

Relationship between Viral Antigen, HBV DNA Level and Hepatic Fibrosis in HBV Infected Patients during the Natural History of Chronic Hepatitis B

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ABSTRACT

Background: Relationship between hepatitis B surface antigen (HBsAg) level, HBV DNA level and hepatic fibrosis during the natural course of hepatitis B virus (HBV) infection are still unknown. Our objective is to correlate HBsAg level, HBV DNA and liver fibrosis in patients with different phases of chronic hepatitis B infection.

Subjects and Methods: One hundred seventy six patients with chronic hepatitis B without previous treatment were recruited. Clinical, demographic, and laboratory data were collected. Liver fibrosis was assessed by transient elastography (Fibroscan). We determined quantitative HBsAg titer, HBV DNA level. Patients were classified into each phase of diseases and analyzed the correlation.

Results: One hundred thirteen males and 63 females at median age of 43 years were studied. Patients were classified into HBeAg positive (n=29), HBeAg negative with low replicative (n = 69) and HBeAg negative hepatitis (n = 78). In patients with HBeAg positive, HBsAg levels were significantly higher than those of patients with HBeAg negative (mean 19797 vs. 4282 IU/mL, $p < 0.01$). No difference in HBsAg level between low replicative and HBeAg negative hepatitis group (mean 3689 vs. 4806 IU/mL, $p = 0.32$). HBsAg levels were significantly correlated with HBV DNA level in HBeAg positive and HBeAg negative with hepatitis groups ($r = 0.698$, $p < 0.01$ and $r = 0.287$, $p = 0.01$, respectively). We neither found any correlation between liver fibrosis measured by Fibroscan and HBsAg level nor HBV DNA level.

Conclusions: HBsAg levels were significantly higher in patients with HBeAg positive than those of patients with HBeAg negative. Some correlations were observed between HBsAg levels and HBV DNA level. No cross-sectional relationship between liver fibrosis measured by Fibroscan and HBsAg level or HBV DNA level.

Key words : HBsAg titer, chronic hepatitis B, transient elastography, Fibroscan, liver stiffness, liver fibrosis, HBV viral load

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BACKGROUND

Chronic hepatitis B infection is major cause of chronic liver disease in many countries and found to be one of major causes of cirrhosis worldwide⁽¹⁾. In Thailand, high prevalence of chronic hepatitis B was observed and considered to be a public-health burden⁽²⁾. Hepatitis B virus (HBV) causes a wide range of clinical consequences, from acute and chronic hepatitis to cirrhosis and hepatocellular carcinoma⁽³⁾. HBsAg was used as diagnostic tool in HBV infection and serologic conversion is the ultimate laboratory marker of cure of the disease. HBsAg was one of a product of viral replication and have been proposed as surrogates for infected liver cell⁽⁴⁾. Recently, standard quantitative serology for hepatitis B surface antigen (HBsAg) had been developed and used widely and comparable⁽⁵⁾. Assessment of liver fibrosis with non-invasive measurement, transient elastography demonstrated reliable results in chronic hepatitis B⁽⁶⁾. Each phase of hepatitis B infection stimulates distinct viral kinetics and host immune responses and also different in level of HBsAg^(7,8). Relationship between hepatitis B surface antigen (HBsAg) level, HBV DNA level and hepatic fibrosis during the natural course of hepatitis B virus (HBV) infection are still unknown. This study aims to correlate HBsAg level, HBV DNA and liver fibrosis in patients with different phases of chronic hepatitis B infection.

PATIENTS AND METHODS

Patients

One hundred seventy six patients with chronic hepatitis B without previous treatment were recruited. Clinical, demographic, and laboratory data were collected. Patients with evidence of HCV or HIV co-infection, alcoholic liver disease, chronic liver disease from other causes and acute viral hepatitis B were excluded from the study. After follow up period, patients were classified into each phase of diseases (HBeAg positive, HBeAg negative with low replicative (LR), HBeAg negative hepatitis (ENH)) according to EASL Clinical Practice Guidelines⁽⁹⁾. We determined ALT level, HBV DNA level to classified phase of disease.

