Morphological characteristics of chronic hepatitis : A comparative study on Turkish patients

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Abstract

Background : **Chronic hepatitis caused by hepatitis B and hepatitis C viruses have characteristic histological features. We aimed to compare these histological features between two groups.**

Methods : **We worked on two groups each contains 50 patients, that are serologically proven, have hepatitis B and hepatitis C. These patients were analysed according to the histological features which are more often seen in chronic hepatitis C.**

Results : **We found higher percentage of lymphoid follicles and aggregates in portal tracts, bile duct damage or loss and lymphoreticular reaction in patients with chronic hepatitis C than chronic hepatitis B (44% - 12%, 96% - 14%, 84% - 40%, respectively). We recorded most patients with severe portal tract inflammation were in group of chronic hepatitis C (86%). Bile duct loss wasn't observed in any patients with chronic hepatitis B , while it was present in 44% of chronic hepatitis C group. There was no significant difference between two hepatitis groups with regard to fatty change. 13 out of 14 patients, having all those above mentioned histological findings, diagnosed with chronic hepatitis C whereas only one patient had chronic hepatitis B.**

Conclusions : **Aggregates and follicles forming portal inflammation, lymphoreticular reaction together with bile duct damage, especially with bile duct loss strongly indicates presence of chronic hepatitis C.** (Acta gastroenterol. belg., **2002**, 65, **146-149**).

Key words : hepatitis C infection, hepatitis B infection, viral hepatitis, pathology.

Introduction

Currently, hepatitis B and hepatitis C viruses (HBV, HCV) are the etiologic agent of the most of the chronic hepatitis (1). It is known that virus caused chronic hepatitis patients have higher risk for development of cirrhosis and hepatocellular carcinoma. Such risk, which differs depending on the etiological agent, is higher in hepatitis C than hepatitis B (2, 3). For this reason, etiological agent should be exactly identified to determine the prognosis of chronic hepatitis. Serological and molecular biological methods such as ELISA and PCR are used to identify the agent (2, 4, 5). Moreover, easily applied thin needle aspiration biopsy, is frequently used to determine existence and severity of hepatic destruction and the agent. The number of studies have been conducted to search the histological characteristics of chronic hepatitis etiology. Some morphological characteristics differ depend on the agent of chronic hepatitis (6, 7). For example, cytoplasm in the form of ground-glass, is observed depending on the endoplasmic reticulum proliferation including hepatitis B surface antigen (HBsAg) in the cytoplasm of hepatocytes infected by HBV, is considered as a histological indication of HBsAg (8, 9). In chronic hepatitis C (CHC) patients ; biliary duct (BD) damage or loss, extensive lymphoid infiltration that causes in aggregate or lymphoid follicle composition in portal areas (PA), fatty change and inflammatory infiltration in the form of intraacinary lymphoreticular reaction are often observed (8, 9, 10). These morphological characteristics that differ depending on viruses are diagnostic but not patognomonic (11).

In this study, the frequency of observing the morphological characteristics, and its specificity to hepatitis C were evaluated in the cases that are known to have only hepatitis B and hepatitis C serologically. In addition we also tried to show whether there are any differences between morphological characteristics of Turkish and foreign chronic hepatitis patients previously reported.

Materials and methods

The diagnostic liver biopsies of 50 CHC and 50 chronic hepatitis B (CHB) cases which were admitted to the Pathology Department of Ankara University Medical School between 1998-1999 were evaluated retrospectively. 50 cases, that were seropositive for HBsAg, 'e' antigen (HBeAg) or antibody-positive to HBeAg (anti-HBe) by ELISA test (Abbott Laboratories, North Chicago, USA) at least for 6 months and HBV DNA-positive by PCR and who had high value of alanine aminotransferases (ALT) that was at least 1.5 times the upper limit was considered as CHB and included in the study. CHC group, on the other hand, was composed of 50 cases which were established as being antibody-positive to the HCV (anti-HCV) by ELISA, HCV RNA-positive by PCR ; and having a similar high value of ALT during at least 1-year monitoring. In both groups, other reasons for hepatitis and steatosis were eliminated through clinical, serological, biochemical and immunological criteria. 20 cases with CHB were female, 30 cases were male and the mean of age was 30 (2-58). 28 cases with CHC were female, 22 cases were male and the mean of age was 48 (20-70).

