease and no history of alcohol consumption, normal platelet count and α fetoprotein, and normal ultrasound scans.

All study participants were seen 2-3 times to provide repeated blood samples at baseline. Chronic HCV patients were also seen periodically to assess the response to treatment. All patients provided written informed consent as stated in the Declaration of Helsinki of 1979, and the ethics research committee of the Hamad Medical Corporation provided ethical approval.

Chronic HCV infection was evidenced by persistently increased alanine aminotransferase (ALT) levels, positive serology for anti-HCV, active virus replication by detection of HCV RNA in serum, and histological findings of chronic active hepatitis according to the Scheuer score. The patients were excluded if they had: (1) concurrent hepatitis B or human immunodeficiency virus infections, autoimmune hepatitis, hemochromatosis, or Wilson’s disease; (2) systemic hypertension or if they reported current use of antihypertensive medication; (3) active alcohol consumption; (4) antiviral or corticosteroid treatment; and (5) chronic renal disease or history of dialysis.

The HCV patients who were candidates for treatment were given 180 μg of peginterferon α-2a (Pegasys®; Hoffmann-LaRoche, Basel, Switzerland) subcutaneously once weekly and ribavirin (Copegus®; Hoffmann-La Roche, Basel, Switzerland) at a daily oral dose of 1000 mg (body weight < 75 kg) or 1200 mg (body weight > 75 mg) for 48 wk.

Laboratory measurement

Testing for anti-HCV was carried out using a commercial ELISA kit (Assym HCV version 3.0; Abbott Laboratories, Chicago, IL, USA). All patients were HCV-G4 as detected by the Inno-LiPA HCV II assay (Innogenetics Inc., Alpharetta, GA, USA). Monitoring serum HCV RNA levels was by Amplicor (version 2.0, Hoffmann-La Roche) with a minimum detection limit of 50 IU/mL.

Microalbuminuria was measured on a spot of second morning urine after an overnight fast, taking the mean of at least 2 samples collected for each subject. Urine albumin measurements were obtained by an automated immunoturbidimetric assay (Roche Hitachi 902, Roche Diagnostics, Indianapolis, IN 46250 USA).

For quantitative determination of creatinine in serum or urine, creatinine blanked kinetic Jaffe (Roche Diagnostics, Hitachi 917, Modulator P analyzer Roche Diagnostics) was used. We estimated GFR (eGFR) using the abbreviated modification of diet in renal disease equation.

After an overnight fast, samples for cryoglobulin measurement were collected at a temperature of 37°C, centrifuged at 37°C for 10 min, and the serum separated; 5 mL of the serum was allowed to stand in a cryocrit tube at 4°C for 2-7 d, with formation of precipitate confirmed visually. If the test was positive we proceeded to electrophoresis and immunofixation for typing. All subjects were tested for cryoglobulin.

Measures of renal insufficiency

Three measures of renal insufficiency were examined, namely eGFR, microalbuminuria, and serum creatinine. Low eGFR was defined as eGFR < 60 mL/min every 1.73 m\(^2\). The presence of microalbuminuria was tested using (1) albuminuria level, or categories of microalbuminuria, defined as individuals with an albuminuria level higher than the upper tertile of the albuminuria level among controls.
The study population consisted of 300 subjects, of whom 233 (77.7%) were HCV-positive. At enrolment 138 (46.6%) were diabetics. The majority of the study participants [239 (79.9%)] were male and 195 (81.6%) of the males were HCV-positive compared with 37 (61.7%) of females. Median age of the study participants was 46 years (inter-quartile range, 41-53 years). The histopathological changes of the liver in HCV patients were classified according to the Scheuer score, and fibrotic changes of Stages I, II, III and IV were seen in 58 (25.1%), 89 (38.3%), 60 (25.7%) and 26 (10.9%) patients, respectively. Necro-inflammatory changes were seen in 23 (9.9%), 109 (46.8%), 58 (25.1%) and 4 (1.7%) patients, respectively. A median microalbuminuria of 8.2 (inter-quartile range, 4.4-16.9) µg/mg was observed, while the median eGFR was 98 (inter-quartile range, 85-112) mL/min every 1.73 m². One hundred and eighteen individuals had microalbuminuria (39.7%), including 23 (37.6%) HCV-positive non-diabetic, 58 (49.6%) HCV-positive diabetic, 7 (6.0%) control non-diabetic, and 8 (6.8%) control diabetic subjects. Forty five individuals had high ACR (15.8%), including 13 (29.6%) HCV-positive non-diabetic, 5 (11.1%) HCV-positive diabetic, 3 (2.0%) control non-diabetic, and 2 (4.6%) control diabetic subjects. Of 85 HCV patients who were on treatment, 44 (51.8%) showed an ETR.

