

Chronic hepatitis delta virus infection in Van region of eastern Turkey

Doğu Anadolu Bölgesi Van yöresinde kronik delta hepatiti infeksiyonu

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Background/aims: Hepatitis delta virus infection is an important cause of liver morbidity and mortality worldwide. In Eastern Turkey, hepatitis B virus infection is the major cause of chronic liver diseases. We aimed to research the role of hepatitis delta virus infection in chronic liver diseases related to hepatitis B virus infection in the Van region of Eastern Turkey. **Methods:** Serological markers of hepatitis B virus and hepatitis delta virus infection [HBsAg, HbeAg, Anti-HBe and Anti-hepatitis delta virus total (IgM+IgG)] were determined by ELISA test in patients with chronic hepatitis and cirrhosis. Serum hepatitis B virus DNA was determined by polymerase chain reaction (PCR) method in chronic hepatitis B patients. **Results:** Hepatitis delta virus infection was detected in 5% (7/138) of asymptomatic hepatitis B virus carriers, in 16% (24/148) of chronic hepatitis B patients and in 45% (34/75) of cirrhotic hepatitis B virus patients. hepatitis delta virus infection showed a three-fold increase in chronic hepatitis ($p<0.01$) and nine-fold increase in cirrhosis ($p<0.001$) compared to hepatitis delta virus carriers. Also, it was three times more frequent in cirrhosis ($p<0.001$) compared to chronic hepatitis. Chronic hepatitis delta virus infection was equally distributed between sexes in patients with chronic hepatitis B virus infection, whereas chronic hepatitis B virus infection alone was three times more frequent in males ($p<0.001$). Mean ages of hepatitis delta virus carriers, chronic hepatitis B and hepatitis delta virus cirrhosis patients were 30.7 ± 8 (14-65), 36 ± 13 (19-70) and 44 ± 16 (25-55), respectively. **Conclusions:** The higher prevalence of hepatitis delta virus infection in more severe form of hepatitis B virus infection suggests that hepatitis delta virus infection increases the severity of chronic hepatitis B virus infection in the Van region. hepatitis delta virus infection remains a second major cause of chronic liver diseases in Eastern Turkey in spite of its decreasing prevalence in Western countries and in Western Turkey.

Key words: HDV, Eastern Turkey

INTRODUCTION

Hepatitis delta virus (HDV) infection is an important cause of liver morbidity and mortality associated with hepatitis B virus (HBV) infection. Ful-

Amaç: Hepatit delta virus infeksiyonu tüm dünyada kronik karaciğer hastalığına bağlı ölümlerin önemli bir nedenidir. Doğu Anadolu Bölgesi'nde Hepatit B virus infeksiyonu kronik karaciğer hastalıklarının başlıca nedenidir. Bu çalışmada hepatitis B virus infeksiyonunun sık görüldüğü Van yöresinde hepatitis B virusuna bağlı kronik hepatit ve karaciğer sirozlu hastalarda hepatitis delta virus infeksiyonunun rolü ve epidemiyolojik özellikleri araştırılmıştır. **Yöntem:** Hepatitis B virus ve hepatitis delta virus infeksiyonunun serolojik göstergeleri (HBsAg, HbeAg, AntiHBe ve Anti hepatitis delta virus total "IgM+IgG") ELISA testi ile kronik hepatit ve karaciğer sirozlu hastalarda araştırılmıştır. Serum hepatitis B virus DNA PCR metodu ile kronik hepatit B'li hastalarda belirlenmiştir. **Bulgular:** Hepatitis delta virus infeksiyonu asemptomatik hepatitis B virus taşıyıcılarının %5'de (7/138), kronik hepatit B'li hastaların %16'da (24/148) ve hepatitis B virus sirozlu hastaların %45'de (34/75) teşhis edildi. Hepatitis delta virus infeksiyonu hepatitis delta virus taşıyıcılarına göre kronik hepatitte 3 kat ($p<0.01$), sirozda ise 9 kat yüksekti ($p<0.001$). Sirozda hepatitis delta virus infeksiyonu sıklığı kronik hepatite göre 3 kat yüksekti ($p<0.001$). Kronik hepatitis B virus infeksiyonu erkeklerde 3 kat daha sık görülürken ($p<0.001$), kronik hepatitis delta virus infeksiyonu kronik hepatitis B virus infeksiyonu olan hastalarda her iki cinste eşit düzeydeydi. hepatitis delta virus taşıyıcıları, kronik hepatit D ve hepatitis delta virus sirozlu hastaların yaş ortalamaları 30.7 ± 8 (14-65), 36 ± 13 (19-70) ve 44 ± 16 (25-55)'di. **Sonuç:** Van yöresinde hepatitis delta virus infeksiyonunun yüksek prevalansı hepatitis B virusuna bağlı kronik karaciğer hastalıklarının progresyonuna neden olmaktadır. hepatitis delta virus infeksiyonu Batı ülkeleri ve Türkiye'nin batı bölgelerinde azalmasına rağmen Doğu ve Güney Doğu Anadolu Bölgeleri'nde kronik karaciğer hastalıklarının 2. büyük nedeni olarak önemini korumaktadır.

