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Operational scores in the diagnosis of chronic hepatitis. A semi-quantitative assessment

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Abstract

Starting from the quantification of the specific lesions for chronic hepatitis B and C, our study focused on (*i*) the correspondence between the necroinflammatory activity and the fibrosis stage ascertained through the Ishak scoring system, (*ii*) the classification overlaps and differences of Ishak *vs.* METAVIR score. The study group consisted of 202 cases with chronic hepatitis B and 751 cases with chronic hepatitis C, diagnosed based on liver biopsies. The fragments of hepatic tissue were routinely processed and stained with Hematoxylin–Eosin, trichrome Szekely, Gordon–Sweet silver impregnation, and Periodic Acid–Schiff. A semiquantitative evaluation was performed using the Ishak (for hepatitis B and C) and the METAVIR (for hepatitis C) scoring systems. Our results revealed that the comparison between hepatitis B and C, based on the necroinflammatory activity and fibrosis, is able to offer through the numeric values of the Ishak scoring system accurate proofs, which support the aggressivity of hepatitis C, because it develops fibrosis more quickly, even on the background of mild necroinflammatory activity. Also, our data showed that the necroinflammatory activity and the fibrosis are not processes which progress in a consistent pattern. The application of the METAVIR scoring system for the cases with chronic hepatitis C confirmed that there is not a direct correlation between necroinflammation and fibrosis. The Ishak scoring system provides through the wide range of numeric values attributed for the evaluation of necroinflammatory activity and fibrosis far more precise criteria for the appraisal of the degree of damage to the hepatic parenchyma at the time of the diagnosis. Supplementary, the METAVIR scoring system allows for the hepatitis C an assessment of the entire histologic activity, including the interface hepatitis and the associated lobular necrosis components. The scoring systems have unavoidably strengths and weaknesses, but the choice of a specific one must reflect the consensus between the pathologists and the clinicians, relying on their experience.

Keywords: chronic hepatitis, liver biopsy, semiquantitative assessment, scoring systems.

Introduction

The infection with hepatotrope viruses, namely hepatitis B virus and hepatitis C virus afflicts 3 up to 5% of the world population [1, 2] representing the ninth most frequently occurring cause of death.

The concept of chronic viral hepatitis appeared during the Second World War, when liver chronic diseases are described at soldiers with jaundice episodes [3].

During the 50's, the terms of chronic persistent hepatitis (corresponding to a non-progressive variation of the viral hepatitis, with prolonged evolution) and chronic active hepatitis [4, 5] are introduced.

Although this new terminology can be found in the monograph "The Chronic Hepatitis" edited in 1966, the first classification authored by an international group of hepatologists, which tries to correlate the histo-pathologic aspects of the liver biopsy with the prognosis of the disease, is published in 1968 [6].

The histopathologic criterion, which differentiated the two forms, is the periportal necrosis occurring in fragments, called "piecemeal necrosis". Soon, it became clear that the parenchymal lesions, especially the "bridging necrosis" constitute an important predictive factor in the progression of the disease (the evolution towards cirrhosis), and thus the chronic lobular hepatitis was defined [7].

Later, the pathologic aspect was broadened, taking into account several other options, such as nonspecific reactive hepatitis – a less aggressive form than chronic persistent and chronic lobular hepatitis, and chronic septal hepatitis – a form of chronic active hepatitis with remission features.

After more than 30 years from the initial classification, important knowledge regarding the etiology, the pathogenesis, the clinical profile and the pathology of the chronic hepatitis bring about new aspects, the old terminology becoming a confusion source, by using the

same diagnosis, such as chronic active hepatitis, for diseases with different origins (viral, autoimmune, metabolic) [8].

These reasons imposed the formulation, by the International Liver Research Association, of a new classification based on etiology, because of the substantial differences between the clinical aspects, the prognosis and the treatment of the different types of chronic hepatitis [9].

In the pathologic exam of liver biopsy, the implicit subjective component was clearly diminished by the introduction and development of certain scoring systems: Scheuer [10], Ludwig [8], Knodell [11], Ishak [12], and METAVIR [13].

The scoring systems have unavoidably strengths and weaknesses in their main objective – the semiquantitative classification of the necroinflammatory activity (NIA) and the staging of fibrosis.

In time, the research groups have published relevant results supporting or contesting the diagnostic and prognostic importance of these scoring systems. Nevertheless, despite all attempts to find and, consequently, introduce a scoring system with gold standard value, this was not possible.

We must stress the fact that each newly proposed scoring system was more and more complete, offering – through the application of several evaluation criteria – the possibility of an evaluation as accurate as possible. However, in the hepatic pathology centers, the decision of choosing a certain scoring system remains in the hands of the pathology team, by consensus with the clinicians, based on their practical experience.

