INVITATION LECTURES

2. INFECTIVE PATHOLOGY

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Ultrastructural hepatocyte changes in HBV and HCV infection

KEYWORDS: Hepatitis C, Ultrastructure, Viral envelope proteins, Viral core proteins

INTRODUCTION

The hepatitis C virus (HCV) is known as a flavivirus and was identified in 1989. Prior to its identification it was referred to as "non A non B" hepatitis, or NANB. HCV is most unusual for an RNA virus because approximately 80% of infected individuals develop persistent infection. The reason for this remains unclear, but it has been suggested that low levels of replication and expression of HCV may result in evasion of the host defense that might promote persistence and ensure survival. Hepatitis C viruses are single stranded, enveloped, positive sense RNA virus-(+)ve-sense ssRNA, which have encoding and nonencoding sequences for virion structural proteins (the core protein, the virion coat glycoprotein E1, coats glycoprotein 2/nonstructural protein 1-E2/NS1). The RNA virion 3' carbocsil end encodes structural proteins which are included in the virion replication (NS2, NS3, NS4, NS5). Hepatitis B virus (HBV) is known as a hepadnavirus, whose DNA structure is double stranded and circular (dsDNA). The hepatitis B viron consists of a surface and a core. The core contains a DNA polymerase and the e antigen. There are four major polypeptide reading frames (genes): the S (surface), the C (core), the P (polymerase) and the X (transcriptional transactivating). Initially there appeared to be three particles associated with hepatitis B infection: a large "complete" particle called the "Dane particle", a small circular 20nm particle and an oblong 40nm particle. Further research identified the Dane particle as the hepatitis B viron and the other two particles as excess surface protein. The S gene consists of three regions, the pre-S1, pre-S2 and encodes the surface proteins (HBsAg). The C gene is divided into two regions, the pre-core and the core, and codes for two different proteins, the Core antigen (HBcAg) and the e antigen (HBeAg).

MATERIAL AND METHODS

Tip biopsies from 38 patients with HBV and 54 patients with HBC were taken at the Clinic Of Tropical And Infectious Diseases. After ordinary fixation and processing, the electron microscope (EM) analysis was performed on Phillips EM 201C and 208 S.

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RESULTS AND DISCUSSION

Hepatitis C virions is not noticeable in hepatocytes, but only ultrastructural alterations that are caused by presence of the virus. Hepatitis B virions are 42nm in diameter and possess an isometric nucleocapsid or "core" of 27nm in diameter, surrounded by an outer coat approximately 4nm thick. The protein of the virion coat is called the "surface antigen" or HBsAg. It is sometimes extended as a tubular tail on one side of the virus particle. The surface antigen is generally produced in vast excess, and is found in the blood of infected individuals. The filamentous particles are identical to the virion "tails" - they vary in length and have a mean diameter of about 22nm. The nucleoli are very active and dynamic components of the infected nucleus. Due to different needs of the cells for protein synthesis and increased need for ribosome, the nucleolus are also different. If the virion replication in the infected cell is active, the nucleoli are morphologically and specifically changed by that virus. The nucleoli are dispersed without clear granular and fibrillar centers. The nucleoli that are changed by the virus, are noticeable in HBV with positive HBeAg, and in HBC most often in group CPHc-CAHa (57.7%). Morphologically expressed forms of HCV (CAH b,c) have a distinct active nucleolus (62%) with multiple and bulky fibrillary centers, without disperse structure. Unchanged nucleoli are found most frequently within chronic persistent hepatitis CPH a, b (75%). These differences are statistically significant. The level of serum transaminases in HBC is increased in patients whose nucleoli are changed by the virus, in relation to the patients with synthetically active nucleoli ($p>0,05$), and to those whose nucleoluses are not changed ($p<0.05$). These patients whose nucleoluses were not changed, but who had a histopathologically active disease, probably didn't have a significant replication of the virus in the moment of biopsy taking. The virus replication had started before taking the biopsy and started the cascade of events. Inclusions in the nucleus, "nuclear bodies" (NB) are a consequence of the increased transport of the nucleolar material, due to increased synthesis of proteins and they are often noticeable in the karyoplasm of the hepatocytes in HCV infection. Presence of NB is noticeable in 72.9% of patients and there is no significant difference between different histological forms of the disease ($p>0.05$). The nucleus of hepatocytes in HBV infection (HBcAg+) are "sandy", filled with small spherical intranuclear viral inclusions of 22nm in diameter. The proliferation of the smooth endoplasmic reticulum (sER) is the constant finding in HBsAg positive patients, because maturation of the complete viron particle occurs in sER. Like in many positive sense RNA viruses, HCV replication and infection is associated with cytoplasmic membrane rearrangements in the cell. The lameral inclusions (85.4% of patients) are present in the cytoplasm of hepatocytes with HCV infection and it is not in correlation with the histological form of hepatitis. It still remains unclear which HCV structural protein is responsible for these morphological changes. The lipid infiltration in hepatocytes are not a specific finding in HBV infection, while they are also very common in HCV infection. The simple and complex lipid inclusions (which are the mark of toxic liver damage) are found in 89.1% of HCV biopsies.