Methods

After patients were enrolled and consented, blood component was test for liver biochemistry and serologic data. Liver fibrosis was assessed by transient

elastography (Fibroscan). We determined quantitative HBsAg titer by ELISA method (Architek, Abbott Laboratories, North Chicago, Ill) and HBV DNA level was measure by realtime PCR.

Statistical analysis

Continuous variables were compared between groups using unpaired *t*-test and one-way ANOVA. Categorical variables were compared between groups using Chi-square/Fisher's exact test. Pearson's correlation coefficient was used to describe the correlation between two continuous, normally distributed variables. A Spearman's correlation was used where variables were not normally distributed. All statistical analyses were performed using SPSS version 16.

RESULTS

One hundred thirteen males and 63 females at median age of 43 years (range 18-79 years) were studied. Patients were classified into HBeAg positive ($n = 29$), HBeAg negative with low replicative ($n = 69$) and HBeAg negative hepatitis ($n = 78$). Demographic and biochemical data of patients are shown in Table 1. The median age and male gender of patient were 29, 48, 42.5 years and 68%, 49% and 76% in HBeAg positive, LR and ENH group respectively. ALT and DNA level were different in each group due to phase of disease. Degree of necroinflammation as described by Knodell and significant hepatic fibrosis described in the Metavir score⁽¹⁰⁾ didn't shown different in HBeAg positive and ENH group. Mean HBsAg titer was statistical significant higher in HBeAg positive group compare with HBeAg negative (19797 and 4282 IU/mL, $p < 0.01$). In HBeAg negative patient no statistical different observed in LR and ENH group (3689 and 4806 IU/mL, $p = 0.32$).

Degree of liver fibrosis measure by Fibroscan was significant lower in LR phase compare with HBeAg positive and ENH (5.28, 7.04 and 7.43 kPa, respectively, $p < 0.01$) as shown in Figure 1.

HBsAg levels were significantly correlated with HBV DNA level in HBeAg positive and ENH groups ($r = 0.698$, $p < 0.01$ and $r = 0.287$, $p = 0.01$ respectively). No significant correlation found in LR group ($r = 0.092$, $p = 0.45$) as shown in Figure 2. In our study, degree of liver fibrosis measure by Fibroscan were also correlated with ALT ($r = 0.515$, 0.398 , 0.277 and 0.426 with $p < 0.01$, 0.033 , 0.028 and < 0.01) in all

Table 1. Baseline characteristic of patients classified with phase of disease.

	HBeAg positive (n = 29)	Low-replicative (LR) (n = 69)	HBeAg negative hepatitis (ENH) (n = 78)	p-value
Median age (yrs)	29	48	42.5	<0.01
Male gender (%)	20 (68)	34 (49)	59 (76)	<0.01
ALT (U/L)	96.38	23.84	61.05	<0.01
HBV DNA (IU/mL)	84,068,000	559	5,823,600	<0.01
Liver stiffness (kPa)	7.04	5.28	7.43	<0.01
HBsAg titer (IU/mL)	19797	3689	4806	<0.01
Significant Inflammation (n = 74) (%)	15/26 (58)	-	21/48 (43.75)	0.252
Significant fibrosis (n = 75) (%)	9/25 (36)	-	26/50 (52)	0.19