Our research is based on the morphologic image resulted from the semiquantitative evaluation of a significant group of liver biopsies performed in chronic hepatitis B and C.

Starting from the quantification of the specific lesions, we focused on (*i*) the correspondence between the NIA and the fibrosis stage ascertained through the Ishak scoring system, (*ii*) the classification overlaps and differences of Ishak *vs.* METAVIR score.

At the same time, our analysis created the necessary support for the critical interpretation of the different classification systems, applicable in the assessment of the chronic hepatitis.

Materials and Methods

The study group consisted of 953 cases of chronic viral hepatitis (202 cases with chronic hepatitis B and 751 cases with chronic hepatitis C), diagnosed in the Pathology Laboratory of the "Sf. Parascheva" Clinical Hospital of Infectious Diseases, Iassy, between 2008 and 2010.

The liver biopsy was performed on all the patients included in the study group through the percutaneous method, using the special Hepafix® kit (B. Braun Melsungen AG, Germany), after echographic guidance.

The fragments of hepatic tissue (dimensions: 2.5/1.5/1 cm) were fixed in formalin and then processed according to the standard protocol of the Pathology Laboratory.

The histology specimens were than stained with Hematoxylin–Eosin (HE), trichrome Szekely, Gordon–Sweet silver impregnation, and Periodic Acid–Schiff (PAS).

The liver biopsies met the minimal criterion necessary for the evaluation – namely a minimum of three portobiliary spaces. The study group had, for each specimen, between 3 and 12 portobiliary spaces, with an average of 6–8.

The evaluation of the lesions was performed in a semiquantitative manner.

For the biopsies of the patients with chronic hepatitis B and C we applied the Ishak score which quantified individually the NIA (score 1–18) and the fibrosis (score 1–6) [12].

At the same time, the biopsies of the patients with chronic hepatitis C were evaluated also through the application of the METAVIR scoring system [13].

Results

The pathologic picture of the investigated liver biopsies was characterized by the presence of the specific basic lesions: inflammatory infiltrate, piecemeal necrosis, bridging necrosis and fibrosis, together with associated lesions developed in the hepatic parenchyma.

In our group, the morphology of the chronic hepatitis ranged from mild to severe forms, some with a tendency towards cirrhosis.

In the mild forms, the inflammatory infiltrate (formed by lymphocytes, macrophages, few plasma cells and rare neutrophils and eosinophils) was restricted to the portobiliary space, the hepatic architecture being preserved.

Gradually, the severity was signaled through: (*i*) the development of piecemeal necrosis, where the chronic inflammatory infiltrate goes beyond the location in the portobiliary space and invades the adjacent parenchyma, accompanied by necrotic hepatocytes from the limiting plates, and the presence of a lobular inflammation with focal hepatocytic necrosis; (*ii*) the development of bridging necrosis (porto-portal, porto-central, and central-central).

We noticed frequent aspects typical for the hepatocyte regeneration, such as binucleated young cells with basophil cytoplasm, rosette-like dispositions of the hepatocytes mimicking the glandular pattern, or local segmental thickenings of the hepatocyte plaques.

Various degrees of Kupffer cells hyperplasia was observed in both types of chronic hepatitis – B and C.

Hepatocytic steatosis was a common morphologic element, more often macrovesicular but also micro-vesicular, with an unsystematic topography most of the times.

The following images illustrate the significant morphologic lesions (Figures 1–10).

The described lesions were interpreted and quantified after the specific algorithms of the used scoring systems.

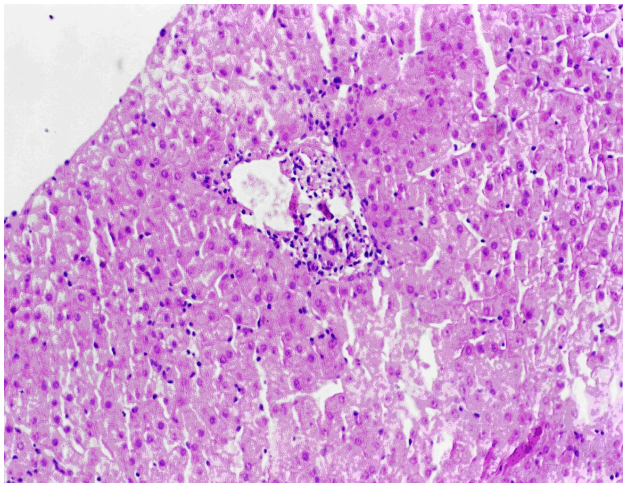

Figure 1 – ***Mild chronic hepatitis: a portobiliary space surrounded by hepatic parenchyma (HE stain, ob. ×10).***

