

Chronic hepatitis type B in childhood: longitudinal study of 35 cases

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SUMMARY Clinical, virological, and histological features of hepatitis B virus infection have been examined in 35 children, aged 1 to 11 years, known to be hepatitis B surface antigen (HBsAg) carriers for at least six months when entering the study. Only 10 patients had a history of acute unresolved hepatitis: in the remaining cases the detection of HBsAg had been an occasional finding. Although 77% of the patients were asymptomatic, all had evidence of hepatic involvement and liver history showed the features of chronic persistent hepatitis in 18 cases and of chronic active hepatitis in 16 cases, with associated cirrhosis in two of them. One patient had only minimal histological changes. A high percentage of children with both chronic persistent and chronic active hepatitis had evidence of active virus replication throughout the observation period. During the follow-up study of one to eight years (mean 3.1 ± 1.7 years), transaminase levels became consistently normal in five patients with chronic persistent hepatitis, and inflammatory infiltrates disappeared in three of them. However, only one of these children cleared HBsAg from serum. Eleven of 16 patients with chronic active hepatitis received immunosuppressive treatment but only one of them achieved a complete and protracted remission, although active viral replication persisted. On the other hand, two of five untreated patients reached complete remission after two and three years of follow-up respectively and one of them cleared HBsAg three years later. These results would suggest the possibility of a spontaneous complete remission of HBsAg positive chronic active hepatitis in children but also raise doubts about the usefulness of immunosuppressive therapy in such patients.

Clinical features of hepatitis B virus (HBV) infection in childhood have been reviewed in recent years¹⁻⁴ and some controversial data have emerged. Dupuy *et al.*⁵ reported a marked incidence of overt disease associated with HBV infection in neonates and infants and conversely a small number of patients who became chronic hepatitis B surface antigen (HBsAg) carriers. On the other hand, Merrill *et al.*,⁶ Schweitzer,¹ Okada *et al.*,⁷ Stevens *et al.*,⁸ and Skinhøj⁹ have observed a number of children born to HBsAg carrier mothers who became chronic HBsAg carriers. In these patients associated liver disease was often asymptomatic and anicteric with only a slight tendency to progression. The majority of these reports, however, included only small num-

bers of chronic HBsAg carrier children or lacked a prospective long-term follow-up study.

In this report we have examined the clinical, biochemical, and histological features of chronic HBV infection in 35 children. Most of them were asymptomatic, although with biochemical and histological evidence of liver disease and with serological features of active virus replication. The long-term outcome of the disease and the effects of immunosuppressive treatment have been also evaluated, as 24 children have been followed up for more than two years.

Methods

PATIENTS

Thirty-five children, known to be HBsAg positive in serum for at least six months, were referred to the Department of Infectious Diseases, Hospital of Padova, between 1972 and 1979, and

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were included in this study. They were between 1 and 11 years old with an average age of 4.8 ± 2.7 (mean \pm SD) years. Nineteen were female and 16 male. In 10 patients (Table 1) an episode of acute hepatitis B, anicteric in seven cases, was recorded six to 12 months before entering this study. While vomiting, anorexia, and abdominal pain were the major complaints during the acute phase, two cases were associated with papular acrodermatitis, which is now considered to be a typical extrahepatic manifestation of HBV infection in childhood.^{10, 11} In all 10 patients the progression to chronic hepatitis was characterised by persistence of increased transaminase levels after the disappearance of jaundice and, in most cases, even of symptoms.

The remaining 25 patients were occasionally found to be HBsAg positive when tested either during a general check-up preceding tonsillectomy or adenoidectomy or after the demonstration of HBV infection in a household contact. The possible sources of infection in our patients are shown in Table 1. Only one of the patients had a history of blood transfusion received soon after birth, although the HBsAg carrier state was discovered 12 months later, when a household contact developed acute hepatitis B. Evidence of past or ongoing HBV infection could often be documented among the family contacts of our patients. Indeed, when the children entered the study, 27% of contacts were found to be HBsAg positive and 24%, including four mothers, remained positive during a follow-up of at least one year and could be therefore considered as chronic HBsAg carriers. In addition another 48% of family contacts of our patients (including 57% of mothers) showed evidence of previous HBV infection as indicated by the finding of antibody to hepatitis B core antigen (anti-HBc) and/or of antibody to HBsAg in serum.

