Liver pathology in chronic hepatitis C virus infection associated with extrahepatic manifestations

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Abstract

We analyzed microscopic changes in the liver from ten patients infected by hepatitis C virus. All patients developed chronic hepatitis and extrahepatic manifestations as cryoglobulinemia, polyneuropathy, purpura cutanea, Sjögren syndrome and nephritic syndrome. The most severe microscopical changes in the liver were connected with cryoglobulinemia and cutaneous manifestations together with Sjögren syndrome and represented intensive inflammatory lymphoid infiltrates in the lobules, portal spaces with prominent interface hepatitis and septal fibrosis with architectural distortion.

Key words: HCV, extrahepatic manifestations, chronic hepatitis, vasculitis

Introduction

Hepatitis C virus (HCV) infection is the second most common viral infection of the liver with a global prevalence of 3% (about 180 million people, but may are unaware of the infection) leading to chronic hepatitis, cirrhosis and hepatocellular carcinoma. Because of its hepatotropic, lymphotropic features and replication in hepatocytes, lymphocytes and macrophages, HCV can induce systemic disease with extrahepatic manifestations [4]. The prevalence of extrahepatic manifestations is low in pediatric population, but about 50% of adult HCV-positive patients during the course of the disease develop at least one extrahepatic manifestation [7]. Among the best reported are cryoglobulinemia, peripheral neuropathy, glomerulonephritis, thrombocytopenic purpura, lichen planus, corneal ulcer, Sjögren syndrome, porphyria cutanea tarda and necrotizing cutaneous vasculitis. [5, 8].

Patients, material and methods

Total number of 10 patients (8 females, 2 males) aged 31 to 69 years (mean age 51,6) with chronic hepatitis C virus (HCV) infection confirmed by serological and virological (HCV-RNA) criteria, underwent percutaneous liver biopsy (Tab. 1). Extrahepatic manifestations of the HCV infection occurred in all patients: cryoglobulinemia in 8 patients, purpura cutanea in 5 patients, Sjögren syndrome in 2 patients, polyneuropathy in 3 cases, nephrotic syndrome in 3 cases. The majority of patients (9 cases) presented more than one feature of extrahepatic manifestations, the rest (1 patient) had

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membranous nephropathy. Patients with alcohol abuse, diabetes and metabolic syndrome were excluded from the study.

All biopsy specimens were fixed in buffered formalin and embedded in paraffin blocks. Four micrometer thick sections, displaying at least 10 portal spaces were routinely stained by Hematoxylin and Eosin, Periodic Acid-Schiff method with and without diastase, Gomori Silver impregnation and Azan or Masson Trichrome Stain. Histological interpretation was performed using internationally accepted criteria [1] and reviewed retrospectively (two independent pathologists). All histological features were finally scored using the four degree scale for the grade (inflammatory activity) and stage (fibrosis) of the disease. The grade was assessed as: 0 – portal inflammation only, without piecemeal necrosis, without lobular inflammation and necrosis; 1 – minimal portal inflammation, minimal, patchy piecemeal necrosis, occasional spotty necrosis; 2 – mild portal inflammation, mild piecemeal necrosis involving some or all portal tracts, little hepatocellular damage; 3 – moderate portal inflammation, moderate piecemeal necrosis involving all portal tracts, moderate lobular inflammation with noticeable hepatocellular change; 4 – severe portal inflammation severe piecemeal necrosis, may have bridging necrosis, severe lobular inflammation with prominent diffuse hepatocellular damage. The stage was assessed as: 0 – no fibrosis, normal connective tissue; 1 – portal fibrosis, fibrous portal expansion; 2 – perportal fibrosis, perportal or rare portal-portal septa; 3 – septal fibrosis, fibrous septa with architectural distortion; 4 – cirrhosis.

Results

The necroinflammatory activity, described as a grade of the disease, was severe in 20%, moderate in 30% and mild in 50% of the examined material. We found lymph follicle formations in portal tracts and inflammatory infiltrates near blood vessel walls (venulitis) in all patients. Hepatocyte steatosis was observed in 4 patients. Liver fibrosis, described as a stage of the disease was characterized by numerous fibrous portal septa and architectural distortion in 20%, while portal fibrosis and portal-portal septa occurred in 60% of biopsy specimens and mild or no fibrosis in 20% of them. Lymph follicle formations were present in all biopsy specimens. They were found in the portal tracts (at least 3 per 10 portal tracts), always involved blood vessels. Lymphoid cells infiltrated also the walls of venous vessels in all portal tracts (Fig. 1).