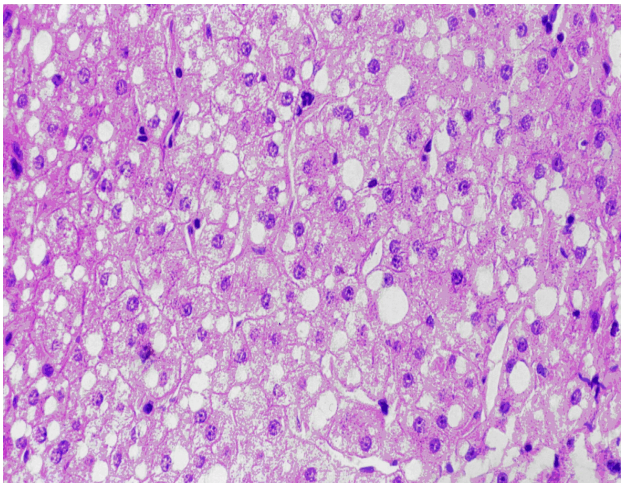

Figure 2 – ***Mild chronic hepatitis: macro- and micro-vesicular steatosis (HE stain, ob. ×20).***

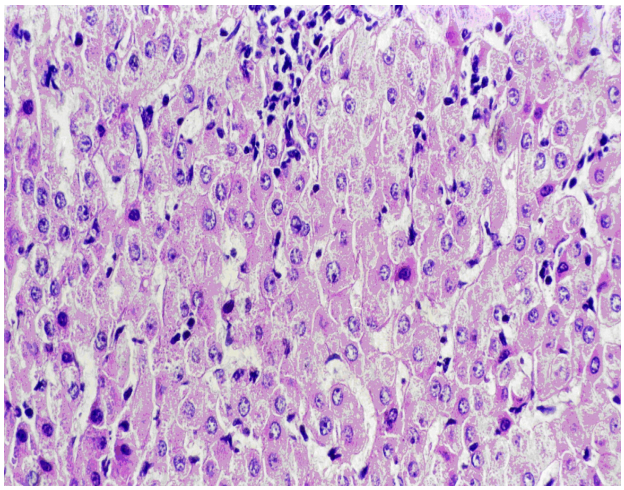

Figure 3 – ***Moderate chronic hepatitis: Councilman bodies (HE stain, ob. ×20).***

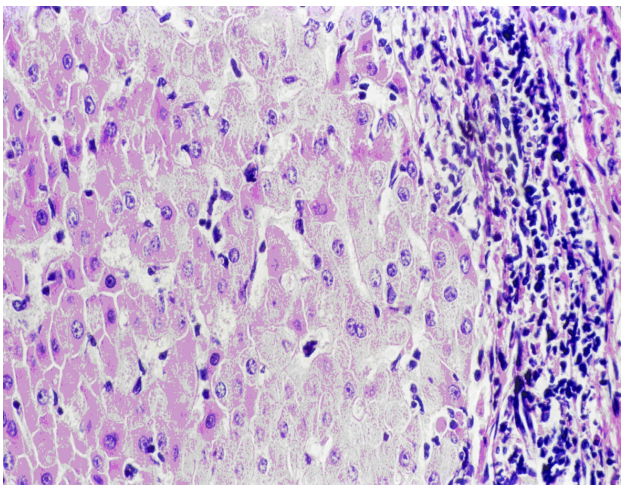

Figure 4 – ***Moderate chronic hepatitis: hepatocytes with "ground glass" cytoplasm (HE stain, ob. ×20).***

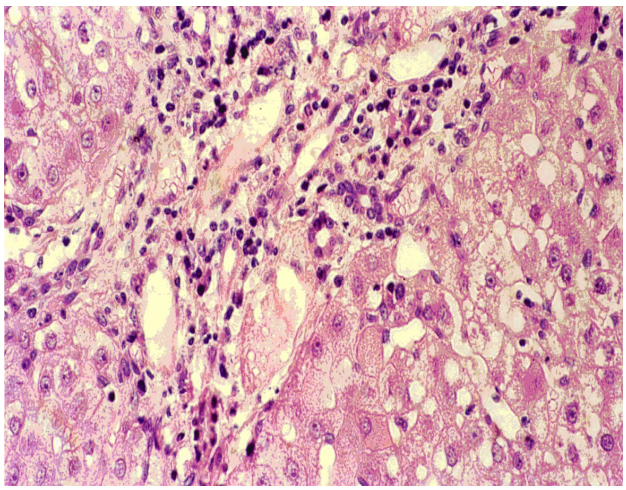

Figure 5 – ***Interface hepatitis: the portal inflammatory infiltrate extends towards the limiting plates (HE stain, ob. ×20).***

