Hepatitis C Virus Heterogeneity: Lipoprotein and Immunoglobulin Binding and Clinical Status

Hepatitis C Virus (HCV) infection is a major cause of liver disease globally.[1] More than 350,000 people die from hepatitis C-related liver diseases each year.[2] Persistent HCV infection can progress to liver cirrhosis and hepatocellular carcinoma.[1] Thus, HCV infection is a major cause of morbidity and mortality worldwide.[3] The range of liver disease severity in individual patients with HCV infection is broad. The spectrum of liver disease in patients infected with HCV ranges from asymptomatic carriers, patients with chronic hepatitis of variable severity to end-stage liver cirrhosis.[4] The rate of progression to the cirrhotic stage also varies widely amongst HCV-infected individuals.

Studies by our group and others[5,6] have demonstrated that the low density HCV fraction represents the "infectious, active" fraction in plasma, and consists of HCV bound to low density lipoprotein (LDL). Agnello and colleagues proposed that the LDL receptor plays a role in the cellular entry of HCV into hepatocytes.[7] Further studies support the role of the LDL receptor in HCV cellular entry and infection.[8] In acute HCV infection, viral RNA is linked with plasma LDL, and in chronic disease states, viral RNA is linked with the high density fraction in plasma, which is thought to be immunoglobulin-bound.[5] An intermediate density fraction has also been identified more recently, which is likely to be lipoprotein-bound.[9] To further investigate the idiosyncratic relationship between HCV, LDL and IgG, we attempted to determine whether high, intermediate and low density HCV RNA levels in plasma are related to the clinical status of patients with liver disease.

HCV RNA titer from six HCV-infected patients with varying severities of liver disease were analyzed by Differential Flotation Ultracentrifugation and HCV RNA quantification, according to the techniques reported by Watson and colleagues[5] and Pumeechockchai and colleagues.[10] The subjects' clinical presentation was compared to the differential HCV density fractions. Baseline and sequential liver function tests were performed and five out of six patients proceeded to liver biopsy for clinical purposes. Hepatitis C virus serology and genotype data were performed (Table 1).

All patients, except Patient 5, had mild disease as demonstrated on liver biopsy. Patient 5 had severe liver disease, including cirrhosis and hepatocellular carcinoma. Patients 1–4 demonstrated inter-patient differences in the amount of quantitative HCV in high density (immunoglobulin-bound) fraction, and low and intermediate density (lipoprotein-bound) fractions. However, this was not statistically significant between patients or within individual patients. Patients 5 and 6 were found to have negative quantitative HCV PCR, thus, fractions could not be determined in these patients. This may be due to difficulties in the RNA extraction process after ultracentrifugation.

Table 1 Clinical and serological data of the hepatitis C infected patients analyzed.

Patient	Sex	Age	HCV genotype	Liver biopsy	Total HCV titer	LD	ID	HD
1	M	47	1*	AG 1 FS1	5.53	4.00	3.67	3.85
2	M	48	1*	AG 1 FS 1	5.92	5.14	5.62	4.47
3	F	51	1a	AG 1 FS 1	5.58	4.20	4.19	3.78
4	M	64	1b	AG 1 FS 0	6.38	5.28	5.07	4.64
5	M	47	3a	ND	0.00	0.00	0.00	0.00
6	F	37	1a	AG 1 FS 1	0.00	0.00	0.00	0.00

Key; AG: activity grade, FS: fibrosis stage, ND: not done, Total HCV titer: Log_{10}total HCV titer in the unfractionated specimen before ultracentrifugation (viral copies/ml), LD: Log_{10} low density HCV RNA (viral copies/ml), ID: Log_{10} intermediate density HCV RNA (viral copies/ml), HD: Log_{10} high density HCV RNA (viral copies/ml), 1*: genotype 1 but subtype unclassified in this patient.

The results of our small pilot study do not clearly demonstrate an association between the severity of clinical liver disease and buoyant density of HCV RNA fractions. A major limitation was the small sample size. However, the technique of differential ultracentrifugation, HCV RNA extraction and viral quantification is extremely labor intensive, which would render a larger sample size difficult. This study demonstrates the complexity of the relationship between HCV infection and LDL binding in the human host. Further investigation of this idiosyncratic association is required.

CONFLICTS OF INTEREST

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