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All biopsy samples were fixated at 10% formaldehid solution. Sections with a thickness of 3-4 μm were stained with hematoxylin and eosin and masson trichrom and prussian blue. All preparations were examined by the same pathologist who did not know clinical, biochemical and serological data of patients. All of the cases were researched regarding PA inflammation, BD damage or loss, fatty change, existence of inflammatory cell infiltration in the form of lymphoreticular reaction which were known as diagnostic in HCV hepatitis. According to this, PA inflammation was assessed as 0, if there was no inflammation ; as 1, if it was only in the form of infiltration ; as 2, if it was in the form to cause aggregate ; and as 3, if it was in the form to cause follicle. BD was assessed as 0, if there was no BD damage ; as 1, if damage was observed ; and as 2, if a loss was observed. Regarding the existence and severity, fatty change was assessed as 0, if no fatty change was observed ; as 1, if it was observed in less than 30% of the hepatocytes ; as 2, if it was observed in 30% and 60% of the hepatocytes ; and as 3, if it was observed in more than 60% of the hepatocytes. Lymphoreticular reaction was subjectively examined in two groups, existing and non-existing. Furthermore, all cases were graded according to Knodell's histologic activity index (HAI) score (12).

The degree of relation between fibrosis and portal inflammation, portal aggregate-follicle formation, periportal-bridging necrosis was calculated by Spearman's Correlation Coefficient. The other results were performed using standard chi-square test.

Results

When hepatitis B and C was compared regarding to the inflammation in PA's, the existence of inflammation in PA's in the form of lymphoid aggregate or follicle is more significant in CHC than CHB (p = 0.0004, Table 1).

The severity of PA inflammation was evaluated according to Knodell's HAI score. According to this, severe PA inflammation was observed in 24% of the cases with CHC and only in 4% of the cases with CHB. It was established that this difference was statistically significant (p = 0.0016, Table 2).

The existence and severity of periportal and bridging necrosis according to types of hepatitis was evaluated, and bridging necrosis was observed in 12% of patients with CHB and 8% of patients with CHC. This difference was found as statistically insignificant (p = 0.37, Table 3).

The relation between the severity of PA inflammation and piece-meal and bridging necrosis was evaluated in cases with CHB (Table 4). According to this, weak portal inflammatory infiltration was observed in 80% of patients who do not have piece-meal necrosis. Whereas moderate or severe portal infiltration was observed in 83% of patients with bridging necrosis. These results were found to be statistically insignificant (p = 0.11). When the relation was evaluated in patients with CHC ; bridging necrosis was observed in 4 of CHC patients and it was also observed that all of these patients had moderate or severe PA inflammation. In this group, 39 patients had moderate or severe PA inflammation, but minimal piece-meal necrosis ; thus, a statistically significant relation was not found (p = 0.38, Table 5).

As table 6 indicates, when BD damage or loss according to hepatitis virus types was evaluated, BD damage or loss was observed in 96% of patients with CHC. On the other hand, BD damage was observed only in 14% of the patients with CHB ; however, BD loss was not observed in any patient. It was also established that this difference was statistically significant (p = 0.00001).

Evaluation on the existence of fatty change according to virus types revealed that 28/50 (56%) of patients with CHB and 35/50 (70%) of patients with CHC had fatty

Table 1. — **PA Inflammation in CHC and CHB**

Hepatitis type	Lymphocytic infiltration n	%	Aggregate / follicle n	%
B	44	88	6	12
C	28	56	22	44

Table 2. — **Severity of PA Inflammation in CHC and CHB**

Hepatitis type	PA Inflammation					
	Mild		Moderate		Severe	
	n	%	n	%	n	%
B	25	50	23	46	2	4
C	11	22	27	54	12	24

Table 3. — **Periportal and Bridging Necrosis in CHC and CHB**

Hepatitis type	Negative		1*		2**	
	n	%	n	%	n	%
B	5	10	39	78	6	12
C	2	4	44	88	4	8

* All of patients whose necrosis degree : 1-3-4 (bridging necrosis-negative).
** All of patients whose necrosis degree : 5-6-10 (bridging necrosis-positive).