In all 35 children, clinical, biochemical, and virological features were studied at presentation and during a follow-up period of one to eight

years (mean 3.1 ± 1.7 years).

As all patients had hepatomegaly and biochemical evidence of liver disease, they all underwent liver biopsy, performed by the Menghini technique, after their parents had given informed consent. In 24 patients one or more subsequent biopsies were performed during the follow-up study. The histological diagnosis in the first and in the follow-up biopsies was assessed according to De Groote *et al.*¹² and to Scheuer.¹³

TECHNIQUES

HBsAg and anti-HBc were tested by commercial radioimmunoassay kits (Abbott). Hepatitis B e antigen (HBeAg) and antibody to HBeAg (anti-HBe) were tested on unconcentrated sera by immunodiffusion using reference reagents known to contain the HBe₁Ag and HBe₂Ag specificities. Specific DNA polymerase activity was measured by the method of Kaplan *et al.*¹⁴ Smooth muscle antibodies (SMA), antinuclear antibodies (ANA), and mitochondrial antibodies (AMA) were investigated by indirect immunofluorescence.

Results

EVIDENCE OF LIVER DISEASE IN 35 CHILDREN

When they entered the study all patients were anicteric and only eight were symptomatic, complaining of mild asthenia, anorexia, or vomiting. However, hepatomegaly was detected in all children and was associated with splenomegaly in 10 of them. All patients showed increased transaminase levels, from 1.5 up to 10 times the upper normal limit, while bilirubin levels were within the normal range. Hypergammaglobulinaemia (gammaglobulin exceeding 12 g/l (1.2 g/100 ml) in children aged 1 to 2 years and 15 g/l (1.5 g/100 ml) in children more than 2 years old) was found in 12 cases, while albumin levels and prothrombin time were always normal in all patients. SMA were transiently detected in three cases, while ANA and AMA were never

Table 1 Age distribution of 35 chronic HBsAg carrier children in relation to clinical onset of disease and possible sources of infection

Age (yr)	No.	Acute onset		Possible source of infection			HBV infection in mothers	
		(No)	(%)	Transfusion	Vertical*	Unknown	Past	Ongoing
0-1	4	1	25	1	3	0	1	2
1-5	21	8	38	0	0	21	13	2
5-11	10	1	10	0	0	10	6	0

*Vertical transmission was reasonably suspected in three patients who were found to be HBsAg carriers within the first year of life; two of their mothers were chronic HBsAg carriers and a third one had acute hepatitis B at the time of delivery.

Table 2 Clinical and serological features of chronic persistent and chronic active hepatitis in 34 HBsAg positive children

	CPH (18 cases)		P*	CAH (16 cases)	
	(No.)	(%)		(No.)	(%)
Acute onset	3	16.6	NS	7	43.7
Symptoms	2	11.1	NS	6	37.5
Hepatomegaly	18	100		16	100
Splenomegaly	6	33.3	NS	4	25
Mean AST (normal < 40IU/l)	81.7 ± 43.2		< 0.01	178.5 ± 96.4	
HBsAg	10	55.5	NS	12	75.0
Anti-HBe	0			2	12.5
Neither	8	44.4	NS	2	12.5
DNA polymerase	11	61.1	NS	12	75.0

*P was calculated by the chi-square test and the Cochran-Cox test.

found. IgM rheumatoid factors were identified by a latex test in six patients.

HBV MARKERS IN 35 CHILDREN

The presence of HBsAg in serum was confirmed by RIA in all cases. All 35 children also had anti-HBc in serum with titres ranging from 4×10^{-3} to 10^{-5} . Twenty-two cases were positive for HBeAg, while three had anti-HBe and 10 were HBeAg/anti-HBe negative. Specific DNA polymerase activity was detected in 23 patients.