Table 1 demonstrates the study participants’ characteristics by HCV status. HCV-positive individuals had significantly higher ALT levels and higher microalbuminuria. No difference was detected in age, body mass index, urea, creatinine or gender (Table 1).

### RESULTS

The study population consisted of 300 subjects, of which 233 (77.7%) were HCV-positive. At enrolment 138 (46.6%) were diabetics. The majority of the study participants [239 (79.9%)] were male and 195 (81.6%) of the males were HCV-positive compared with 37 (61.7%) of females. Median age of the study participants was 46 years (inter-quartile range, 41-53 years). The histopathological changes of the liver in HCV patients were classified according to the Scheuer score, and fibrotic changes of Stages I, II, III and IV were seen in 58 (25.1%), 89 (38.3%), 60 (25.7%) and 26 (10.9%) patients, respectively. Necro-inflammatory changes were seen in 23 (9.9%), 109 (46.8%), 58 (25.1%) and 4 (1.7%) patients, respectively. A median microalbuminuria of 8.2 (inter-quartile range, 4.4-16.9) µg/mg was observed, while the median eGFR was 98 (inter-quartile range, 85-112) mL/min every 1.73 m². One hundred and eighteen individuals had microalbuminuria (39.7%), including 23 (37.6%) HCV-positive non-diabetic, 58 (49.6%) HCV-positive diabetic, 7 (6.0%) control non-diabetic, and 8 (6.8%) control diabetic subjects. Forty five individuals had high ACR (15.8%), including 13 (29.6%) HCV-positive non-diabetic, 5 (11.1%) HCV-positive diabetic, 3 (2.0%) control non-diabetic, and 2 (4.6%) control diabetic subjects. Of 85 HCV patients who were on treatment, 44 (51.8%) showed an ETR.

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### Outcomes and covariates definitions

Subjects were considered diabetic if they had a fasting blood glucose level ≥ 5.6 mmol/L, or self-reported as being diabetic or on anti-diabetic medication. End-of-treatment response (ETR) was defined as loss of detectable serum HCV RNA at the end of treatment. Normal ALT was defined as ALT ≤ 40 U/L for women and ≤ 40 U/L for men.

### Statistical analysis

Unadjusted association between renal insufficiency and HCV infection was evaluated. For categorical variables, the unadjusted association was tested using the Chi-square test (Fisher exact test if there was limited sample size thus violating the Chi-square test assumptions), while for continuous variables, analysis of variance or the Kruskal-Wallis test (when the assumption of normality was violated) were employed. Multivariate logistic regression (for categorical variables) and multivariate linear regression (for continuous variables) were used to test for the null hypothesis of no significant association between renal insufficiency and HCV infection controlling for possible confounders and covariates. Microalbuminuria, ACR and eGFR data were log transformed to adjust for skewness.

We tested for the presence of an interaction between HCV and diabetes, by adding the cross product term of HCV × diabetes in addition to HCV and diabetes in the regression model predicting microalbuminuria.

In addition, we employed mediation analysis to test whether the effect of HCV on the risk of developing microalbuminuria was mediated by diabetes. Baron and Kenny’s criteria was used to assess the presence of mediation. Mediation was expected if diabetes accounted for part or all of the relationship between HCV and microalbuminuria, as manifested by a decrease in the magnitude of the previously significant association between HCV and microalbuminuria upon controlling for diabetes. The significance of the mediation pathway was tested using the Sobel test as described elsewhere.