CONCLUSION

Based on HCV ultrastructural findings, we can assume that in the first phase of its direct cytopathogenic influence, the virus causes toxic damages of hepatocytes. When viron replication is on the certain level, synthesis of proteins intensifies. New proteins incorporate in hepatocyte cytomembranes, the hepatocyte surface antigen structure changes, new proteins become a target of immunocompetent cells, and all that leads to clinical and morphological activation of the disease.

REFERENCES

1. Alter H. Discovery of non-A, non-B hepatitis and identification of its etiology. Am J Med 1999;27:16S-20S.

2. Adinolfi LE, Andreana A, Utili R, Zampino R, Ragone E, Ruggiero G. HCV RNA levels in serum, liver, and peripheral blood mononuclear cells of chronic hepatitis C patients and their relationship to liver injury. Am J

Gastroenterol 1998;93:2162-6.

3. Meehyein K, Hajeong K, Seong-PCh, Mi-Kyung M, Template requirements for De Novo RNA synthesis by hepatitis C virus nonstructural protein 5B polymerase on the viral X RNA. J Virol 2002;76: 6944-56.

4. Guidotti G, Eggars M, Raney K, Chi Y, Peters M, Gonzalez J, McLachian A. In vivo regulation of hepatitis B virus replication by peroxisome proliferation. J Virol 1999;73:10377-82.

5. Keith DT, Kazutoshi M, Aleem S. Hepatitis C Virus subgenomic replicons induce endoplasmic reticulum stress activating an intracellular signaling pathway. J Virol 2002; 76:7453-59.

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Immunomorphological characteristics of inflammatory infiltrates in the liver with chronical hepatitis B and C

KEYWORDS: Hepatitis C; Hepatitis B; Lymphocytes; Immunohistochemistry

Cell-mediated immune reactions could be important for the hepatocellular damage in viral chronic hepatitis. We investigated immunomorphological characteristics of intrahepatic lymphocyte subtypes and non-lymphoid effector cells in relation to histopathological lesions with similar histological types of these diseases, separately and as compared to the liver parenchyma in chronic hepatitis B and C. Does the number of lymphocytes with different immunofenotypes depend on etiology, grade or stadium of hepatitis? We analyzed 72 puncture and 2 surgery biopsies, 38 with chronic hepatitis B and 36 with chronic hepatitis C, with different activity grades and fibrosis. The paraffin-embedded liver tissue sections were stained with antibodies to CD3, CD8, CD20, CD45RO, CD57, CD68, HLA-DR, S- 100 and EMA by DAKO LSAB2 System. The slides were analyzed with computerized image analysis CAMIA program. The imaging findiongs were statistically processed.There is a great level of variation in the number of cells with examined fenotypes (NCEF) independent of the etiology, grade and the stage of hepatitis. There is a slight positive linear correlation between the grade and NCEF in both B and C hepatitis. The gradient of the NCEF increase is higher in hepatitis C than in B. There is no correlation between the stage of hepatitis and the NCEF. The current grading sistem of chronic hepatitis is arteficial and is not based on the relevant factors which are important for pathogenesis of chronic hepatitis.

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Testing of histological parameters for predicting the fibrosis in drug abusers with chronic C viral infection

KEYWORDS: Fibrosis; Drug abusers; Hepatitis C

Chronic Hepatitis C (CHC) is present in drug abusers of young age. Among other effects, the toxic effects on the immunologic system in these patients modulate the total immunological response to the viral infection, so it is important to find appropriate histological parameters for the evaluation and prognosis of the formation of fibrosis in these patients. Testing of the histomorphologic features combined with the likelihood of fibrosis formation in drug abusers with CHC. Forty liver biopsies with CHC (15 active drug abusers, 15 persons who quit narcotics 3-5 months before the liver biopsy, and 10 patients with CHC without a history of drug use). A semi-quantitative analysis was performed using histological parameters that are routinely analyzed during the evaluation of the histological index of activity using a standardized questionnaire and a comparison with the extent of fibrosis that is established using a statistical method. The histological parameter which predicts a development of fibrosis in CHC cases is a lobular activity of persons who are not drug addicts. In two other groups of patients this particular parameter does not correspond to the extent of actual fibrosis found in the patients. The histomorphological changes and the extent of fibrosis are in a direct correlation with the duration of drug use. The morphological expression of the immunologic toxicity in drug usage and CHC make the CHC in this population group a special entity to which it is not possible to apply the standard pathologic-histological parameters.