Anahtar kelimeler: HDV, Doğu Anadolu Bölgesi

minant hepatitis, liver cirrhosis and hepatocellular carcinoma are the main fatal diseases complicating HDV infection. Fifteen million individuals

are infected with HDV throughout the world. In Turkey, HBV infection is still the most serious cause of chronic liver diseases. Liver morbidity and mortality are especially higher in Eastern and Southeastern Turkey than in other regions of the country. In this study, we aimed to investigate the role of HDV infection in chronic hepatitis and liver cirrhosis related to HBV infection in the Van region of Eastern Turkey.

MATERIALS AND METHODS

Serum samples of 138 asymptomatic HBV carriers (95 males, 43 females), 148 patients with chronic hepatitis B (110 males, 38 females) and 75 patients with HBV cirrhosis (55 males, 20 females) were analyzed for determination of Anti-HDV total (IgM+ IgG). Levels of serological markers of HBV infection (HBsAg, HBeAg, Anti-HBe) were measured using microparticle enzyme immunoassay (MEIA) technique (Abbott AxSYM System, Illinois). Positive serum samples were collected and saved at -20°C . Anti-HDV total (with Giuliana Diagnostici S.r.l., Italy) levels were then determined using enzyme-linked immunosorbent assay (by TKA 4HD, Teknolabo A.S.S.I. S.r.l., Italy) method in these HbsAg-positive sera. Serum HBV DNA was determined by quantitative polymerase chain reaction (PCR) assay with a detection limit of 100 copies/ml in chronic hepatitis B and D patients. The gender distribution and mean age of patients were determined. Serum liver function tests (transaminases - ALT, AST, etc.) were analyzed in all chronic liver patients, and two-fold or greater elevation of transaminases was evaluated as the biochemical activity of chronic hepatitis. Liver biopsy was performed in all chronic hepatitis patients and in some of the cirrhotic patients with the suspicion of hepatocellular carcinoma. Biopsy spe-

cimens were stained by hematoxylin and eosin, and the histopathological activity index (HAI) of chronic hepatitis patients was determined according to Knodell's scoring system. In patients without biopsies, the diagnosis of decompensated liver cirrhosis was based mainly on the physical, ultrasonographic and endoscopic examinations and laboratory findings.

Statistical methods

The differences between HDV prevalence ratios (%) in the three groups were analyzed according to the standard normal distribution, named "z-test". Mean ages of chronic HBV and HDV patients were compared by Student's-t test. The prevalences of HBV infection in both sexes in the three groups were compared by Pearson χ^2 test. HBeAg and HBV DNA prevalences were analyzed by z-test in chronic hepatitis B and D patients. Mean values are presented with standard errors in (Table 1, 2).