LIVER HISTOLOGY

Liver biopsies showed the features of chronic persistent hepatitis in 18 children and of chronic active hepatitis in 16 cases, with associated cirrhosis in two of them. A 13 months old, asymptomatic child with hepatomegaly and slightly increased transaminases, who was anti-HBe positive in serum, had only minimal histological changes. During a 15 months' follow-up this patient, who had been presumably infected by his HBsAg carrier mother, remained HBsAg and anti-HBe positive, with slightly increased transaminases. Clinical and serological findings in chronic persistent and in chronic active hepatitis patients are compared in Table 2. An acute onset and a symptomatic presentation were more common in chronic active than in chronic persistent hepatitis and the prevalence of HBeAg and of DNA polymerase

activity was higher in the former group of patients but the differences were not statistically significant. On the other hand transaminase levels were significantly higher in chronic active than in chronic persistent hepatitis cases.

FOLLOW-UP OF CHRONIC PERSISTENT HEPATITIS (Table 3)

None of the 18 patients with chronic persistent hepatitis who could be followed up for a period of one to eight years received immunosuppressive treatment. The clinical course of the disease was characterised by complete well-being. Transaminase levels fluctuated in time in 13 cases including all HBeAg positive children, while they became consistently normal after one to three years in five HBeAg/anti-HBe negative cases. Two of the latter patients became anti-HBe positive at the time when liver enzymes had become normal and one of them cleared HBsAg eight months later. None of the HBeAg positive patients seroconverted to anti-HBe. Subsequent liver biopsies were obtained 15 months to seven years after entering the study in 11 cases. In the five children with sustained normalisation of transaminases, liver histology performed at the time of normalisation showed the features of chronic persistent hepatitis in two cases, minimal histological changes in two other cases, and portal fibrosis in the female patient who later on cleared HBsAg. Among patients with fluctuating transaminases liver histology remained unchanged in four of six cases and progressed to chronic active hepatitis in the remaining two cases, after two and three years respectively.

FOLLOW-UP OF CHRONIC ACTIVE HEPATITIS PATIENTS

Eleven of 16 patients with chronic active hepatitis were started on immunosuppressive treatment with prednisone (2 mg/kg/day) or with a combination of prednisone (1 mg/kg/day) and azathioprine (2 mg/kg/day). When stable improvement of biochemical parameters was achieved—that is, transaminase levels lower than twice the upper

Table 3 Follow-up study (one to eight years) of 18 children with HBsAg positive chronic persistent hepatitis

Transaminases	HBe/anti-HBe system		Subsequent liver histology (11 cases)
	At presentation	Last detection	
Sustained normalisation (5 cases)	HBsAg	0	{ CPH: 2 minimal changes: 2 portal fibrosis: 1
	Anti-HBe	2	
	Neither	3	
Fluctuating (13 cases)	HBsAg	10	{ CPH: 4 CAH: 2 not available: 7
	Anti-HBe	0	
	Neither	3	

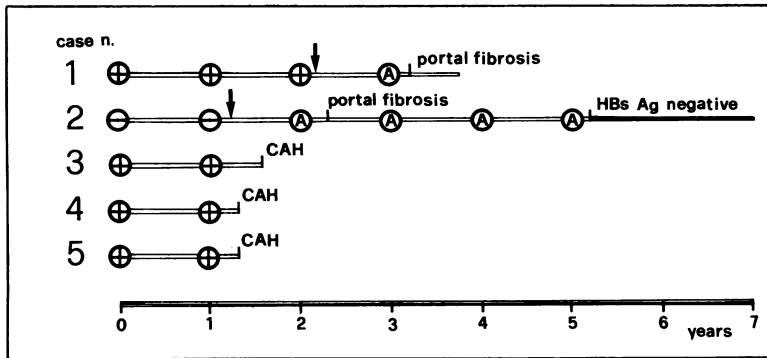


Fig. 1 Follow-up study of five untreated patients with HBsAg positive chronic active hepatitis.
 ⊕ HBeAg positive.
 ⊗ Anti-HBe positive.
 ⊖ HBeAg/anti-HBe negative.
 ↓ Sustained normalisation of transaminases.

normal limit—drug intake was gradually reduced and maintenance therapy (prednisone 0.5–1 mg/kg/alternate day) was given for periods of one to four years. When relapse occurred after withdrawal of therapy, treatment was started again at full dosage. Side-effects of prolonged immunosuppressive therapy were observed in six cases who had a transient cushingoid appearance. Five of the 10 asymptomatic patients did not receive immunosuppressive treatment because their parents were unwilling. Asthenia and anorexia were the only symptoms transiently recorded in patients with chronic active hepatitis during the follow-up period of one to eight years. None developed liver failure or clinical evidence of portal hypertension.

