

# Steatosis and Chronic Hepatitis C Virus Infection

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### Abstract

**Objective:** To assess the presence of steatosis and other histological changes in patients with chronic hepatitis C.

**Methods:** Liver biopsy samples were reviewed for presence of steatosis, its degree and other histological changes of hepatitis C including necro-inflammatory score, fibrosis grade and these changes were correlated with liver function tests.

**Results:** A total of 109 liver biopsy samples were reviewed. Mean age of the patients was  $44.46 \pm 13.93$  years and 62 (56.8%) were male. Mean necro-inflammatory score was  $2.32 \pm 0.95$ . Mean fibrosis grade was  $1.69 \pm 1.12$ . No Steatosis was found in 42 (38.5%) samples and mild to severe degree of steatosis was found in 67 (61.5%) samples.

**Conclusion:** In this study, nearly 62% of liver biopsy samples had some degree of steatosis. Whether this steatosis increases occurrence of severe form of fibrosis or is an associated phenomena with chronic inflammation needs to be further clarified (JPMA 54:110;2004).

### Introduction

Chronic hepatitis C (CHC) virus infection is a predominant cause of chronic liver disease not only in many parts of the world but also in Pakistan.<sup>1,2</sup> In certain public hospitals, the presentation of chronic hepatitis C virus (HCV) infection has been in the form of chronic liver disease and they have presented in advanced condition with the complications of portal hypertension and hepatic failure.<sup>1</sup> Hepatitis C in our country has become a major cause of chronic liver disease. The sources of spread include unsafe injections and body piercing including tattooing and acupuncture, in addition to improperly screened blood and blood products.<sup>3</sup> Histological features of chronic hepatitis C have been well documented which include lymphoid follicles and aggregates, bile duct injury and fibrosis.<sup>4-6</sup> However, these histological features of hepatitis may differ in various countries and various locations.<sup>7,8</sup>

Steatosis as a part of histological feature of chronic hepatitis C has been described<sup>9</sup> and epidemiologic studies have shown that HCV-related steatosis correlates with both patient factors, such as obesity, as well as various viral factors such as HCV genotype.<sup>9-11</sup> The degree of steatosis has been linked to the extent of hepatic fibrosis and patients with steatosis and genotype 3 were found to be at risk of accelerated fibrosis<sup>10</sup> and an implication that steatosis may be contributing to the disease progression in CHC infection.<sup>9</sup> Furthermore, it was noted that hepatitis C genotype 3 may interfere with pathways of hepatic lipid metabolism<sup>11</sup> and oxidative damage may lead to steatosis in CHC.<sup>9</sup> In addition to all this, it has also been noted that steatosis may play an important role in response to HCV therapy.<sup>9</sup>

The aim of this study was to review the histological features especially the steatosis in liver biopsies performed on patients of chronic hepatitis C in past 4 years at Shifa International Hospital Islamabad.

### Material and Methods

Liver biopsy samples were reviewed which were taken from hepatitis C patients who had elevated ALT and positive HCV RNA (PCR) during 1998 to 2002. Biopsy samples were obtained using Menghini needles or in several cases a spinal needle.<sup>12</sup> The specimens were processed with 10% formalin and were stained by standard methods of hematoxylin and eosin. For evaluation of fibrosis, trichrome stain was used which highlights collagen tissue around hepatocytes.

All the biopsy material was interpreted by a single pathologist (FI) using METAVIR score. This scoring has four stages of fibrosis, stage 1 being the early changes and stage 4 being cirrhosis.<sup>9</sup> Presence of lymphoid aggregate or lymphoid follicle, bile duct injury and necro-inflammatory score and fibrosis grades were noted. Steatosis was also noted and its presence and severity was recorded. Degree of steatosis was categorized as mild, moderate and severe.

### Statistical Analysis

Categorical variables are given as means  $\pm$  standard deviations. Continuous variables were assessed with students 't' test and 95% confidence intervals were calculated. Spearman correlation was used to assess correlation. Statistical significance was considered to be present with the  $p < 0.05$ . Statistical software package SPSS 10 (SPSS, Chicago, Illinois, USA) were used to make all statistical calculations.

## Results

A total of 109 liver biopsy samples were reviewed. The mean age was  $44.46 \pm 13.9$  years. Males were 62 (56.8%) and females were 47 (43.2%). Relevant information for liver function test, necro-inflammatory score and fibroses grade are shown in Table 1.