To test for treatment effect, we examined differences between pre- and post-treatment using the Wilcoxon Signed Rank test which is a non-parametric equivalent of the paired t-test. In the multivariate analysis we tested for significant predictors of microalbuminuria post-treatment compared to pre-treatment, using the generalized estimating equation model.

All analyses were conducted using SAS 9.1 software.
Microalbuminuria
Levels of microalbuminuria were significantly higher among HCV-positive individuals than HCV-negative patients (median 9.5 vs 5.9, respectively, Kruskal-Wallis $P = 0.017$). A significantly higher prevalence of microalbuminuria (defined as albuminuria $> 0.175$ mg/mmol) was observed among HCV-positive individuals (53.7%) compared to HCV-negative individuals (31.8%), ($\chi^2 = 9.8, P = 0.002$).

Log microalbuminuria was significantly correlated with grade ($r = 0.13, P = 0.036$), borderline correlated with older age ($r = 0.11, P = 0.069$) and more fibrosis ($r = 0.12, P = 0.061$), but not significantly associated with viral load ($r = -0.03, P = 0.610$), or ALT levels ($r = -0.03, P = 0.617$).

There was no significant interaction between HCV status and diabetes such that the odds ratio (OR) of microalbuminuria among diabetics compared to non-diabetics was not significantly higher in HCV-positive individuals compared to HCV-negative controls ($P$ for interaction $= 0.720$) (Table 2).

For sensitivity analysis we restricted the analysis to non-diabetics. Among non-diabetics, the prevalence of microalbuminuria was significantly higher in HCV-positive individuals (50%) compared to HCV-negative controls (25.8%) ($\chi^2 = 5.9, P = 0.015$). Restricting the analysis to individuals with no cryoglobulinemia revealed that microalbuminuria was significantly higher in HCV-positive individuals (53.3%) compared to HCV-negative controls (31.8%) ($\chi^2 = 9.4, P = 0.002$).

To adjust for potential confounders and important covariates we employed multivariate regression to test for the significance of HCV as a predictor for microalbuminuria. Log microalbuminuria was used in the linear regression analysis because of the skewedness of the microalbuminuria data. Similar to the unadjusted analysis, we did not detect a significant interaction between diabetes and HCV ($\chi^2 = 1.2, P = 0.272$), thus no interaction term was included in the final model.

After adjusting for age, gender, diabetes, cryoglobulinemia, urea and creatinine, there was a significant association between HCV status and log microalbuminuria ($\chi^2 = 4.97, P = 0.026$). Microalbuminuria was significantly associated with urea ($\chi^2 = 8.2, P = 0.004$), creatinine ($\chi^2 = 27.0, P < 0.0001$), diabetes ($\chi^2 = 8.2, P = 0.004$), but not with age ($\chi^2 = 0.0, P = 1.0$) or gender ($\chi^2 = 0.4, P = 0.530$) or cryoglobulinemia ($\chi^2 = 0.2, P = 0.703$).

We tested whether diabetes mediated the effect of HCV on microalbuminuria risk. Diabetes was not a significant mediator of the effect of HCV on microalbuminuria risk, as manifested by the non fulfillment of the Baron and Kenny criteria for mediation, such that HCV did not significantly predict diabetes ($\chi^2 = 0.56, P = 0.45$), and therefore the Sobel test for mediation was not significant ($P = 0.49$).

Albumin creatinine ratio (ACR)
Median ACR was 1.0 (range, 36.8) mg/mmol in HCV-negative subjects vs 1.0 (range, 193.9) mg/mmol in HCV-positive subjects. Positive HCV status was not associated with a significantly high ACR [OR: 1.7, 95% confidence interval (CI): 0.7-4.1], which also did not change by stratifying according to diabetic status ($P$ for interaction $= 0.752$) (Table 2).