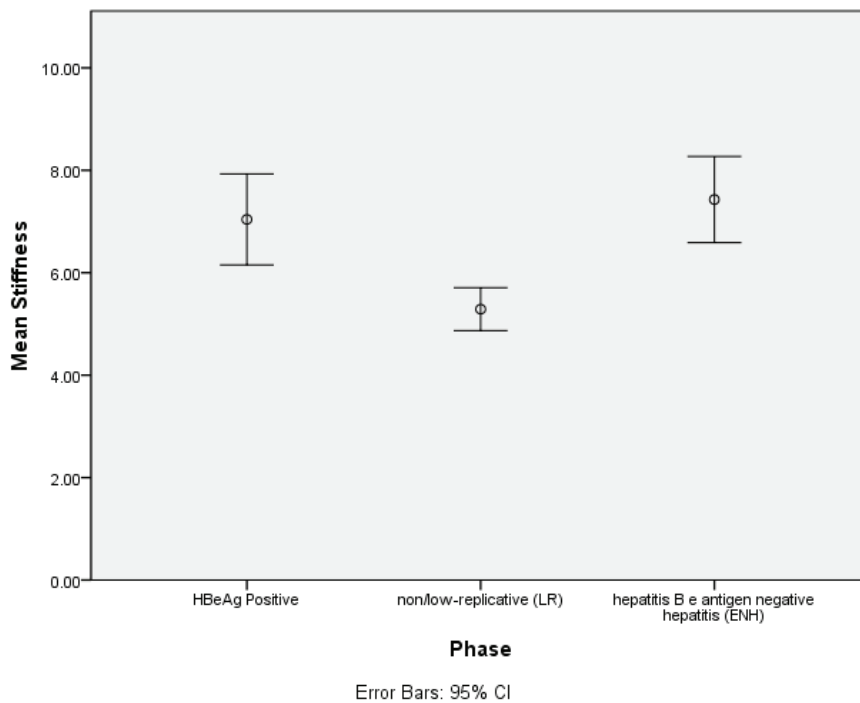


Figure 1. Degree of liver fibrosis measure by Fibrosan (kPa) in each phase of disease.

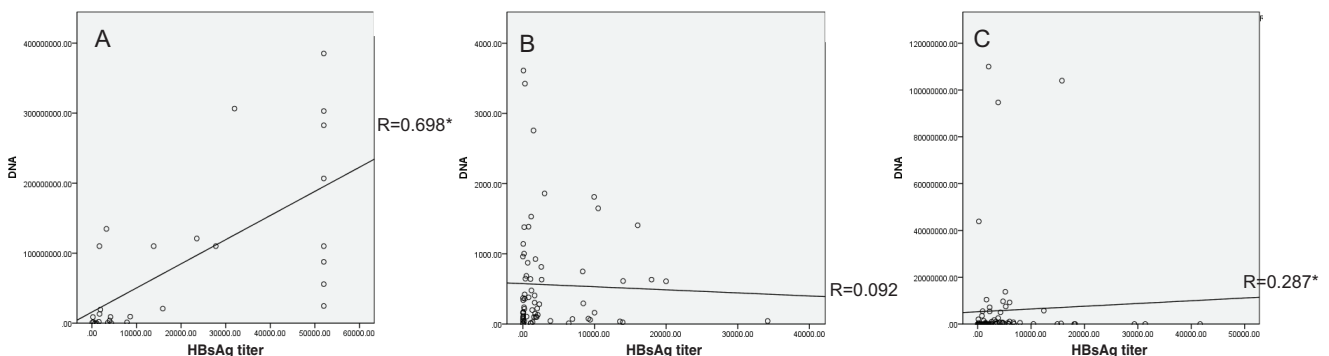


Figure 2. Correlation between HBsAg titer and DNA level in groups of patients. (A: HBeAg positive, B: Low replicative (LR), C: HBeAg negative hepatitis (ENH)) (*statistical significant).

groups, HBeAg positive, LR and ENH group respectively as shown in Figure 3. Significant correlation between degree of liver fibrosis measure by Fibrosan and patient age was found in only LR group ($r = 0.333$, $p < 0.01$).

We neither found any correlation between liver fibrosis measured by Fibrosan and HBsAg level nor HBV DNA level. Correlation between HBsAg titer and liver stiffness in HBeAg positive, LR and ENH group were $r = 0.174$, -0.101 and -0.047 with $p = 0.38$, 0.41 , 0.68 respectively. Correlation between HBV DNA level and liver stiffness in HBeAg positive, LR and ENH group was $r = 0.025$, -0.077 and 0.194 with

$p = 0.896$, 0.527 and 0.089 as shown in Figure 4.