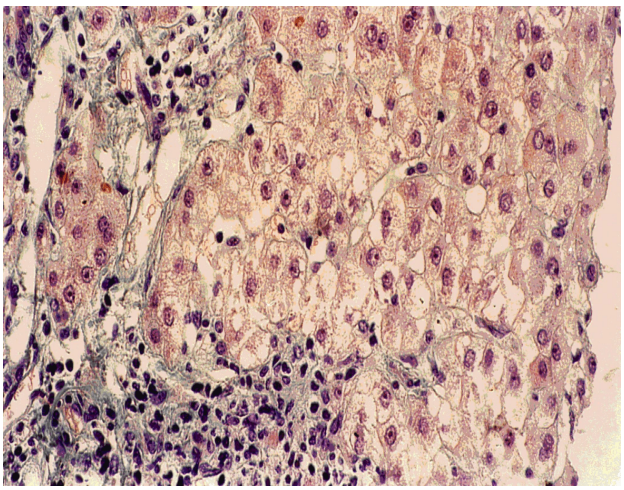

Figure 6 – ***Interface hepatitis: the portal inflammatory infiltrate penetrates into the hepatic lobule (HE stain, ob. ×20).***

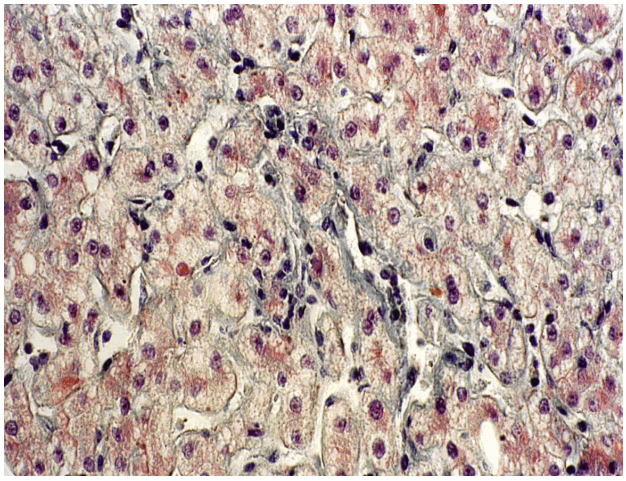

Figure 7 – ***Moderate chronic hepatitis: hepatocytes with "ground glass" cytoplasm, necroinflammatory foci (trichrome Szekely stain, ob. ×20).***

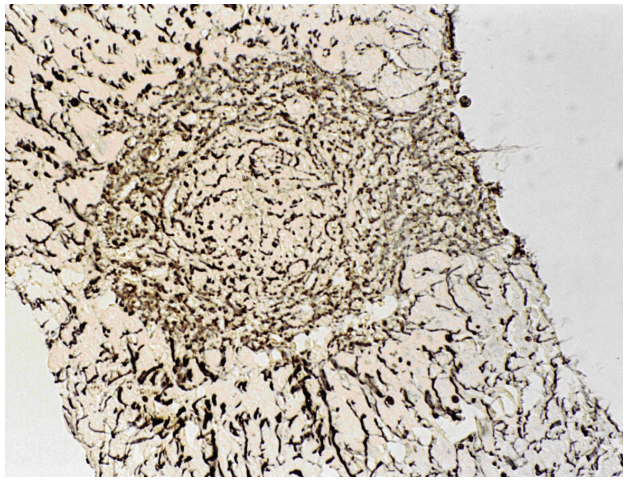

Figure 8 – ***Severe chronic hepatitis: disruption of the hepatic architecture (silver impregnation, ob. ×4).***

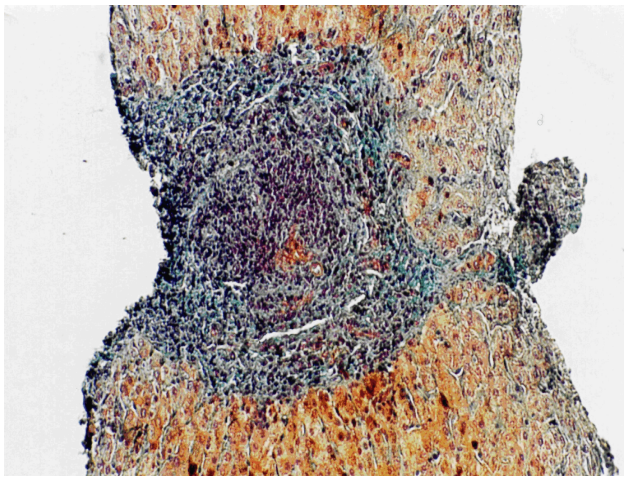

Figure 9 – ***Severe chronic hepatitis: massive inflammatory infiltrate in the portobiliary space, with invasion of the adjacent hepatic parenchyma (trichrome Szekely stain, ob. ×4).***