In multivariable regression analysis, and adjusting for age and gender, there was no significant association between HCV status and high ACR ($\chi^2 = 0.86, P = 0.35$).

eGFR
The prevalence of low eGFR $< 60$ mL/min every 1.73 m$^2$ was lower in HCV-positive individuals than HCV-negative individuals (4.4% vs 8.9%). Positive HCV status was not associated with significantly low eGFR $< 60$ mL/min every 1.73 m$^2$ (OR: 0.5, 95% CI: 0.2-1.4). Median eGFR was 99.5 (range, 317.0) mL/min every 1.73 m$^2$ in HCV-negative subjects and was 98.0 (range, 155.0) mL/min every 1.73 m$^2$ in HCV positive subjects. We did not detect a significant interaction between HCV status and diabetic status ($P$ for interaction $= 0.175$) (Table 2).

Multivariate regression analysis revealed that, upon adjusting for age, and gender there was no significant association between HCV status and low eGFR ($\chi^2 = 1.12, P = 0.29$).

Serum creatinine
Median creatinine was 0.75 (range, 248) mg/dL in HCV negative subjects, vs 0.77 (range, 230) mg/dL in HCV positive subjects. Negative HCV status was borderline associated with high serum creatinine (OR: 2.6, 95% CI: 1.02-6.68, $P = 0.05$). There was no statistically significant difference in serum creatinine comparing diabetics to non-diabetics among HCV positive and HCV negative controls (Table 2).

In the multivariable regression analysis, and adjusting for age, and gender, there was significant association between HCV negative status and high serum creatinine (OR: 3.3, 95% CI: 1.2-9.2, $P = 0.02$).

Table 2  Renal insufficiency in diabetics compared with non-diabetics among HCV-positive and HCV-negative controls

| Renal function                  | Diabetes | OR (95% CI) | $P$-value$^1$ |
|--------------------------------|----------|-------------|---------------|
| Microalbuminuria                |          |             |               |
| HCV-positive                    |          | 1.4 (0.8-2.3)| 0.720         |
| HCV-negative                    |          | 1.7 (0.6-5.1)| 0.175         |
| eGFR                            |          |             |               |
| HCV-positive                    |          | 3.0 (0.8-11.9)| 0.752         |
| HCV-negative                    |          | 0.6 (0.1-4.0)| 0.627         |
| Albumin creatinine ratio        |          |             |               |
| HCV-positive                    |          | 2.9 (1.4-6.1)| 0.627         |
| HCV-negative                    |          | 2.1 (0.4-12.7)| 0.627         |
| Creatinine                      |          |             |               |
| HCV-positive                    |          | 0.9 (0.3-2.8)| 0.627         |
| HCV-negative                    |          | 0.5 (0.1-2.5)| 0.627         |

$^1$ For interaction is the $P$-value of the cross product term of HCV $\times$ diabetes in the logistic regression models predicting renal insufficiency. OR: Odds ratio.
**Microalbuminuria and treatment effect**

Log microalbuminuria pre-treatment median level was 2.26 (inter-quartile range, 1.5-3.1), while median post-treatment level was 2.04 (inter-quartile range 1.5-2.6), (Wilcoxon signed rank test, P = 0.09). Adjusting for age, sex, fibrosis, grade, log ACR, ALT, diabetes and viral load the decline was more pronounced in individuals with ETR compared to individuals without ETR ($\chi^2 = 8.19$, $P = 0.004$).

The pre-to post-treatment log microalbuminuria difference was significantly correlated with pre-treatment older age ($r = 0.37$, $P < 0.001$), fibrosis ($r = 0.26$, $P = 0.017$), grade ($r = 0.23$, $P = 0.042$) and log ACR ($r = 0.38$, $P < 0.001$), but not correlated with male gender ($r = 0.14$, $P = 0.222$), diabetes ($r = 0.12$, $P = 0.265$), urea ($r = 0.015$, $P = 0.896$), creatinine ($r = -0.05$, $P = 0.658$) or ALT ($r = -0.07$, $P = 0.508$).