Among the untreated patients (Fig. 1) transaminases fluctuated in time in three cases, all HBeAg positive when entering the study, and became normal, after 14 months and two years respectively, in two other cases: one of the latter (case 1) who was HBeAg positive at presentation, seroconverted to anti-HBe, while the other (case 2), who was initially HBeAg/anti-HBe negative, became anti-HBe positive at the time when liver enzymes had become normal and cleared HBsAg from serum about three years later. In both these cases chronic active hepatitis was of moderate activity at first biopsy. Subsequent liver histology performed after biochemical remission and after the appearance of anti-HBe was characterised by substantial changes in parenchymal and mesenchymal lesions. These were represented by a striking reduction or complete disappearance of intra-lobular inflammation and necrosis, leaving a diffuse hypertrophy of sinusoidal lining cells. The portal tracts appeared to be well delineated from the parenchyma, with minimal cellular inflammation, but prevalent fibrosis. Features of

nodular regeneration were absent. No significant changes of liver histology were observed in the untreated patients with fluctuating transaminases.

The behaviour of transaminases and of liver histology in the 11 treated patients is shown in Fig. 2. A sustained normalisation of liver enzymes was obtained after nine to 17 months of treatment in three cases who remained HBeAg positive throughout the observation period. Liver histology at this time was unchanged in one case (case 2) and showed the features of chronic persistent hepatitis in the remaining two children (cases 1 and 3). One of the latter, however, relapsed one year after withdrawal of therapy and liver histology is now again consistent with chronic active hepatitis. In the other eight patients transaminases showed periodical fluctuations despite continued maintenance therapy and liver histology remained unchanged, after one to seven years, in all five children who underwent a second liver biopsy.

Discussion

Of the 35 children with HBsAg positive chronic hepatitis included in this study, only 10 had a history of acute unresolved hepatitis. In all the other cases the detection of HBsAg in serum had been an occasional finding, thus suggesting that, as HBV infection is endemic in Italy, a number of chronic carrier children could remain undetected.

Only one of our children had received blood transfusions and maternoneonatal transmission could be reasonably suspected in three other cases. Thus the source of infection remained undefined in most children, although it can be suggested that, as 57% of mothers had evidence of past HBV infection, vertical transmission of HBV could have occurred in a larger number of

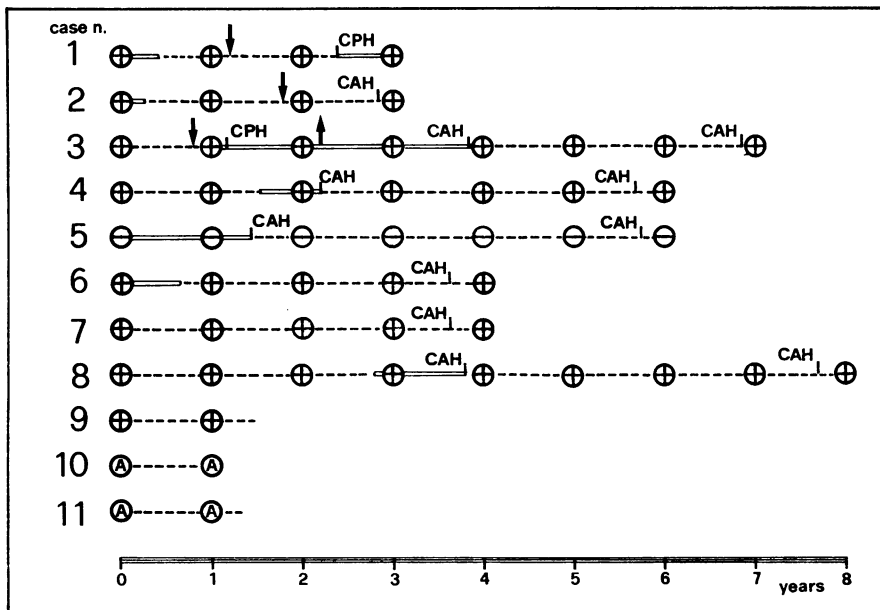


Fig. 2 Follow-up study of 11 treated patients with HBsAg positive chronic active hepatitis.
 ⊕ HBeAg positive.
 ⊕ Anti-HBe positive.
 ⊖ HBeAg/anti-HBe negative.
 ↓ Sustained normalisation of transaminases.
 ↑ Biochemical relapse.
 --- Immunosuppressive therapy.