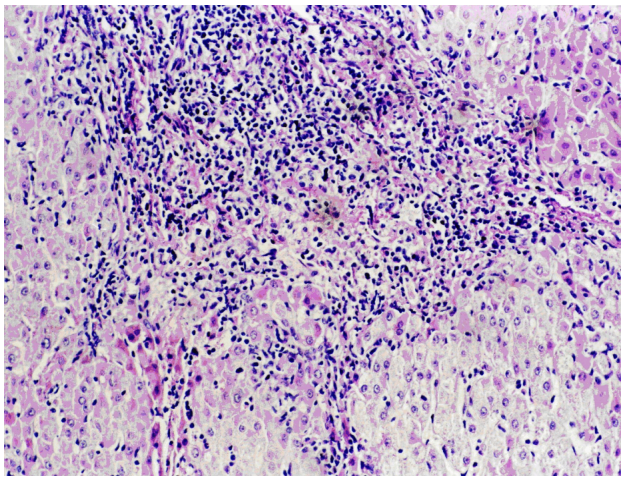

Figure 10 – ***Severe chronic hepatitis: important inflammatory (lymphocytic) infiltrate, numerous Councilman bodies (HE stain, ob. ×10).***

Chronic hepatitis B

For the 202 cases diagnosed with hepatitis B, the values conferred by the semiquantitative evaluation after the application of the Ishak scoring system for the NIA and fibrosis are presented in Table 1.

Table 1 – ***Distribution of the cases with hepatitis B based on NIA and fibrosis – Ishak scoring system***

NIA	No. of cases	NIA score	No. of cases	Fibrosis score	No. of cases
Mild Score 1–6	48	4	1	1	1
		5	38	1	38
		6	3	1	3
Moderate Score 7–10	148	7	92	1	82
				2	3
				3	7
		8	4	3	4
		9	49	3	49
		10	3	3	3
Severe Score 11–18	6	11	5	3	4
				4	1
		12	1	4	1

Thirty-nine cases from the total of 202 presented also chronic hepatitis D with the following NIA profile: seven mild cases, 27 moderate and five severe.

In the investigated group, the steatosis was associated with the NIA lesions as follows:

- For the mild NIA: from the total of 48 cases, in 34 it was absent, in 11 it was present in 10% of the whole specimen and in three cases it was present in 20% of the whole specimen;
- For the moderate NIA: from the total of 148 cases, in 99 it was absent, in 18 it was present in 10% of the whole specimen, in 27 cases it was present in 20% of the whole specimen and in 4 cases it was present in 30% of the entire specimen;
- For the severe NIA: from the total of six cases, in four it was absent, in one case it was present in 10% of the entire specimen and in one case it was present in 20% of the entire specimen.

Chronic hepatitis C

For the 751 cases diagnosed with hepatitis C, the values conferred by the semiquantitative evaluation after the application of the Ishak scoring system (for the NIA and fibrosis) as well as that of the METAVIR scoring system are presented in Table 2.

Table 2 – ***Distribution of the cases with hepatitis C in correlation with the Ishak (NIA, fibrosis) and the METAVIR scoring systems***

NIA	No. of cases	NIA score	No. of cases	Fibrosis score	No. of cases	METAVIR score	No. of cases
Mild Score 1–6	26	2	1	1	1	A1F1	23
		5	12	1	8		
				2	4		
		6	13	1	4		
				2	6		
				3	3	A1F2	3
Moderate Score 7–10	672	7 -10	672	1	3	A2F1	3
				2	30	A2F1	30
				3	606	A2F2	606
				4	31	A2F3	3
						A3F3	28
				6	2	A2F4	1
						A3F4	1
Severe Score 11–18	53	11	45	4	52	A3F3	53
		12	8	5	1		

In the investigated group, the steatosis was associated with the NIA lesions as follows:

▪ For the mild NIA: from the total of 26 cases, in nine it was absent, in eight it was present in 10% of the entire specimen, in six it was present in 20% of the entire specimen and in three cases it was present in 30% of the entire specimen;

▪ For the moderate NIA: from the total of 672 cases, in 328 it was absent, in 149 it was present in 10% of the entire specimen, in 167 it was present in 20% of the entire specimen and in 28 cases it was present in 30% of the entire specimen;

▪ For the severe NIA: from the total of 53 cases, in 16 it was absent, in 15 it was present in 10% of the entire specimen, in 17 it was present in 20% of the entire specimen and in five cases it was present in 30% of the entire specimen.

From the total of 751 cases, 16 cases presented also hepatitis B, with the following NIA profile: mild in two cases and moderate in 14 cases.