In multivariate regression, after adjusting for gender, age, pre-treatment ALT, log ACR, diabetes, fibrosis and grade, only log ACR, ETR, and fibrosis were moderately associated with a greater decline in log microalbuminuria post-treatment ($\chi^2 = 8.98$, $P = 0.003$; $\chi^2 = 8.19$, $P = 0.004$; $\chi^2 = 9.35$, $P = 0.053$, respectively), while age, gender, ALT, diabetes, and grade were not associated with log microalbuminuria decline ($\chi^2 = 0.70$, $P = 0.401$; $\chi^2 = 0.13$, $P = 0.718$; $\chi^2 = 1.31$, $P = 0.253$; $\chi^2 = 0.0$, $P = 0.969$; $\chi^2 = 1.33$, $P = 0.722$, respectively).

**DISCUSSION**

Hepatitis C infection is known to have a higher prevalence of some components of metabolic syndrome and to be associated with chronic renal disease. Renal involvement in the course of HCV infection is attributed to a high incidence of intrinsic diabetic renal disease or cryoglobulinemia. Studying microalbuminuria in HCV-G4 patients and its relationship to response to treatment is a novel report, especially after recent evidence for diabetes-inducing effects of HCV-G4[10].

In the current study, using the same definition of microalbuminuria as Liangpunsakul et al[11], the prevalence of microalbuminuria in HCV-G4 was 20%, similar to that reported by the Third National Health and Nutrition Examination Survey (12.4%). In contrast to the limitations of the NHANES III study, we were able to study the mean of multiple microalbuminuria readings, adjusting for stage of hepatic fibrosis, grade of inflammation, viral load and cryoglobulinemia.

In our study, not only was the prevalence of microalbuminuria higher among HCV-positive individuals but significantly higher levels were noted compared to non-HCV subjects. Although the prevalence of microalbuminuria was higher among diabetic HCV patients, testing for the effect of diabetes did not reveal a significant interaction with HCV infection nor a significant mediation of the HCV effect. In contrast to a previous suggestion of a link between HCV infection and diabetes[12], our results revealed that HCV infection was not associated with type 2 diabetes mellitus. Our results are in accordance with other reports that revealed no interaction between HCV infection and diabetes, and the association between HCV infection and albuminuria was not altered upon adjustment for diabetes[13,14]. In addition HCV infection was not associated with cryoglobulinemia. Therefore, our results suggested that HCV-G4 was associated with microalbuminuria independently of cryoglobulinemia or diabetes status.

HCV-induced glomerulonephropathy in the absence of cryoglobulinemia was explained in previous reports by direct or indirect pathways: deposition in the glomerulus of a monoclonal IgM rheumatoid factor with particular affinity for the glomerular matrix, which is produced by permanent clones of B lymphocytes infected by the virus or immune complexes composed of HCV antigens and anti-HCV IgG antibodies can deposit directly in the glomerular structures in the absence of a concomitant type II mixed cryoglobulin. HCV RNA genomic sequences and HCV core protein detected in kidney glomerular and tubular structures point to distinct pathways of HCV-related damage in glomeruli and tubules[15]. Kidney disorders constitute patho- and morphogenesis of systemic infection in HCV[16].

In spite of HCV-induced microalbuminuria, which was in agreement with Liangpunsakul et al[11], Moe et al[17] and Tsui et al[18] but in contrast to Dalrymple et al[19] and Tsui et al[18], we did not find an increased risk for renal disease in HCV-G4 as reflected by undetectable changes in creatinine, ACR or eGFR. This discrepancy might be explained by long-term infection in some studies, the definition of renal insufficiency, or the viral genotype. These findings suggested that the principal clinical manifestation of glomerular disease in HCV-G4-infected patients is the presence of microalbuminuria without impaired kidney function[19].

The lack of increased risk of renal disease persisted even after restricting the analysis to diabetics which is consistent with a previous report[20].