**Table 1. Demographic features of CHC patients (n=109).**

Age (years)	$44.46 \pm 13.93$ (95% CI 41.82 - 47.11)
Sex	62 Males (56.8%) 47 Females (43.2%)
Total Bilirubin (mg/dl)	$2.77 \pm 9.85$ (95% CI 0.55 - 4.99)
ALT (IU/L)	$113 \pm 109$ (95% CI 90 - 136)
AST (IU/L)	$109 \pm 96$ (95% CI 87 - 2.50)
Necro-inflammatory Score	$2.32 \pm 0.95$ (95% CI 2.14 - 2.50)
Fibrosis Grade	$1.69 \pm 1.12$ (95% CI 1.48 - 1.91)

**Table 2. Steatosis, Necro-inflammatory score and fibrosis grade of CHC patients (n=109).**

Steatosis	Degree	No.	Percent
	None	42	38.5
	Mild	35	32.1
	Moderate	1	0.9
	Severe	31	28.5
Necro-inflammatory score	Grade	No.	Percent
	1	25	22.7
	2	36	32.7
	3	36	32.7
	4	12	10.9
Fibrosis grade	Stage	No.	Percent
	0	5	4.5
	1	62	56.4
	2	16	14.5
	3	13	11.8
	4	13	11.8

Histological evaluation showed that 56 (46.4%) patients had lymphoid follicles. Steatosis was found in 67 (61.5%) patients. All biopsies showed necro-inflammatory changes and scores are shown in Table 2. Fibrosis grade from 1 to 4 were found in 95% of patients and 5 specimens (4.5%) had no fibrosis. None of the patients had bile duct injury.

Steatosis was found in 67 (61.5%) patients. Most of the cases had mild or severe degree of steatosis. Necro-inflammatory grade in most cases showed grade II and III and fibrosis stage patients generally belonged to grade I. These are shown in Table 2.

Correlation of steatosis with various liver function tests, and for age and sex was assessed using spearman correlation. Correlation coefficient was 0.38 (P=NS) for age, coefficient was 0.93 (P = NS) for ALT, coefficient was 0.057 (P = NS) for AST, coefficient was 0.22 (P = NS) for sex and correlation coefficient was 0.158 (P = NS) for total bilirubin. Spearman correlation coefficient for steatosis to fibrosis was 0.004 (P = NS).

## Discussion

Our patients showed the histological features of lymphoid aggregate, follicles and necro-inflammatory changes which were similar to most of the earlier reports. However, bile duct damage was not seen in these patients as has been reported in several other European studies.<sup>4,8</sup> Our patients showed various degrees of steatosis in 62% cases which has been found in other studies.<sup>9-11</sup> Hepatic steatosis is a common feature and has been linked with acceleration of fibrosis development specially in patients who have hepatitis C virus genotype 3.<sup>9,10</sup> It has been proposed that there could be oxidative damage which may result in steatosis<sup>9</sup> or certain genotypes hepatitis C virus may interfere with the pathways of hepatic lipid metabolism.<sup>11</sup> Patients with chronic hepatitis C and significant steatosis have been found to have progressive fibrosis on liver biopsies.<sup>13</sup> As our population in Pakistan has shown predominant serotype 3<sup>14</sup> and although, the treatment response to antiviral therapy in these patients has been excellent<sup>15,16</sup>, it has been recommended that patients with genotype 3 and steatosis should have early therapeutic intervention.<sup>10</sup>

Patients with steatosis are known to have increased liver echogenicity on ultrasound examination and a recent study showed that 60% of the patients with raised echogenicity had various degrees of liver steatosis.<sup>17</sup> However, liver biopsy remains the gold standard for the evaluation of steatosis. Since steatosis is assuming greater importance with fibrosis and accelerated progression of disease, quantitative assessment of fibrosis and steatosis using stereological techniques have been shown to yield better results for assessment of steatosis and fibrosis.<sup>18</sup>

There have been various factors that have been associated with steatosis which include alcohol intake, obesity, diabetes mellitus and high cholesterol levels, among others.<sup>11</sup> However, hepatitis C patients have been noted to have steatosis and acinar fibrosis<sup>19</sup> and obesity has

been noted along with hepatitis C in these patients.<sup>19,20</sup> There have been studies to show that weight reduction may be helpful in these patients.<sup>20</sup> Weight reduction had been shown to decrease the Knodell score of fibrosis and decrease in activated stellate cells on periodic biopsies<sup>21</sup> in these patients.

Although our study had shown that steatosis has been present in these patients but there are limitations of the study. We did not have the blood glucose levels, cholesterol levels or the weight of these patients which may have contributed to some degree to the steatosis. Nevertheless, the percentage of patients having steatosis in the study are just about as has been found in various other studies.<sup>22</sup>

In conclusion, this study has shown that over 60% of patients with hepatitis C had varying degrees of steatosis. As steatosis has been noted more in patients with genotype 3 and has been associated with more rapid progression of fibrosis, this should re-emphasize the need for early intervention and treatment of patients in our population who generally have genotype 3 and have associated steatosis.

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