Discussion

The liver biopsy was and still remains a major diagnosis tool in the evaluation of chronic hepatitis patients, because it allows the assessment of the morphological background of the disease and consequently of the severity and expansion degree of the morphological changes in the hepatic parenchyma. In the last decade, the advances in the development of the biologic and imaging techniques lead to the introduction of new methods, less invasive than liver biopsy. These methods are based on the follow-up of the seric biochemical markers [14] and the imagistic evaluation of the liver through elastography, in order to measure its rigidity, with a correspondence in fibrosis [15–17]. However, the experts still rely on liver biopsy – not necessarily for the confirmation of the clinic diagnosis but rather for the estimation of the prognosis and the choice (or not) of the antiviral therapy [18].

The pathologic picture of the chronic hepatitis may vary considerably from case to case, but in essence these variations are due to the relationship between the three possible components: inflammatory reaction, fibrosis (or cirrhosis) and hepatocellular changes. The morphologic features are generally similar to all forms of chronic, viral or other types of hepatitis. Nevertheless, there are some distinct elements, which we will discuss in correlation with our study. In chronic hepatitis B, the hepatocytes with "ground glass" cytoplasm were evident, appearance caused by a massive production of AgHBs in the hepatocytes, which determines the proteic coating of the HBV to be accumulated in excess within the RER cisternae [19].

The morphologic image for the hepatitis C was characterized by the presence of lymphoid aggregates in the portobiliary spaces – which is considered to be a specific signature [19] and by lesions of the biliary pathways. Regarding the evolution of the disease, we must stress the caution with which the piecemeal and the bridging necrosis must be interpreted. Although these lesions do not imply an unavoidable progression of the disease, the loss of the hepatocytes continuity leads to the formation of fibrous septae, which, associated with the hepatocytic regeneration, triggers the development of cirrhosis [20–22].

Our study is focused on a morphologic analysis performed on almost 1000 liver biopsies investigated during a three years interval. The semiquantitative evaluation of the microscopic specimens was achieved based on a previous rigorous instruction; the scores granted being periodically ascertained through an inter-observers diagnostic appraisal.

Within this context, our results create the premises for a three-directional discussion: (*i*) the significance of the analysis on the correlation between the NIA and fibrosis, (*ii*) the motivation of the choice and application of a certain scoring system and (*iii*) the minimal condition necessary for the assessment of a liver biopsy.

Correspondences *vs.* differences between the NIA and the fibrosis

The correlation between the NIA and the fibrosis is essentially the main element which permits a prognostic evaluation and, at the same time, an adequate therapeutic decision.

In the chronic hepatitis B, our results reveal the fact that the moderate NIA (score 7–10) involves simultaneously an extremely variable development of fibrosis from the limited, restricted form (score 1) to the expanded form, characterized by fibrous bridges (score 4). On the contrary, for the mild and severe NIA, the correlation with a specific degree of fibrosis respects (in a certain manner) a better-defined pattern. Thus, the mild form (score 1–6) associates fibrosis score 1 (mild) while the severe form (score 11–16) associates fibrosis score 3 and 4 (severe).

Conversely, in the chronic hepatitis C, the analysis of the correspondence between the NIA and the fibrosis revealed a different pattern, with a wider variability. Firstly, the mild NIA (score 1–6) correlated with mild fibrosis (score 1), moderate (score 2) but also severe (score 3). We believe it is necessary to underline this variability recorded in the expression of fibrosis. Thus, fibrosis score 1 was present in the case of NIA score 2 but also in the case of a NIA score 6. At the same time, the NIA score 6 was associated with mild fibrosis (score 1), moderate (score 2) but also severe (score 3). Comparing the values of the scores, the development pattern of the fibrosis is similar with the image present in moderate NIA (score 7–10) in hepatitis B. For the moderate NIA (score 7–10), the expansion degree of the fibrosis ranges from score 1 to score 6 – similar to the results obtained in the mild NIA for the hepatitis B – with the remark that the overwhelming majority of the cases (606 out of 672) presented fibrosis score 3 – which is the first step on the scale of severe fibrosis quantification. Moreover, the results obtained for the severe NIA (score 11–18) were similar to the ones in hepatitis B cases, associated with severe fibrosis – with the remark that in this class of the classification the severity degree was higher (score 4 and 5).

The comparison between hepatitis B and C based on the NIA and fibrosis offers, through the numeric values of the Ishak scoring system, accurate proofs, which support the aggressivity of hepatitis C, because it develops fibrosis more quickly, even on the background of mild NIA. Also, our results reveal the fact that the NIA and the fibrosis are not processes which progress in a consistent pattern. The extreme diversity of the fibrosis degree reflects, in essence, the biologic individuality that defines the response to the viral aggression.

The application of the METAVIR scoring system for the cases with chronic hepatitis C confirmed the above-mentioned observation, namely that there is not a direct correlation between necroinflammation and fibrosis. Our data showed that a moderate NIA might be accompanied by fibrosis scores from 1 to 6. However, our findings revealed the occurrence of overlaps in the classification of some cases. Thus, score A3F3 resulted both for moderate NIA (score 7–10) with fibrosis F4, and for severe NIA (score 11–12) with fibrosis F4/F5. Another example is the score A2F1 – resulted for moderate NIA (score 7–10) with fibrosis F1 and F2, respectively. Hence, we must draw the attention on the possibility of an identical score for lesions with different intensity – which can be designated as an important limit of the METAVIR scoring system.