RESULTS

Epidemiological characteristics of HDV infection are presented in Table 1. HDV infection was detected in 7 (5 males, 2 females; 5%) of 138 asymptomatic HBV carriers, in 24 (18 males, 6 females; 16%) of 148 chronic hepatitis B patients and in 34 (25 males, 9 females; 45%) of 75 cirrhotic HBV patients. Mean ages of HDV carriers, chronic hepatitis D and HDV cirrhosis patients were 31 ± 8 (14-65), 36 ± 13 (19-70) and 44 ± 16 (25-55), respectively. Mean ages of chronic hepatitis B and cirrhotic HBV patients were 31 ± 9 (11-62) and 48 ± 12 (14-70), respectively. Mean age of cirrhotic HDV patients was eight years older than that of chronic hepatitis D patients and four years younger than that of cirrhotic HBV patients. Mean age of chronic hepatitis B patients was five years younger than that of chronic hepatitis D patients. Thus, there is

Table 1. Epidemiological characteristics of HDV infection in Van region

HDV	Males (%)	Females (%)	Total (%)	Mean Age
Carriers	5.2 (5/95)	4.6 (2/43)	5.0 (7/138)	31 ± 8
Chronic Hepatitis	16.3 (18/110)	15.7 (6/38)	16.2** (24/148)	36 ± 13
Cirrhosis	45.4 (25/55)	45.0 (9/20)	45.3*** (34/75)	$44 \pm 16^*$

* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

Table 2. Biochemical, virological and pathological findings in chronic hepatitis B and D

Chronic Hepatitis	ALT (IU)	AST (IU)	HBe Ag (+) (%)	HBV DNA (+) (%)	HAI (pts)
HBV	127 ± 18	101 ± 19	22 (27/124)	85* (105/124)	8 ± 1
HDV	187 ± 75	110 ± 34	13 (3/24)	13 (3/24)	11 ± 1

HAI: histopathological activity index

* $p < 0.001$

an approximately eight-year lapse before the development of decompensated cirrhosis following chronic HDV infection, whereas this period is approximately two times longer (mean: 17 years, $p<0.05$) in chronic HBV infection, suggesting a more rapid progressive prognosis in chronic HDV infection than in chronic HBV infection.

HDV infection ratios were not significantly different between sexes in carriers, chronic hepatitis and cirrhotic patients ($p>0.05$). However, chronic HBV infection was 2.2 times (95/43), 2.9 times (110/38) and 2.75 times (55/20) more frequent in males than in females in carriers, chronic hepatitis and cirrhotic patients, respectively ($p<0.001$).

Biochemical, virological and HAI findings are presented in (Table 2). Serum mean ALT values were three to six times elevated and mean AST values were approximately three times elevated in chronic hepatitis B and chronic hepatitis D, respectively. Serum HbeAg was positive in 22% (27/124) and in 13% (3/24) of chronic hepatitis B and hepatitis D patients, respectively ($p>0.05$). Serum HBV DNA was significantly positive in chronic hepatitis B compared to chronic hepatitis D (85% vs. 13%, $p<0.001$). Also, HAI scores were 8 ± 1 and 11 ± 1 points (pts) in chronic hepatitis B and D patients, respectively ($p>0.05$).

The prevalence of HDV infection was three- and nine-fold increased in chronic hepatitis and cirrhotic patients compared to HDV carriers ($p<0.01$, $p<0.001$). Also, it was three times more frequent in cirrhotics compared to chronic hepatitis patients ($p<0.001$). HDV infection in HBV carriers and in chronic liver diseases is represented in (Figure 1).

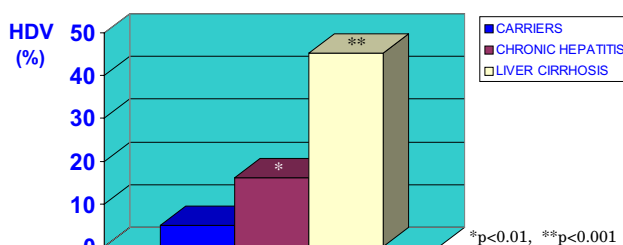


Figure 1. HDV infection in chronic liver diseases

DISCUSSION

Five percent of HBV carriers (15 million people) are infected with HDV worldwide, and areas of high prevalence include Italy, certain parts of Eastern Europe, the Amazon basin, Colombia, Vene-

zuela, Western Asia, and some Pacific Islands. However, in Southern Europe, HDV infection has greatly declined in the last decade owing to the prevention of HBV infection by means of vaccination, avoidance of paid donors and the introduction of disposable needles and syringes. Genotype 1 predominates in most areas of the world. Genotype 2 was originally discovered in Japan and Taiwan, where an association with less severe disease has been proposed. Genotype 3 predominates in South America and is associated with a more severe form hepatitis. HDV superinfection leads to chronic liver diseases in 80-90% of patients. HDV-related cirrhosis develops one to two decades earlier than HBV- and HCV-related cirrhosis in endemic regions (1-6).