Table 4. — **The Relation between Periportal and Bridging Necrosis and PA Inflammation in CHB**

Periportal and bridging necrosis	Severity of PA inflammation			
	Mild		Moderate / severe	
	n	%	n	%
0	4	80	1	20
1*	20	51	19	49
2**	1	17	5	83

* All of patients whose necrosis degree : 1-3-4 (bridging necrosis-negative).
** All of patients whose necrosis degree : 5-6-10 (bridging necrosis-positive).

Table 5. — **The Relation between Periportal and Bridging Necrosis and PA Inflammation in CHC**

Periportal and bridging necrosis	Severity of PA inflammation			
	Mild		Moderate / severe	
	n	%	n	%
0	0		2	100
1*	11	25	33	75
2**	0		4	100

* All of patients whose necrosis degree : 1-3-4 (bridging necrosis-negative).

** All of patients whose necrosis degree : 5-6-10 (bridging necrosis-positive).

Table 6. — **BD Damage or Loss in CHC and CHB**

Hepatitis type	No damage		BD damage		BD loss	
	n	%	n	%	n	%
B	43	86	7	14	0	
C	2	4	26	52	22	44

Table 7. — **Lymphoreticular Reaction in CHC and CHB**

Hepatitis type	Negative		Positive	
	n	%	n	%
B	30	60	20	40
C	8	16	42	84

change. Fatty change was a little bit more in CHC patients ; however, this difference was not statistically significant ($p = 0.15$).

Evaluation on the relation of the existence of lymphoreticular reaction with types of viral hepatitis revealed that such reaction was observed in both hepatitis groups. Such reaction was observed in 40% of cases with CHB, and 84% of patients with CHC. It was established that this relation was statistically significant ($p = 0.00001$, Table 7).

When the relation between the severity of PA inflammation and aggregate and follicle formation was evaluated in patients with CHB, a weak statistically significant relation was found ($r = 0.334$, $p < 0.05$). However, a statistically significant relation was found in patients with CHC ($r = 0.727$, $p < 0.001$).

The presence and the severity of fibrosis was examined in patients with CHB and CHC. But there was no difference between these two groups ($p > 0.05$).

The relation between the severity of PA inflammation and fibrosis in both CHC and CHB patients was researched. A statistically weak significant relation was found in CHB ($r = 0.317$, $p < 0.05$) but no significant correlation was found for CHC ($r = 0.238$, $p > 0.05$).

When the relation between the severity of periportal and bridging necrosis and fibrosis according to types of hepatitis was researched, a statistically significant relation was found both in CHB and CHC patients. (for CHB $r = 0.552$, $p < 0.001$, for CHC $r = 0.450$, $p < 0.001$).

The distribution of cases, where PA inflammation, BD damage or loss, fatty change and lymphoreticular reaction were observed together, were evaluated according to the viral hepatitis type. It was observed that 13 of 14 (93%) of such patients were in CHC group and 1 patient (7%) was in CHB group ($p = 0.0005$).

Only 2 of 50 patients with CHC showed iron deposition in liver biopsies. And in these two patients, iron deposition were detected in a few Kupffer cells.

Discussion

Etiology of chronic hepatitis is the most important parameter, which leads the treatment and prognosis. Various studies revealed morphological characteristics, which are specific to CHC and CHB (8,9,10). Such morphological indicators with clinical and other laboratory data assist to etiological diagnosis. Sometimes in other chronic liver disease, like autoimmune chronic hepatitis and alcoholic liver diseases, HCV coinfection may be take place. In such cases, HCV-specific morphological findings may be valuable for diagnosing of CHC (10,13). Furthermore, in HBV and HCV coinfection, that is observed generally in frequent transfusion required patients, morphological indicators may be used to differentiate the factor, on which the chronic hepatitis diagnosis is histologically dependent.