Score system selection

Our activity centered on the liver biopsy required the accumulation of abilities specific for this type of pathology. In time, the experience increased mainly through the comparative application of several scoring systems, in order to create our own opinion with respect to the informational advantages offered by each of them. Although the Scheuer classification, the METAVIR system and the Batts and Ludwig visual interpretation provide simple and easy to apply criteria [19], from our point of view the information obtained is not refined. Most interpretation issues regard the extremely different aspects of fibrosis. In these cases, the specialists recommend the Ishak staging system, because it makes available several classification steps and hence it increases the staging accuracy of the lesions [19].

The final purpose of the scoring systems used in the interpretation of the liver biopsy, by the evaluation of the degree and activity stage of the inflammatory disease and fibrosis, is to convey clear and precise information to the clinician [19]. Consequently, the choice and the usage of an operational scoring system must be founded on [19]: (1) the existence of a consensus between pathologists and clinicians on the pathologic criteria quantified in the selected scoring system; (2) the concise and consistent formulation, in the pathology report, of the information about classification and staging; (3) the possibility to apply the obtained information in diagnosis and therapy.

Based on our experience in the semiquantitative evaluation of liver biopsies, both for hepatitis B and C, we use the Ishak scoring system. Our choice is supported by the fact that the wide range of numeric values attributed for the evaluation of NIA and fibrosis provides far more precise criteria for the appraisal of the degree of damage to the hepatic parenchyma at the time of the diagnosis. Supplementary to the Ishak scoring system, for the hepatitis C we perform at the same time a METAVIR scoring system assessment, because it allows an assessment of the entire histologic activity, with the addition of the interface hepatitis and of the associated lobular necrosis components.

Minimal requirements in the evaluation of a liver biopsy

The controversies in the literature on the histologic grading and the staging systems may be explained by the possible sampling errors. We believe that the liver biopsy is a representative sample for the hepatic parenchyma. However, we must take into account the fact that the liver biopsy, regardless of its basic length or width, is only a finite segment from an organ with an immense potential diversity in the expression of disease [19]. The pathologists obviously prefer larger samples for the interpretation [23], and comparative data indicate the fact that 20 mm long samples are necessary, which have to include at least 11 portobiliary spaces in order to have an optimal specimen for grading and staging [24]. In our study group, each microscopic specimen had between 3 and 12 portobiliary spaces, with an average of 6 to 8 – a value that can support a correct diagnostic evaluation.

The diagnosis based on the semiquantitative assessment of the lesions, according to the parameters included in the scoring system, relies on the examination of the microscopic specimens in special stains: trichrome Szekely, Gordon–Sweet silver impregnation, PAS. A last comment must be made with respect to the expertise in the field – which corresponds to the recommendations in the literature [25] – the main investigator having 10 years experience in liver pathology – fact that increases the coherence and the accuracy of the diagnosis and decreases the issues associated with the dimensions of the sample.

Conclusions

The qualitative and semiquantitative appraisal of the liver biopsy allows an adequate evaluation of the severeness of the liver injury. However, a certain degree of subjectivity unavoidably intervenes in the use of the operational scores for the quantification of the lesions. The significant decrease of the subjective factor in the assessment implies a solid experience in the field which leads to the choice of a particular score system or the comparative application of several systems in complete consensus with the clinician.

References

[1] Lai M, Liaw YF, *Chronic hepatitis B: past, present, and future*, Clin Liver Dis, 2010, 14(3):531–546.

[2] Munir S, Saleem S, Idrees M, Tariq A, Butt S, Rauff B, Hussain A, Badar S, Naudhani M, Fatima Z, Ali M, Ali L, Akram M, Aftab M, Khubaib B, Awan Z, *Hepatitis C treatment: current and future perspectives*, Virol J, 2010, 7:296.

[3] Kalk H, *Die chronische Verlaufsform der Hepatitis epidemica in ihrer Beziehung zu ihren anatomischen Grundlagen*, Dtsch Med Wochenschr, 1947, 72:308–313.

[4] Smetana HF, *The histologic diagnosis of viral hepatitis by needle biopsy*, Gastroenterology, 1954, 26(4):612–625; discussion 640–644.

[5] Saint EG, King WE, Joske RA, Finckh ES, *The course of infectious hepatitis with special reference to prognosis and the chronic stage*, Australas Ann Med, 1953, 2(2):113–127.