Histological activity index varied from mild inflammation to severe necroinflammatory activity with the mean activity of grade 3 (moderate inflammation). The stage of the

<table>
<thead>
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<th>No</th>
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<td>Portal lymph follicles portal vasculitis, hepatocyte steatosis</td>
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</table>

Table 1

Characteristics of HCV positive patients with extrahepatic manifestations
disease varied also from 0 (without fibrosis) to 3 (architectural distortion) with the mean stage of 2 (portal-portal fibrosis without cirrhosis). The most severe microscopic changes in the liver were connected with cryoglobulinemia and cutaneous manifestations together with Sjögren syndrome and represented intensive inflammatory lymphoid infiltrates in the lobules, portal spaces with prominent interface hepatitis and septal fibrosis with architectural distortion.

**Discussion**

Extrahepatic manifestations in HCV infected patients involve primarily the skin and joints. The most frequent immunologic abnormalities include mixed cryoglobulinemia, renal and thyroid diseases and lymphoproliferative disorders [3]. We present patients with chronic hepatitis connected with cryoglobulinemia, Sjögren Syndrome, skin, joints renal and neurological lesions.

Vascular changes and lymph follicle formations occurred in all liver biopsies. This phenomena have been also observed in other patients of HCV infection without extra-hepatic involvement, but the intensity of this changes appear to be more pronounced than in other chronic hepatitis cases. Similar morphological vascular changes have been observed in transplanted liver with acute rejection as autoimmune phenomena. Vascular changes in these patients can be considered as immunological reaction to generalized HCV infection.

The most severe changes in the liver were found in two patients with Sjogren syndrome, cryoglobulinemia and cutaneous disorders. Despite these findings we did not found any clear correlation between presence of these symptoms and staging score of chronic hepatitis, because these lesions occurred also in a patient with mild changes in the liver. The prospective study of 45 consecutive HCV infected patients revealed similar histopathological changes in the liver, but also the association between older age and liver disease activity [6].

The association between the presence of serum cryoglobulins and intensity of hepatic microscopic changes was found, with no difference in patients age or duration of infection [12]. It is possible that cryoglobulinemia results in

![Fig. 1 Microscopical changes in the liver in patients with extrahepatic manifestations of HCV infection](image-url)

A: Lymphoid infiltration in the wall of a vein in the portal tract, the ducts remain unchanged (H.E. 400x)
B: Inflammatory infiltrates of the portal tract with interface hepatitis, involvement of blood vessels, the ducts remain unchanged (H.E. 400x)
C: Lymph follicle near blood vessels in the portal tract (Azan stain 200x)
D: Inflammatory infiltrate and fibrosis in the portal tract (Azan stain 200x)
more rapid hepatic fibrosis in HCV infected patients. In our
group of ten patients we did not found this association.

Renal diseases associated with HCV infection are rela-
tively common in adult patients but rare in children. We
found them in five patients together with other extrahepatic
manifestations, but in two cases as single manifestation.
Interestingly they occur in the youngest patients from our whole
group (female 31 and male 32 years old). The hepatic histo-
logical activity of these patients is moderate, without pro-
gression into cirrhosis. The frequent lesions associated with
HCV are membranoproliferative glomerulonephritis, mem-
branous nephropathy, focal segmental glomerulosclerosis and
mesangial proliferative glomerulonephritis with IgA [10]. In our study HCV-related glomerulopathy consisted of
membranoproliferative glomerulonephritis in 2 patients and
membranous nephropathy in 1 patient.

Neurologic complications in HCV infected patients
can involve the peripheral and central nervous system, the
most frequently subacute, distal, symmetric, sensory and mo-
tor polyneuropathy in the presence or without mixed cry-
globulinemia [10]. Polyneuropathy increased significantly with age, which is the only independent predictor. Cryoglo-
bulinemia and the intensity of fibroinflammatory changes in
the liver do not influence the prevalence of polyneuropathy
[11]. In our study polyneuropathy occurred in 4 patients;
three of them were more than 60 years old (mean age 65) and
the histological activity was mild to moderate. The fourth pa-
tient was younger (51 years old) but he had also other extra-
hepatic manifestations as Sjögren syndrome, nephritic syn-
drome and skin lesions. Interestingly, the histological activi-
ty was not very high but the vascular involvement was
prominent (Fig. 1A).

Systemic autoimmune diseases are also connected
with HCV infection. In a large study of 1020 patients from
Europe, America, Asia and Australia (HISPAMEC Registry)
systemic autoimmune symptoms in HCV infection are most
commonly described in Sjögren syndrome, rheumatoid ar-
tritis and SLE [9].

In summary, infections with HCV can involve not only
the liver but also various organs. The histological activity
of hepatitis does not influence the incidence of extrahepatic
involvement. Vascular involvement and prominent lymph
follicles in portal tracts are constant findings in patients with
HCV chronic hepatitis and extrahepatic manifestations.

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