e Antigen and antibody in outbreaks of hepatitis B in two renal dialysis units

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SUMMARY In two renal dialysis units which had outbreaks of hepatitis B, patients were studied for the presence of e antigen and antibody. In one unit, in which the illness was mild and most of the patients (78%) became carriers of HBsAg, all but one patient had e antigen. Most of the HBsAg carriers had persisting e antigen and most of the transient patients had also transient e antigen. In the other unit, in which the illness was more severe, none of the patients became a carrier. Only 30% had e antigen during the acute phase of illness and 40% developed anti-e just before clearing HBsAg. It seems possible that the differences between these two units may reflect strain differences in the infectivity of hepatitis B virus.

The e antigen/antibody system was first described by Magnius and Espmark (1972), who suggested that the presence of e antigen might indicate infectivity while the antibody occurred only in non-infectious HBsAg positive sera. The correlation of e antigen with large numbers of 'Dane' particles, and its absence in sera containing no 'Dane' particles (Nielsen *et al.*, 1974; Neurath *et al.*, 1976), also seems to suggest a relationship to infectivity. Neurath *et al.* (1976) suggested that e antigen is present as an additional antigenic site on the surface of the 'Dane' particles and tubular forms of HBsAg as well as in the form of a soluble antigen. Nielsen *et al.* (1974) suggested that it might be a valuable prognostic marker because it was present in the sera of 58% of cases of acute hepatitis B which progressed to chronic hepatitis or cirrhosis.

In the present study, the presence was determined of e antigen and antibody in sera from patients in two renal dialysis units in which the outbreaks of hepatitis B (during 1969-71) were quite different (Polakoff *et al.*, 1972). In dialysis unit A, 32 of the infected patients became carriers of HBsAg. Of these, consecutive serum samples were available from 27. In dialysis unit B, none of the infected patients became a carrier. Consecutive serum samples were available from all 10 individuals.

Material and methods

Sera taken from patients with hepatitis B infection in the two renal dialysis units were examined for e

antigen and antibody. In unit A, consecutive serum samples were available from the patients for periods of five months to five years after the onset of antigenaemia. In unit B, serum samples were available from the patients from the onset of antigenaemia until they cleared HBsAg. Serum samples taken just before hepatitis B infection in the patients of both units were also examined for e and anti-e.

All the samples were first tested by immunoelectrophoresis (IEOP) for HBsAg activity.

The immunodiffusion test was used to detect e antigen and antibody. The gel consisted of 0.9% agarose (Indubiose, l'Industrie Biologique Francaise) in tris buffer. The test was performed in 5 cm Petri plates with the gel 3 mm thick. Six wells were cut surrounding a central well, each 6 mm in diameter with approximately 10 mm between the centres of the wells. Each well was twice filled with sera.

The anti-e serum was obtained from a blood donor who was a carrier of HBsAg. The standard e antigen was serum obtained from a carrier of HBsAg who had chronic liver disease. Reference e antigen and anti-e sera were kindly supplied by Dr J. O. Nielsen (Copenhagen) and Dr L. O. Magnius (Stockholm).

Results

The frequency of e antigen and antibody in the HBsAg carriers of two renal dialysis units is shown in Table 1. In unit A, in which most of the individuals with HBs antigenaemia progressed to become carriers of HBsAg, all of those followed up except one

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Table 1 *Presence of e antigen and antibody in two dialysis unit outbreaks of hepatitis B*

Category	Total	Persisting e	Transient e	Anti-e	Neither e nor anti-e
Unit A					
HBsAg carriers	21 (78%)	19	2	0	0
Transient HBsAg	6 (22%)	0	5	1	1
Total	27	19	7	1	1
Unit B					
HBsAg carriers	0	0	0	0	0
Transient HBsAg	10 (100%)	0	3	4	3
Total	10	0	3	4	3

Table 2 *Duration of e antigen and antibody in postinfection sera from transient carriers of hepatitis B in two dialysis unit outbreaks*