cases. On the other hand serological evidence of past HBV infection was frequent among the family contacts of our children and the prevalence of chronic HBsAg carriers was relevant as compared with the mean incidence (2.5%) of the chronic HBsAg carrier state among asymptomatic blood donors in Italy.¹⁵ These findings, as suggested in a previous report,¹⁶ could reflect the importance of person to person transmission of infection. The role of chronic carrier children in the epidemiology of HBV infection in our region could be of relevance when considering that, although they were either asymptomatic or had very few symptoms, about two-thirds of our patients had evidence of active virus replication. Indeed, specific DNA polymerase activity was detected in 67% of cases and HBeAg was found in the serum of most children at presentation and throughout the course of the disease; this suggests that these patients remain infectious for long periods of time.

The high frequency of HBeAg in this series of chronic HBsAg carriers is not surprising, as a prevalence of HBeAg positivity has been demonstrated in young as compared with old chronic carriers.¹⁷ It also seems remarkable that both patients with features of chronic active hepatitis and associated cirrhosis had anti-HBe at presentation. These data are in agreement with the frequent occurrence of seroconversion from HBeAg to anti-HBe observed in our adult

patients with HBsAg positive chronic active hepatitis when the disease had progressed to liver cirrhosis.¹⁸

Transaminase levels at presentation were significantly higher and DNA polymerase activity was more frequently detected in chronic active than in chronic persistent hepatitis cases; however, clinical, biochemical, and virological features of the infection overlapped in the two groups of children.

Independently of the histological lesions all patients have been clinically well or with only minor complaints during the follow-up study and no growth retardation was observed, although transaminase levels became permanently normal in only 10 patients and DNA polymerase activity disappeared in only one of the initially positive cases. In spite of the often severe biochemical and virological features, clinical evidence of liver failure did not develop in any of our children.

A mild clinical course has been commonly described in children with HBsAg positive chronic persistent hepatitis, and it is generally considered as a benign disorder.^{2,4} Indeed, so far, in five of our 18 children with chronic persistent hepatitis transaminases have become normal and liver histology confirmed the disappearance of inflammatory infiltrates in three of these five cases. All five patients were HBeAg negative, thus suggesting that a favourable course of the disease is most common among patients

without evidence of active virus replication. The long-term outcome of such patients remains, however, unpredictable, as only one of five cases has cleared HBsAg from serum. Otherwise in two HBeAg positive patients with fluctuating transaminases a progression has been observed from chronic persistent to chronic active hepatitis.

Our follow-up study of children with chronic active hepatitis has shown evidence of the possibility of a spontaneous histological remission with or without final clearance of HBsAg from serum. Indeed, although bioptic sampling errors cannot be excluded, this favourable outcome has been observed in two of five asymptomatic children who did not receive immunosuppressive treatment. On the other hand, only one of 11 treated patients, including also five asymptomatic cases, has so far reached histological remission and no one seroconverted from HBeAg to anti-HBe. In one other patient histological remission was not maintained after withdrawal of therapy and another child achieved only biochemical remission. Therefore these preliminary results of immunosuppressive treatment have not been encouraging in our patients, although no definite conclusions can be drawn because of the small number of patients and the lack of a randomised control group.

Our results are at variance with the findings of Dupuy *et al.*⁵ Indeed, these authors reported that 12 out of 12 children with chronic active hepatitis progressed to cirrhosis, and in eight of them the disease became inactive under immunosuppressive treatment, with clearance of HBsAg from serum in six cases. These discrepancies might be due to a different selection of patients; however, they further indicate the need for a controlled trial of immunosuppressive therapy in HBsAg positive chronic active hepatitis of children, similar to that carried out in HBsAg negative chronic active hepatitis,¹⁹ to evaluate the real benefit of this treatment and the opportunity of new therapeutic approaches.

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