In our study, we showed that PA inflammation in the form of aggregate or follicle was one of the morphological characteristics findings of CHC, but not for CHB, and these results were strongly correlated with previous reports (14,15,16). Moreover degree of PA inflammation in CHC group was also strongly correlated with aggregate or follicle infiltration. On the other hand lymphoreticular reaction was the other morphological characteristics in our CHC group as previously reported (17,18). Whereas BD loss was not showed in any of CHB patients, it was existed in 44% of CHC group. BD damage or loss was observed in 96% of CHC patients, but only 14% of CHB patients had BD damage alone. In two studies, Lefkowitch and Wong *et al*. have indicated that BD damage was between 31-30% (9,16). Moreover Bach *et al*. showed 91% BD loss in CHC patients and this is higher than ours (6). According to this, we have the opinion that in cases, in which BD damage and/or loss is clearly observed, HCV is more probable etiologic factor than the other viral hepatitis agents. It is reported that, fatty change in hepatocytes in CHC, is frequently detected (10,19). However, we did not find any difference among two groups. In two other studies, fatty change in CHB patients is as high as ours (69%-51% and 70%-67% in CHC and CHB patients, respectively) (9,20).

PA inflammation, BD damage and/or loss, fatty change and lymphoreticular reaction was observed together in our 14 patients. It is considerable that 13 of 14 patients were in CHC group. The results we obtained are similar to the studies, defining such morphological

characteristics as common histological indicators of CHC patients (9,10).

The morphological features of CHC in Turkish patients have been previously evaluated by Akpolat *et al*. (21). They have evaluated in 44 biopsy specimens from CHC patients. But we could not find any comparative study on the morphological changes between CHB and CHC from Turkey on MEDLINE database. In their study, lymphoid follicles and aggregates in PA's, lymphoreticular reaction, BD loss and steatosis were 32%, 80%, 25% and 80%, respectively. Whereas BD loss is lower than our results, other ratios are similar. This difference may be related to the patient group or geographic differences. For explaining this difference comparative studies including these parameters must be examined on Turkish patients.

In both hepatitis groups, we observed that the degree of piece-meal and bridging necrosis correlated with the degree of fibrosis. This correlation has been shown previously for CHC (16). There are studies indicating that PA inflammation is more and piece-meal necrosis is less severe in CHC than in CHB (17,22). Similarly severe inflammatory infiltration was observed in CHC patients. However, contrary to other reports there were no difference for piece-meal necrosis between patients CHC and CHB groups. There were no difference for fibrosis among two groups. However, it has been reported that fibrosis was more severe in CHB than CHC (17). The degree of inflammation in our CHB patients was parallel with fibrosis but there were no correlation in CHC group. In contrast to our result this correlation has been shown in CHC group previously (16). Difference in these three histopathological comparisons may depend on different patient population or interpretation of histopathological features.

In conclusion, PA inflammation in aggregate or follicle form, BD damage and lymphoreticular reaction together should consider the existence of CHC. Furthermore, BD loss may be the most characteristic finding of CHC. To detail fatty change in CHB and BD loss in CHC we need further studies extended to CHC genotyping, increased number of patients and differences between geographic distributions.