In Turkey, it is estimated that approximately 4 million persons are infected by HBV. HBV carriage rate is approximately 2-5% in Western and Central Anatolia Turkey. However, it is significantly elevated (8-10%) in Eastern and Southeastern Anatolia, where people have lower educational and socioeconomical level and less hygienic living conditions (7). HBV infection alone is responsible of 54-80% of chronic liver diseases in Eastern and Southeastern Turkey (8).

An epidemiological study revealed that the prevalence of HDV infection was significantly higher in patients with chronic liver disease (32.7%) than in asymptomatic carriers of HBsAg (5.2%) in Turkey (9). The highest prevalence (45.6%) of HDV infection was found in patients at high risk of acquiring hepatitis virus infection (health care workers, hemodialysis patients, polytransfused patients) with chronic liver disease. The frequency of "severe" or fulminant hepatitis was similar in HBV-infected patients (7.8%) and in HBV/HDV-coinfected individuals (10%). Another epidemiological study concerning risk factors of hepatocellular carcinoma (HCC) in Turkey reported that 56% of patients had hepatitis B and 19% of them (11% of all HCC patients) had hepatitis D (10).

HDV seroprevalence has declined progressively in Western and Central Anatolia Turkey, from 7% to 0.9%, in the last two decades. However, in Eastern and Southeastern Anatolia, it remains at higher rates (4-10 %). Actually, HDV infection is the second major cause of chronic liver diseases in these regions, ranging from 8% to 51% (7, 8, 11). In Diyarbakir, the largest city of Southeastern Anatolia, a high prevalence rate (47%) of HDV infection in liver cirrhosis was reported (12).

In the Van region of Eastern Anatolia, HBV infection alone is presently the major etiological factor of chronic liver diseases. Its prevalence has been reported as 54-79% in liver cirrhosis (13, 14). Moreover, we previously observed that HDV infection is the second major cause of chronic liver diseases in this region (15). In this new study, we detected that 16% of chronic hepatitis B and 45% of HBV cirrhosis are related to HDV infection in the Van region. HDV carriage rate was 5% in asymptomatic HBV carriers. HDV infection showed a three-fold increase in chronic hepatitis ($p < 0.01$) and nine-fold increase in cirrhosis ($p < 0.001$) compared to HDV carriers. It was also three times more frequent in cirrhosis ($p < 0.001$) compared to chronic hepatitis. Interestingly, we observed no significant sexual difference in HDV infection in terms of chronic liver diseases, although chronic HBV infection was three times more frequent in males than in females ($p < 0.001$). We also observed that there is an eight-year lapse before the development of decompensated cirrhosis following chronic HDV infection, whereas this lapse is approximately two times longer (mean: 17 years) in chronic HBV infection for the development of end stage liver disease ($p < 0.05$). The mean age of cirrhotic HDV patients was also four years younger than of

cirrhotic HBV patients. These findings reveal the more severe prognosis of chronic HDV infection than of chronic HBV infection alone. The young age (mean: 36 years) of chronic hepatitis D patients also suggests that the infection has been acquired in childhood or in the adolescence period. Intrafamilial transmission rate of both HBV and HDV infections has been reported as 25% in Eastern and Southeastern Anatolia (8).

In conclusion, the higher prevalence of HDV infection in the more severe form of HBV infection suggests that HDV infection increases the severity of HBV infection. HDV infection is the second major etiological factor of chronic liver diseases in the Van region of Eastern Anatolia as in Southeastern Anatolia. HBV vaccination in the childhood period, hygienic living conditions and improved socioeconomical and educational status are the cornerstones in the prevention of liver morbidity and mortality in Eastern Turkey.

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