ANTIGEN IN THE SERUM OF HBs ANTIGEN-POSITIVE PATIENTS ON MAINTENANCE DIALYSIS AND AFTER TRANSPLANTATION

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Summary

The 'e antigen' (eAg) is specifically associated with hepatitis B virus infections and appears to be a marker for the infectivity and a prognostic indicator of the chronicity of liver disease. Therefore we examined by immunodiffusion the presence of eAg in the serum of HBsAg-positive patients on maintenance dialysis. The dialysis patients had a significantly higher incidence of positive eAg compared with a group of unselected HBsAg-positive patients without renal failure. In most of the dialysis patients the microscopic findings in the liver revealed only 'minimal changes'. Three eAg-positive patients received a renal transplant. Afterwards they displayed an appreciably increased eAg-yield on immunodiffusion and histology revealed chronic persistent hepatitis. It is assumed therefore that the immunodeficiency of patients undergoing chronic haemodialysis is possibly a supporting factor in the synthesis of eAg, and will perhaps induce a more subacute and prolonged course of hepatitis. The synthesis of eAg after renal transplantation may be enhanced by the additional immuno-suppressive therapy.

Introduction

Hepatitis B virus infection is one of the most frightening complications in regular dialysis therapy. In the majority of the European dialysis centres therefore, periodical screening for HBs antigen in patients and staff members is usually performed early to detect the presence of hepatitis and to control the disease. In patients on maintenance dialysis hepatitis B generally results in the persistence of HBs antigen; the importance of this persistence for the infectivity and the prognosis is not clear. In 1972 Magnus and Espmark [1] detected a new antigen-antibody system in HBsAg-positive sera, the 'e system'. The e antigen or HBes antigen according to the new WHO nomenclature has been identified, as yet, only in association with HBs antigen. The corresponding antibody (anti-HBes) is often found in HBsAg carriers with clinically in-
apparent disease. Meanwhile it has been shown that the e system probably represents an antibody/anti-antibody system [2]. eAg has the physiochemical and immunological properties of an immunoglobulin, predominantly of the IgG 4 subclass. Presumably the ‘e’-determinants represent idiotypic determinants on antibodies. A higher frequency of eAg in HBsAg-positive haemodialysed patients was reported by Magnus and Espmark in 1972 [1] and by Thamer et al in 1976 [3]. Several investigations point to the fact that eAg displays a prognostic value [4–6]. In the presence of eAg, rapid development of chronic liver disease is said to be possible. Even the infectivity of HBsAg carriers may be defined particularly by means of the e system: the blood of eAg-positive carriers is said to be infectious, anti e-positive blood is said to be non-infectious [7,8].

These aspects of the e system are important in differential diagnosis and it is therefore necessary to determine eAg in patients as regular dialysis treatment, and after renal transplantation.

Patients and Methods

The sera of 22 patients on maintenance dialysis (n = 12 male, 10 female patients, mean age 49 years — range 16 to 65) with persistence of HBs antigen, and of 51 unselected HBsAg-positive patients (n = 29 male, 22 female patients, mean age 45 years — range 16 to 66) without renal failure were investigated. HBs antigen was determined by radioimmunoassay. The test for e antigen was performed by a standard immunodiffusion technique [9]. The details of the technique are shown in Figure 1. Laboratory standards (e antibody) were tested for a reaction of identity with standard e antibody kindly provided by

![Image](Methods.png)

**Figure 1. Principles of e antigen detection by a standard immunodiffusion technique**

Dr L Bouvier, Yale University. Statistical analysis was done by χ² and the Student t test.

Liver biopsy was performed on most of the patients by the Menghini technique.
Results

In 7 of the 22 patients on maintenance haemodialysis with persistent HBs-antigenaemia the e antigen could be detected. All individuals were still HBsAg- and eAg-positive when retested more than one year later. The frequency of the eAg in the group of 51 unselected HBsAg-positive patients without any renal failure was significantly lower (χ² = 6.97, P < 0.01). Only 4 out of 51 were positive for the e antigen (Table I).