We found a significant correlation between microalbuminuria and necroinflammatory changes, but not fibrotic changes; this represents a relation with viral activity rather than progression of liver disease. In the absence of a significant correlation between microalbuminuria and viral load, hepatic fibrotic changes or platelet count, we suggest that HCV infection per se in HCV-G4 and not the stage of liver disease is the cause of microalbuminuria.

There was a moderate reduction in microalbuminuria after pegylated interferon therapy which was more pronounced in patients with an ETR, those with higher pre-treatment fibrosis and higher pre-treatment log ACR, regardless of the grade of inflammation, diabetes or liver function. This post-treatment reduction in proteinuria in HCV-G4 indirectly suggested an improvement in renal function. This post-treatment reduction in proteinuria in HCV-G4 patients and its relationship to response to treatment level was 2.04 (inter-quartile range 1.5-2.6), (Wilcoxon signed rank test, $P = 0.09$). Adjusting for age, sex, fibrosis, grade, log ACR, ALT, diabetes and viral load the decline was more pronounced in individuals with ETR compared to individuals without ETR ($\chi^2 = 8.19$, $P = 0.004$).
In conclusion, renal disorders in the form of microalbuminuria increased in HCV-G4 infection, especially in older patients, regardless of the stage of liver disease or viral load. In our study, diabetes did not mediate or moderate the effect of HCV infection, suggesting that HCV independently affected the development of microalbuminuria. Combination therapy of pegylated interferon-ribavirin had a positive effect in reducing microalbuminuria.

COMMENTS

Background
Hepatitis C virus (HCV) is a significant cause of glomerulopathy in countries with a high prevalence of HCV. The principal clinical manifestations of glomerular disease in HCV-infected patients are the presence of proteinuria and microscopic hematuria with or without impaired kidney function.

Research frontiers
Microalbuminuria is observed in HCV-positive individuals. However, the prevalence of microalbuminuria in HCV-genotype 4 (HCV-G4) patients compared to HCV-negative controls and its association with liver histology, viral load, and response to treatment has not been indisputably addressed. In this study, the authors demonstrate that antiviral therapy with pegylated interferon and ribavirin could have a beneficial effect on microalbuminuria.

Innovations and breakthroughs
Latest reports have provided evidence of a significant increase in renal disease in HCV infection, in particular HCV-induced microalbuminuria. This is the first study to report the increase in microalbuminuria in HCV-G4 patients. Furthermore, the study demonstrated that antiviral therapy with pegylated interferon and ribavirin could have a beneficial effect on the microalbuminuria.

Applications
Understanding how microalbuminuria is related to HCV infection, grade, fibrosis and response to treatment, may provide an insight for future strategy for post-treatment follow-up and for predicting response to treatment.

Terminology
Microalbuminuria, estimated glomerular filtration rate, and serum creatinine are measures of renal insufficiency. End-of-treatment response is the loss of detectable serum HCV RNA at the end of treatment.

Peer review
In the current study, the authors demonstrated the independent effect of HCV-G4 infection on the development of microalbuminuria through prospective comparison to HCV-negative control groups and the positive effects of antiviral therapy. This study included some interesting points.

