SHORT ARTICLE

A study of the evolution of specific and non-specific immune complexes in acute hepatitis B and chronic hepatitis

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Summary. Circulating immune complexes (ICs) containing IgG and HBsAg, and IgG and HBeAg, in sera from groups of patients with various liver diseases were sought by ELISA and immunodiffusion. A correlation was found between the absence of ICs and the disappearance of HBsAg in patients who had recovered from acute hepatitis B, but complexes containing HBsAg were always found in chronic hepatitis.

Introduction

The lesions of “immune complex disease” are caused by the deposition in tissues of circulating antigen-antibody complexes (Germuth *et al.*, 1967; McCluskey *et al.*, 1971). Immune complexes (ICs) have been demonstrated in numerous conditions, in health and disease. The presence of ICs has been demonstrated in hepatitis B (Almeida and Waterson, 1969; Araki *et al.*, 1982) and certain diseases related to hepatitis B virus (HBV) infection (Alpert *et al.*, 1971; Prince and Trepo, 1971; Carella *et al.*, 1977; Furuse *et al.*, 1982).

This study of 39 patients with acute hepatitis B and chronic HBsAg-positive hepatopathy examines the occurrence and development of ICs.

Materials and methods

The progress of 22 patients with acute hepatitis B (AHB) and 17 patients with chronic HBsAg-positive hepatopathies (CH HBsAg+) was studied for 2–12 months, depending upon the condition of each patient.

Sera were tested by ELISA (Abbot) for HBsAg, antiHBs, antiHBe, HBeAg and antiHBe.

Blood was tested for circulating ICs by two methods. (i) Precipitation with polyethylene glycol (PEG) 6000 (2% final concentration) and subsequent identification of ICs by simple immunodiffusion (ID) (Behring). This test detects ICs formed by IgG and any type of antigen. For this reason we refer to them as “non-specific” (non-sp) ICs. (ii) Precipitation with PEG 6000 (2% final concentration) and identification of HBsAg ICs and HBeAg ICs

in the precipitate by ELISA, Auszyme II and HbeAg/antiHBe ELISA, respectively.

These techniques were slightly modified by a change of the controls and the introduction of a correction factor for interpreting the results (Leyva *et al.*, 1987).

Results

Table I shows the results obtained with the 22 AHB patients studied. In the first serum samples, 19 patients had HBsAg as well as HBs ICs and three patients had only HBsAg. Second samples showed no change in 13 of the 19 patients, whereas the other six had lost both antigen and complexes. The three patients with HBsAg only also had negative results with second samples.

HBe ICs were found in four of the five HBeAg-positive patients and there was no change during the period of study.

When non-specific ICs were studied in relation to the presence of HBsAg, the number of patients with both HBsAg and non-sp ICs stayed the same in the two samples. Of the 18 HBsAg-positive patients without non-sp ICs, seven remained unchanged, two retained HBsAg and acquired non-sp ICs and nine lost their HBsAg.

We did not attempt to establish a relationship between non-sp ICs and HBeAg because the latter is not found in the absence of HBsAg.

HBsAg ICs were found in all 17 cases of CH HBsAg+ (table II).

HBeAg ICs were found in five out of the seven patients with HBeAg in serum. These findings did not change during the study. The only change in

Table I. Evolution of HBsAg ICs, HBeAg ICs and non-specific ICs in relation to HBs and HBe antigens in 22 patients with acute hepatitis B

Results obtained with first serum sample		Results obtained with second serum sample	
HBsAg + ; HBsAg IC +	19	13 HBsAg + ; HBsAg IC +	
		6 HBsAg - ; HBsAg IC -	
HbsAg + HBsAg IC -	3	3 HBsAg - ; HBsAg IC -	
HBeAg + ; HBeAg IC +	4	4 HBeAg + ; HBeAg IC +	
HBeAg + ; HBeAg IC -	1	1 HBeAg + ; HBeAg IC -	
HBeAg - ; HBeAg IC -	17	17 HBeAg - ; HBeAg IC -	
HBsAg + ; non-sp IC +	4	4 HBsAg + ; non-sp IC +	
		7 HBsAg + ; non-sp IC -	
HBsAg + non-sp IC -	18	2 HBsAg + ; non-sp IC +	
		9 HBsAg + ; non-sp IC +	

second samples from these patients was the appearance of non-sp ICs in three initially without them.

Discussion

Recently, the importance of circulating immune complexes in numerous diseases has been discussed more frequently (Report, 1977). It is generally assumed that ICs in slight antigen excess may be responsible for the development of IC disease (Pernice and Sedlacek, 1979).

The surveillance of 39 patients diagnosed as having acute hepatitis B or chronic HBsAg-positive hepatopathies has enabled us to make some noteworthy observations.

The absence of HBsAg ICs in three patients with AHB who were still HBsAg-positive (table I) could

be a finding indicating a favourable outcome for the patients, because their second serum samples showed that they had lost their HBsAg and were recovering from the disease. Moreover, in all the patients who retained their ICs, as noted by Pernice and Sedlacek (1979), a good correlation was found between the level of HBsAg-containing immune complexes and the clinical state. ICs could be detected simultaneously with HBsAg and either decreased or disappeared before the appearance of free antiHBs. However, authors such as Nydegger *et al.* (1974) observed that the order of appearance of the various reactants in building up immune complexes during HBV infection may be variable; circulating immune complexes may be formed 2-3 months after HBV infection. Careoda *et al.* (1982) observed that the persistence of ICs in the early phase of acute hepatitis could indicate impending

Table II. Evolution of HBsAg ICs, HBeAg ICs and non-specific ICs in relation to HBs and HBe antigen in 17 patients with chronic hepatopathies

Results obtained with first serum sample		Results obtained with second serum sample	
HBsAg + ; HBsAg IC +	17	17 HBsAg + ; HBs IC +	
HBsAg + ; HBsAg IC -	0	...	
HBeAg + ; HBeAg IC +	5	5 HBeAg + ; HBeAg IC +	
HBeAg + ; HBeAg IC -	2	2 HBeAg + ; HBeAg IC -	
HBeAg - ; HBeAg IC -	10	10 HBeAg - ; HBeAg IC -	
HBsAg + ; non-sp IC +	8	8 HBsAg + ; non-sp IC +	
		6 HBsAg + ; non-sp IC -	
HBsAg + non-sp IC -	9	3 HBsAg + ; non-sp IC +	

chronicity. These findings agree with our results in that HBs ICs were always detected in the chronic hepatopathies. Nevertheless, others (Trepo *et al.*, 1974) believe that no relationship exists between the presence of this type of IC and an unfavourable outcome of the disease.

As the number of patients with HBe ICs is small we are not able to determine whether the absence of HBe ICs in the presence of HBeAg has any prognostic significance.

The detection of non-specific ICs showed no change in the AHB patients who eliminated HBsAg. On the other hand, the number of non-sp IC-positive cases rose over the period of study in the AHB patients who did not show signs of recovery and in the CH patients. This observation seems to indicate that the ID technique, which is less sensitive than ELISA, requires time for a build-up of circulating ICs in order that they may be detected.

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