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<th>e antigen</th>
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<th>χ² = 6.97</th>
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<td>Patients on maintenance dialysis</td>
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Another numerous group of HBsAg-negative subjects undergoing chronic haemodialysis always had a negative e antigen. In the serum of two dialysis patients eliminating the HBs antigen in course of time, eAg was never detected. The mean SGOT and SGPT levels of the HBsAg-positive dialysis patients were only moderately elevated, but showed large fluctuations. There was no difference between the eAg-positive and the eAg-negative group in this respect. In most of the dialysis patients (14 out of 22) the microscopic findings of the

Figure 2. Typical histological findings in liver biopsies of dialysis patients with HBsAg-persistence: 'Minimal changes' with focal proliferation of Kupffer's cells and necrosis of single liver cells (haematoxylin and eosin stain, X 250 – reduced for publication)
liver only revealed 'minimal changes'. Figure 2 demonstrates typical 'minimal changes' with focal proliferation of Kupffer cells and necrosis of single liver cells. Other histological diagnoses were: chronic persistent hepatitis (four patients) and liver cirrhosis (four patients). In all cases opalescent hepatic cells ('ground glass' cells) which are typical of HBsAg-carriers were a striking feature.

Three eAg-positive patients with 'minimal changes' in the liver histology received a renal transplant and were treated with prednisone and azathioprine.

![Graph showing SGOT and SGPT levels for different groups of patients](image)

Figure 3. Average values of sequential SGOT and SGPT determinations over a period of at least 12 months (n\textsubscript{v} = number of values)

Several months after transplantation these three patients displayed an appreciably increased eAg-yield by immunodiffusion. Histology now revealed chronic persistent hepatitis, and the mean SGPT levels were significantly higher than those before transplantation (Figure 3).

Discussion

Previous clinical studies suggested that eAg was associated with the development of chronic hepatitis in HBsAg carriers [5,10–12]. In our HBsAg-positive dialysis patients we found a large proportion with chronic persistent hepatitis and liver cirrhosis: a direct correlation with the e antigen could not be verified,
but it was confirmed that the persistent carriers of HBsAg in this patient group have a high frequency of e antigen. The HBsAg-positive dialysis patients are known to be remarkably infectious. This fact might indicate a correlation between the present of e antigen and infectivity.

The immune response of chronic stable haemodialysis patients is better than that of uraemic patients who are not efficiently treated. But the cell-mediated immunity of the former is markedly diminished in comparison with healthy subjects [13,14]. They have a qualitative defect of T lymphocytes [15]. It is likely that the course of hepatitis B is influenced by the differing degrees of immune response. The subacute anicteric but prolonged course of hepatitis in patients undergoing chronic haemodialysis is presumably a result of their immunodeficiency [16]. The immunodeficiency may also support the synthesis of eAg. This hypothesis is supported by the finding of an increased eAg-yield after renal transplantation. The synthesis of eAg after transplantation is possibly enhanced by the additional immunosuppressive therapy. Our three transplanted patients only showed slight transplant rejections. After more than one year the function of their transplants is very good. The presence of eAg can be interpreted as an indicator of a low immune response, and perhaps it can be considered as favourable for graft tolerance. On the other hand one has to consider the progress of 'minimal changes' to chronic persistent hepatitis under the immunosuppressive treatment, which may shorten patient survival.

References

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OPEN DISCUSSION

BÜHLER (Kaiserslautern) As you have shown the e antigen has a certain prog-
nostic value in the later stage of hepatitis B. Why does it not have a prognostic value in the earlier stage of the infection too? Secondly, to which part of the Dane Particle does the eAg belong. Is it a part of the surface antigen or is it a part of the core antigen or is it a product of the immune response?

BRISAM  About the prognostic value: several studies have suggested that the e antigen at the acute stage of hepatitis has a prognostic value, but authors from Japan and Germany have disqualified e antigen in the acute phase of the disease as an indicator of the prognosis because of the quick disappearance of the e antigen and because of the low sensitivity of the method of immune diffusion to detect the e antigen. I think that this problem will be solved when the methods to detect the e antigen improve so we can show more e antigen positive patients in the acute stage of the disease. It can be said that the persistence of e antigen may be an indicator which shows that the disease will become chronic. About the relationship to the Dane Particle: in the eAg positive sera it was found that the concentration of the Dane Particle was very much higher in comparison with those which were e antigen negative.

PARSONS (London) We have seen the disappearance of e antigen and Dane Particles in three patients after a long period of transplantation, in excess of two years. Have you followed your patients for that long and seen the disappearance of the antigen with low doses of steroids and immunosuppression?

BRISAM  Well I cannot definitely give an answer to this question. Our patients after renal transplantation have an increased eAg yield, probably because of an impairment of their cell-mediated immunity. We have not observed disappearance of eAg after renal transplantation.