DISCUSSION

HBsAg has significant role in virologic point of view and also in clinical point, more knowledge in HBsAg enable us to deepen our insights, and more clinical application. Some studies had showed benefit of HBsAg titer as predictor PEG-Interferon treatment response^(11,12). Currently transient elastography (Fibrosan) was used as noninvasive tool to predict degree of liver fibrosis, with strong correlation with histopathology in chronic hepatitis B⁽¹³⁾. Multiple causes were related with progression and degree of

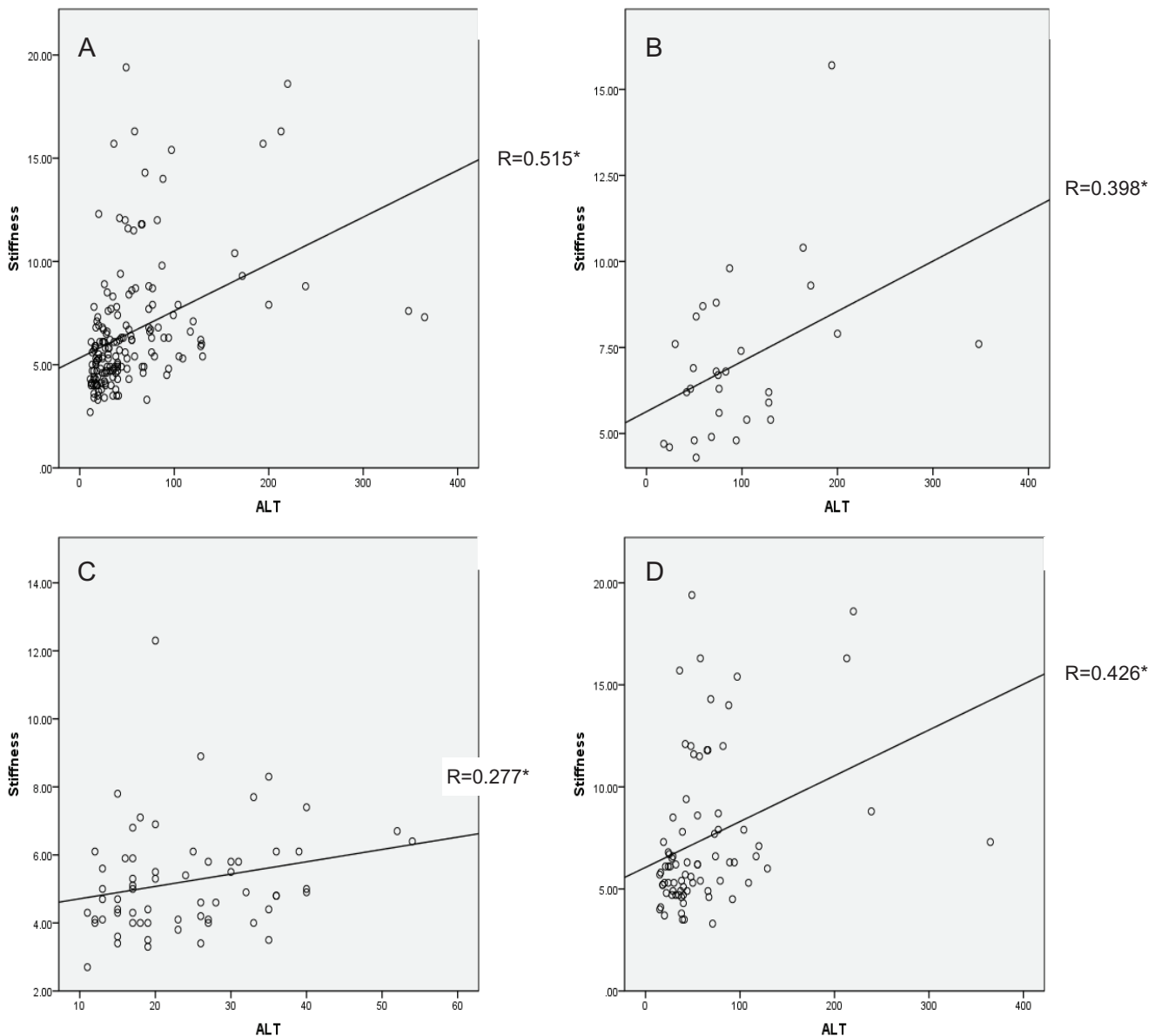


Figure 3. Correlation between degree of liver fibrosis measure by Fibrosan and ALT. (A: All groups, B: HBeAg positive, C: Low replicative, D: HBeAg negative hepatitis) (*statistical significant).