[6] De Groote J, Desmet VJ, Gedigk P, Korb G, Popper H, Poulsen H, Scheuer PJ, Schmid M, Thaler H, Uehlinger E, *A classification of chronic hepatitis*, Lancet, 1968, 2(7568): 626–628.

[7] Popper H, Schaffner F, *The vocabulary of chronic hepatitis*, N Engl J Med, 1971, 284(20):1154–1156.

[8] Batts KP, Ludwig J, *Chronic hepatitis. An update on terminology and reporting*, Am J Surg Pathol, 1995, 19(12): 1409–1417.

[9] Desmet VJ, Gerber M, Hoofnagle JH, Manns M, Scheuer PJ, *Classification of chronic hepatitis: diagnosis, grading and staging*, Hepatology, 1994, 19(6):1513–1520.

[10] Scheuer PJ, *Classification of chronic viral hepatitis: a need for reassessment*, J Hepatol, 1991, 13(3):372–374.

[11] Knodell RG, Ishak KG, Black WC, Chen TS, Craig R, Kaplowitz N, Kiernan TW, Wollman J, *Formulation and application of a numerical scoring system for assessing histological activity in asymptomatic chronic active hepatitis*, Hepatology, 1981, 1(5):431–435.

[12] Ishak K, Baptista A, Bianchi L, Callea F, De Groote J, Gudat F, Denk H, Desmet V, Korb G, MacSween RNM, Phillips MJ, Portmann BG, Poulsen H, Scheuer PJ, Schmid M, Thaler H, *Histological grading and staging of chronic hepatitis*, J Hepatol, 1995, 22(6):696–699.

[13] Bedossa P, Poynard T, *An algorithm for the grading of activity in chronic hepatitis C. The METAVIR Cooperative Study Group*, Hepatology, 1996, 24(2):289–293.

[14] Poynard T, McHutchison J, Manns M, Myers RP, Albrecht J, *Biochemical surrogate markers of liver fibrosis and activity in a randomized trial of peginterferon alfa-2b and ribavirin*, Hepatology, 2003, 38(2):481–492.

[15] Colletta C, Smirne C, Fabris C, Toniutto P, Rapetti R, Minisini R, Pirisi M, *Value of two noninvasive methods to detect progression of fibrosis among HCV carriers with normal aminotransferases*, Hepatology, 2005, 42(4):838–845.

[16] Castéra L, Vergniol J, Foucher J, Le Bail B, Chanteloup E, Haaser M, Darriet M, Couzigou P, De Lédinghen V, *Prospective comparison of transient elastography, Fibrotest, APRI, and liver biopsy for the assessment of fibrosis in chronic hepatitis C*, Gastroenterology, 2005, 128(2):343–350.

[17] Ghany MG, Doo E, *Assessment of liver fibrosis: palpate, poke or pulse?* Hepatolog, 2005, 42(4):759–761.

[18] Almasio PL, Niero M, Angioli D, Ascione A, Gullini S, Minoli G, Oprandi NC, Pinzello GB, Verme G, Andriulli A, *Experts' opinions on the role of liver biopsy in HCV infection: a Delphi survey by the Italian Association of Hospital Gastroenterologists (A.I.G.O.)*, J Hepatol, 2005, 43(3):381–387.

[19] Lefkowitch JH, *Liver biopsy assessment in chronic hepatitis*, Arch Med Res, 2007, 38(6):634–643.

[20] Okuno T, Okanoue T, Takino T, Mori K, *Prognostic significance of bridging necrosis in chronic active hepatitis*, Gastroenterol Jpn, 1983, 18(6):577–584.

[21] Marcellin P, Asselah T, Boyer N, *Fibrosis and disease progression in hepatitis C*, Hepatology, 2002, 36(5 Suppl 1): S47–S56.

[22] Neuman MG, Sha K, Esguerra R, Zakhari S, Winkler RE, Hilzenrat N, Wyse J, Cooper CL, Seth D, Gorrell MD, Haber PS, McCaughan GW, Leo MA, Lieber CS, Voiculescu M, Buzatu E, Ionescu C, Dudas J, Saile B, Ramadori G, *Inflammation and repair in viral hepatitis C*, Dig Dis Sci, 2008, 53(6):1468–1487.

[23] Demetris AJ, Ruppert K, *Pathologist's perspective on liver needle biopsy size?* J Hepatol, 2003, 39(2):275–277.

[24] Colloredo G, Guido M, Sonzogni A, Leandro G, *Impact of liver biopsy size on histological evaluation of chronic viral hepatitis: the smaller the sample, the milder the disease*, J Hepatol, 2003, 39(2):239–244.

[25] Rousselet MC, Michalak S, Dupré F, Croué A, Bedossa P, Saint-André JP, Calès P; Hepatitis Network 49, *Sources of variability in histological scoring of chronic viral hepatitis*, Hepatology, 2005, 41(2):257–264.

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