Unit	Duration of HBs antigenaemia	Number	Detectable e at (months)			Detectable anti-e at (months)			Neither e nor anti-e
			1	2	3	1	2	3	
A									
(6 cases)	<1 month	1	1	—	—	0	—	—	0
	1-2 months	4	3	2	—	0	0	1*	1
	2-3 months	1	1	1	1	0	0	0	0
	Total	6	5	3	1				
B									
(10 cases)	<1 month	5	3	—	—	1	—	—	1
	1-2 months	4	0	0	—	0	2	—	2
	2-3 months	1	0	0	0	0	0	1	0
	Total	10	3	0	0				

*This specimen was HBsAg negative

had e antigen in their sera. No anti-e was detected in the carriers and only one of those with transient HBsAg had detectable anti-e. In this individual e antigen was detected within the first month of HBs antigenaemia. At two months the serum was HBsAg positive and e negative. At three months the serum was HBsAg negative and contained anti-e.

In unit B, in which no patient became a carrier of HBsAg, 30% of the transient cases had detectable e while 40% had detectable anti-e. In these patients (Table 2) e antigen was found only within the first month of antigenaemia and anti-e was found only just before the disappearance of HBsAg from the serum. In this unit, the presence of e antigen did not imply a longer period of HBs antigenaemia. In fact, in all three patients in whom e antigen was detected, HBs antigenaemia lasted for less than one month while three or the four patients in whom anti-e was detected had HBsAg in the sera for periods longer than one month. In contrast, the transient HBsAg carriers in unit A in whom e was detected had detectable e for almost the complete duration of their HBs antigenaemia. There appeared to be no difference in the duration of HBs antigenaemia in the transient carriers between the two units although e antigen did tend to persist longer in unit A.

The presence or absence of e antigen in the serum

was not related to the titre of HBsAg, as seen in Table 3.

Table 3 *Non-correlation of e antigen with the titre of HBsAg in different serum samples*

Serum	e Ag	HBsAg titre (IEOP)
1	+	64
2	—	32
3	+	4
4	+	16
5	—	8
6	— (anti-e)	16
7	—	16
8	— (anti-e)	16
9	+	0
		(detectable only by HA and RIA)

Discussion

Many studies of the e antigen/antibody system have related the features of individual cases to the presence of e antigen or antibody (Eleftheriou *et al.*, 1975; El Sheikh *et al.*, 1975; Feinman *et al.*, 1975; Nordenfelt and Kjellen, 1975; Vogten *et al.*, 1976). This report is concerned with the epidemiological significance of the e system.

The two dialysis unit outbreaks have previously been compared (Polakoff *et al.*, 1972). In unit A, the classical pattern was observed, many of the patients experiencing very mild symptoms of hepatitis at the time HBsAg was first detected. Of the infected patients, 78% became established HBsAg carriers and all of these had e antigen detectable from the acute phase onwards. In only two cases (10%) did the e antigen disappear. Similar findings have been reported by Nordenfelt *et al.* (1975). In their series, most of the patients showed histological evidence of chronic hepatitis. Of the six patients in unit A who cleared HBsAg five had e antigen at the onset of hepatitis. From this it appears that determinations of e have no prognostic value in predicting the outcome of individual cases.

In unit B there were fewer patients and these experienced much more acute symptoms of hepatitis than is expected in dialysis unit outbreaks. Moreover, all cleared the antigen within four months of onset. Only 30% of the infected patients in the unit produced e antigen during their illness. Contrary to the results of Norkrans *et al.* (1976) in their study of acute hepatitis B, the presence of e antigen was not related to a longer duration of HBs antigenaemia.

There were marked differences in the detection of e antibody in these outbreaks. Only one patient in unit A produced anti-e shortly after the appearance of e antigen, and both were present transiently. In unit B, however, four of the 10 infected patients produced anti-e transiently just before they finally cleared HBsAg. It is unlikely that the difference in the courses of the hepatitis B outbreaks is due to different methods of selection or treatment of patients, nor can it be attributed to a difference in the subtype since ay was involved in both units. However, it does seem that e antigen and antibody determinations may be useful in predicting the course of dialysis outbreaks.

These results also suggest that the occurrence of e antigen or antibody could be related to the nature of the infecting strain rather than to the individual patient's response.

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