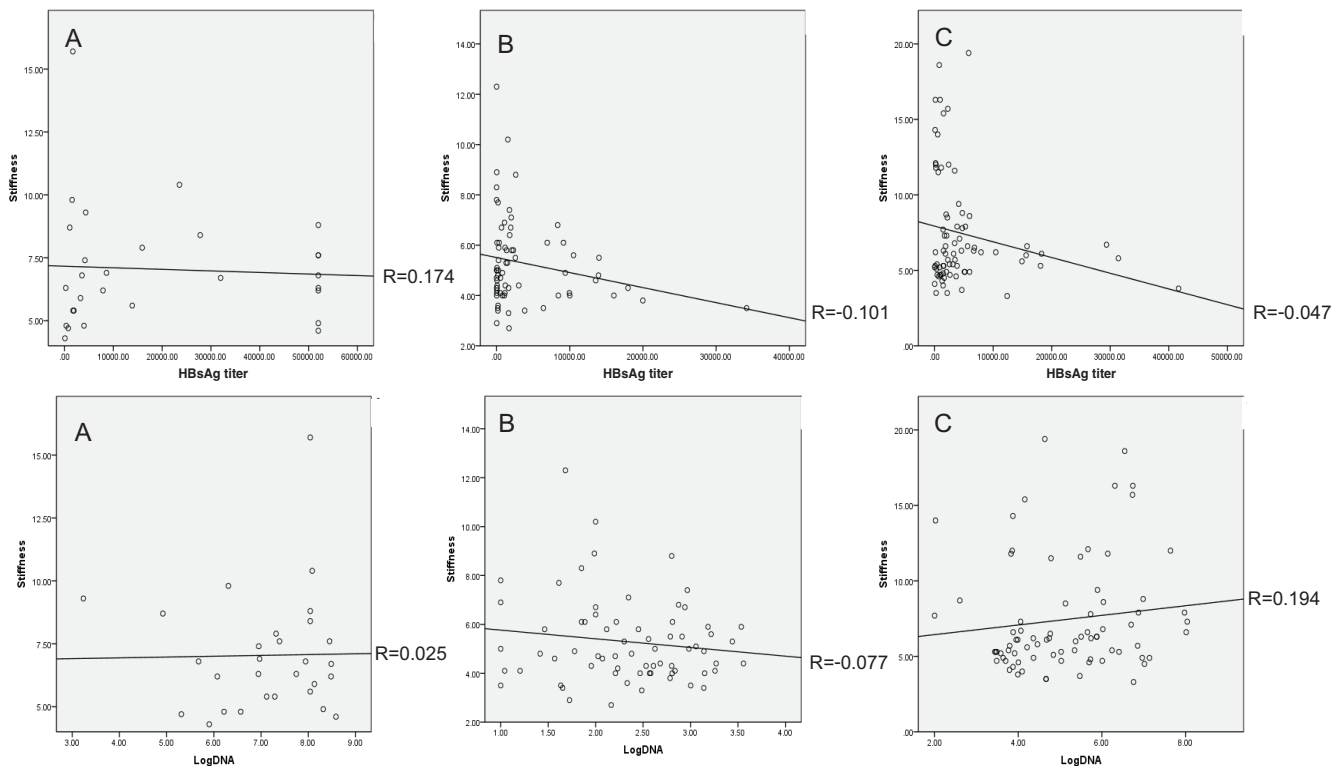


Figure 4. Correlation between HBsAg titer and liver stiffness, DNA level and liver stiffness in groups of patients (A: HBeAg positive, B: Low replicative, C: HBeAg negative hepatitis).

hepatic fibrosis in chronic hepatitis B such as level of DNA, HBV genotype, age, co-infection with HCV or HIV, alcohol consumption^(14,15). This study tried to find out any correlation between HBsAg level and degree of liver fibrosis. Result of study revealed that HBsAg levels were significantly higher in patients with HBeAg positive than those of patients with HBeAg negative as previous few report. Some correlations were observed between HBsAg levels and HBV DNA level. We didn't found cross sectional relationship between liver fibrosis measured by Fibroskan and HBsAg level or HBV DNA Level. More prospective data are needed to confirm possibility of association between HBsAg level and liver fibrosis.

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