REFERENCES
1 Kamal SM. Hepatitis C genotype 4 therapy: increasing options and improving outcomes. Liver Int 2009; 29 Suppl 1: 39-48
2 Perico N, Cattaneo D, Bikbov B, Remuzzi G. Hepatitis C infection and chronic renal diseases. Clin J Am Soc Nephrol 2009; 4: 207-220
3 Yamabe H, Johnson RJ, Gretch DR, Fukushima K, Osawa H, Miyata M, Inuma H, Sasaki T, Kaizuka M, Tamura N. Hepatitis C virus infection and membranoproliferative glomerulonephritis in Japan. J Am Soc Nephrol 1995; 6: 220-223
4 Lo KY, Chen CY, Lee CS. Hepatitis C virus-associated type II mixed cryoglobulinemia vasculitis complicated with membranous proliferative glomerulonephritis. Ren Fail 2009; 31: 149-152
5 Nishiyama Y, Shimatsu A, Arakawa M, Nagao M, Saito A, Kido A, Kaganeya H, Wada T, Okuda T, Ichiyama S. Characterization of cryoglobulin M protein, low molecular weight IgM in a patient with chronic hepatitis C and type II mixed cryoglobulinemia. Rinsho Byori 2001; 49: 1139-1145
6 Sabry A, E-Agroudy A, Sheashaa H, El-Husseini A, Mohamed Tah N, Elbaz M, Sobh M. HCV associated glomerulopathy in Egyptian patients: clinicopathological analysis. Viralology 2005; 334: 10-16
7 Chehadeh W, Abdella N, Ben-Nakhi A, Al-Arouj M, Al-Nakib W. Risk factors for the development of diabetes mellitus in chronic hepatitis C virus genotype 4 infection. J Gastroenterol Hepatol 2009; 24: 42-48
8 Baron RM, Kenny DA. The moderator-mediator variable distinction in social psychological research: conceptual, strategic, and statistical considerations. J Pers Soc Psychol 1986; 51: 1173-1182
9 Jasti S, Dudley WN, Goldwater E. SAS macros for testing statistical mediation in data with binary mediators or outcomes. Nurs Res 2008; 57: 118-122
10 Mohamed MK. Risk factors for the development of diabetes mellitus in chronic HCV genotype 4 infection. J Gastroenterol Hepatol 2009; 24: 11-18
11 Liangpunsaikul S, Chalasani N. Relationship between hepatitis C and microalbuminuria: results from the NHANES III. Kidney Int 2005; 67: 285-290
12 Kamar N, Izopet J, Alic R, Buemi-Manfrini C, Rostaing L. Hepatitis C virus-related kidney disease: an overview. Clin Nephrol 2008; 69: 149-160
13 Dalrymple LS, Koespel T, Sampson J, Louie T, Dominitz J, Young B, Kestenbaum B. Hepatitis C virus infection and the prevalence of renal insufficiency. Clin J Am Soc Nephrol 2007; 2: 715-721
14 Tsui JI, Vittinghoff E, Shlipak MG, Bertenthal D, Inadomi J, Rodriguez RA, O’Hare AM. Association of hepatitis C seropositivity with increased risk for developing end-stage renal disease. Arch Intern Med 2007; 167: 1271-1276
15 Sansonno D, Lauetta G, Montrone M, Grandaliano G, Schena FP, Dammacco F. Hepatitis C virus RNA and core protein in kidney glomerular and tubular structures isolated with laser capture microdissection. Clin Exp Immunol 2005; 140: 498-506
16 Tov NL, Nepomnyashchikh LM, Aidagulova SV, Onishchenko AA. Ultrastructure of kidney cell population in patients with markers of HCV- and HBV-infections (analysis of biopsy specimens). Bull Exp Biol Med 2004; 138: 624-628
17 Moe SM, Pampalone AJ, Ofner S, Rosenman M, Teal E, Hui SL. Association of hepatitis C virus infection with prevalence and development of kidney disease. Am J Kidney Dis 2008; 51: 885-892
18 Tsui JI, Vittinghoff E, Shlipak MG, O’Hare AM. Relationship between hepatitis C and chronic kidney disease: results from the Third National Health and Nutrition Examination Survey. J Am Soc Nephrol 2006; 17: 1168-1174
19 Fabrizi F, Messa P, Martin P, Takkouche B. Hepatitis C virus infection and post-transplant diabetes mellitus among renal transplant patients: a meta-analysis. Int J Artif Organs 2008; 31: 675-682
20 Poussier A, Lebourvier M, Penformis A, Di Martino V, Buffier P, Vorges B, Hillon P, Petit JM. Specific phenotype associated with diabetes mellitus secondary to chronic hepatitis C infection. Diabet Med 2008; 25: 1237-1240
21 Abbas G, Hussein S, Shafi T. Effect of antiviral therapy on hepatitis C virus related glomerulopathy. Saudi J Kidney Dis Transpl 2008; 19: 775-780

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