References

1. SHERLOCK S., DOOLEY J. Diseases of the Liver and Biliary System. Oxford : Blackwell Scientific, 1997, 303-35.
2. SALLIE R., BISCEGLIE A.M. Viral hepatitis and hepatocellular carcinoma. *Gastroenterol. Clin. North. Am.*, 1994, **23** : 567-9.
3. OKUDA K. Clinicopathological features of virus associated hepatocellular carcinoma : B vs C. VIII. International Symposium on Viral Hepatitis Jan 22-24, 1998 Madrid, Spain, 117-21.
4. SHAW-STIFFEL T.A. Chronic hepatitis. In : Mandell G.L., Bennett J.E., Dolin R., (eds). Principles and Practice of Infectious Diseases. Philadelphia : Churchill Livingstone, 2000, 1297-331.
5. TREPO C., ZOULIM F., ALONSO C., PETIT M.A., PICHOUD C., VITVITSKI L. Diagnostic markers of viral hepatitis B and C. *Gut*, 1993, Suppl. S20-S5.
6. BACH N., THUNG S.N., SCHAFFNER F. The histological features of chronic hepatitis C and autoimmune chronic hepatitis : A comparative analysis. *Hepatology*, 1992, 15 : 572-7.
7. Lee R.G. Diagnostic Liver Pathology. London : Mosby, 1994 : 57-79.
8. BARWICK K.W., ROSAI J. Chronic hepatitis. In : ROSAI J. (ed). Ackerman' s Surgical Pathology. New York : Mosby, 1996, 861-7.
9. LEFKOWITCH J.H., SCHIFF E.R., DAVIS G.L., PERRILLO R.P., LINDSAY K., BODENHEIMER H.C. *et al.* Pathological diagnosis of chronic hepatitis C : A multicenter comparative study with chronic hepatitis B. *Gastroenterology*, 1993, **104** : 595-603.
10. MORIYA K., YOTSUYANAGI H., SHINTANI Y., FUJIE H., ISHIBASHI K., MATSUURA Y. *et al.* Hepatitis C virus core protein induces hepatic steatosis in transgenic mice. *J. Gen. Virol*., Jul 1997, **78** (Pt 7) : 1527-31.
11. GERBER M.A., KRAWCZYNSKI K., ALTER M.J., SAMPLINER R.E., MARGOLIS H.S. Histopathology of community acquired chronic hepatitis. *Mod. Pathol*., 1992, **5** : 483-6.
12. KNODELL R.G., ISHAK K.G., BLACK W.C., CHEN T.S., CRAIG R., KAPLOWITZ N. *et al*. Formulation and application of a numerical scoring system for assesing histological activity in asymptomatic chronic active hepatitis. *Hepatology*, 1981, **1** : 431-5.
13. PERRILLO R.P. The role of liver biopsy in hepatitis C. *Hepatology*, 1997, **26** (Suppl. 1) : S57-S61.
14. COLOMBARI R., DHILLON A.P., PIAZZOLA E., TOMEZZOLI A.A., ANGELINI G.P., CAPRA F. *et al.* Chronic hepatitis in multiple virus infection : histopathological evaluation. *Histopathology*, 1993, **22** : 319-25.
15. SCHEUER P.J., ASHRAFZADEH P., SHERLOCK S., BROWN D., DUSHEIKO G.M. The pathology of hepatitis C. *Hepatology*, 1992, **15** : 567-71.
16. WONG V.S., WIGHT D.G., PALMER C.R., ALEXANDER G.J. Fibrosis and other histological features in chronic hepatitis C virus infection : a statistical model. *J. Clin. Pathol*., 1996, **49** : 465-9.
17. HAHM K.B., CHON C.Y., KIM W.H., HAN K.H., CHUNG J.B., LEE S.I. *et al.* Histologic study of chronic active hepatitis C ; comparison with chronic active hepatitis B. *Korean J. Intern. Med*., 1992, **7** : 102-10.
18. SHAH H.A., KAYANI N., SHEIKH H., JAFRI S.W., HAMID S., KHAN A.H. Comparison of liver histology in chronic active hepatitis C and chronic active hepatitis B. *Indian J. Gastroenterol*., 1995, **14** (3) : 91-4.
19. MIHM S., FAYYAZI A., HARTTMAN H., RAMADORI G. Analysis of histopathological manifestations of chronic hepatitis C virus infection with respect to virus genotyping. *Hepatology*, 1997, **25** : 735-9.
20. MALHOTRA V., SAKHUJA P., GONDAL R., SARIN S.K., SIDDHU M., DUTT N. Histological comparison of chronic hepatitis B and C in an Indian population. *Trop Gastroenterol*., 2000, **21** (1) : 20-1.
21. AKPOLAT I., ÖZYILKAN E., KARAGÖZ F., KANDEMYR B. Histopathological characteristics of chronic hepatitis C virus infection in Turkey. *Acta Gastro-Enterologica Belgica*, 1997, **60** (4) : 251-4.
22. GIUSTI G., PASQUALE G., GALATE D., RUSSO M., SARDARO C., GALLO C. *et al*. Clinical and histological aspects of chronic HCV infection and cirrhosis. *Hepatogastroenterology*, 1993, **40** : 365-9.