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Prevalence, characteristics, and virologic correlations of hepatitis delta (D) among patients with hepatitis B surface antigen in Mongolia

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Abstract: Clinical and biochemical features of hepatitis delta virus (HDV) infections in Mongolia remain largely unknown. We aimed to investigate the clinical characteristics of HDV patients in Mongolia using several markers. The 143 hepatitis B surface antigen (HBsAg)-positive patients were divided into 122 HDV-positive and 21 HDV-negative patients by HDV RNA positivity. Subgroup analysis was performed between hepatitis B e antigen (HBeAg)-positive and -negative HDV-positive patients. Liver function, quantitative HBsAg (qHBsAg), anti-HDV Immunoglobulin (Ig) M, Mac-2 binding protein glycosylation isomer (M2BPGi), hepatitis B virus (HBV) DNA level, and HDV RNA level were tested. HDV RNA was positive in 85.3% (122/143) of patients showing anti-HDV IgG. Liver disease activity was higher in HDV-positive patients than in HDV-negative patients. The HDV-positive group included a higher proportion of patients with high qHBsAg and M2BPGi levels ($p < 0.001$). The positivity rate for anti-HDV IgM was significantly higher in the HDV-positive group ($p < 0.001$). HDV RNA levels showed an inverse correlation with qHBsAg levels in HBeAg-positive-HDV-positive patients ($r = -0.49$, $p = 0.034$), and a positive correlation with qHBsAg levels in HBeAg-negative patients ($r = 0.35$, $p < 0.001$). Hepatitis B virus (HBV) DNA and HDV RNA levels did not show any correlation. M2BPGi levels likewise did not correlate with HDV RNA levels. A high positivity rate for HDV RNA was observed for HBV patients in Mongolia using the highly sensitive HDV RNA assay. The positivity rate for anti-HDV IgM was high in HDV RNA-positive patients. Severity of liver disease and M2BPGi levels were both high in the HDV RNA-positive group.

***Keywords*:** highly sensitive HDV RNA assay, anti-HDV IgM, M2BPGi, hepatitis B virus, quantitative HBsAg, HBeAg

Introduction

Four decades ago, Rizetto *et al.* discovered the hepatitis delta virus (HDV), which requires the hepatitis B virus surface antigen (HBsAg) helper function for viral assembly and propagation (*1*). Mongolia is known as an endemic country with a high prevalence of viral hepatitis. The studies reported the seroprevalence of HDV to be approximately 60–65% among Mongolian patients with chronic hepatitis B (*2,3*).

Sodium taurocholate co-transporting polypeptide (NTCP) was recently identified as an entry receptor for both hepatitis B virus (HBV) and HDV (*4*). HBV uses a retrograde trafficking route for infection that is facilitated by dedicator of cytokinesis 11 (DOCK11) and serves to maintain HBV covalently closed circular DNA (*5*).

The presence of anti-HDV immunoglobulin (Ig) M antibodies is used in clinical practice not only to differentiate between acute and chronic HDV infection (*6*), but also to identify persistent viremia during chronic hepatitis delta (*7*).

Polymerase chain reaction (PCR) is the most reliable diagnostic tool for detecting HDV RNA, but commercial assays have been unavailable because of the genetic heterogeneity of HDV (*8*). In 2017, the World Health Organization (WHO) approved the standard method for quantitative determination of HDV RNA levels (*9*), which was recently validated as a highly sensitive method for accurately evaluating

disease and antiviral treatment outcomes (*10*). Mac-2 binding protein glycosylation isomer (M2BPGi) is a new glyco-biomarker for liver fibrosis found by Japanese researchers (*11-13*) They reported that serum M2BPGi level offers a reliable, non-invasive marker of liver fibrosis in patients with chronic hepatitis B, C, non-alcoholic fatty liver disease, and primary biliary cholangitis (*14*).

Although the prevalence of HDV infection is reportedly high, data is lacking for anti-HDV IgM, M2BPGi, and highly sensitive HDV RNA testing in chronic hepatitis delta patients in Mongolia. We therefore aimed to investigate the clinical characteristics of HDV patients using a combination of sensitive biomarkers such as anti-HDV IgM, HDV RNA testing, qHBsAg, and M2BPGi levels.

Materials and Methods

Patients

Study subjects were selected from among 363 HBV patients being followed-up at Intermed Hospital and the University Hospital of the Mongolian National University of Medical Sciences between June 2015 and August 2016. All patients were negative for HCV infection. The flowchart for study participation is displayed in Figure 1. Among these 363 hepatitis B surface antigen (HBsAg)-positive patients, 306 patients (84.3%) tested positive for anti-HDV IgG. The 158 patients who were not tested for HDV RNA were excluded from the present study. Another five patients who responded to pegylated interferon-α therapy were excluded from this study. All the remaining 143 patients were tested using the highly sensitive HDV RNA assay and were selected for further investigation. Of these 143 patients, 71 (49.7%) had chronic hepatitis and 72 (50.3%) had cirrhosis. We then compared clinical, virologic, and biochemical findings between groups according to HDV status, comprising 122 HDV-positive (HDV RNA-detectable) patients and 21 HDV-negative (HDV RNA-undetectable) patients. We also compared detailed characteristics between hepatitis B e antigen (HBeAg)-positive and HBeAg-negative HDV-positive patients. Liver biopsy was not performed.

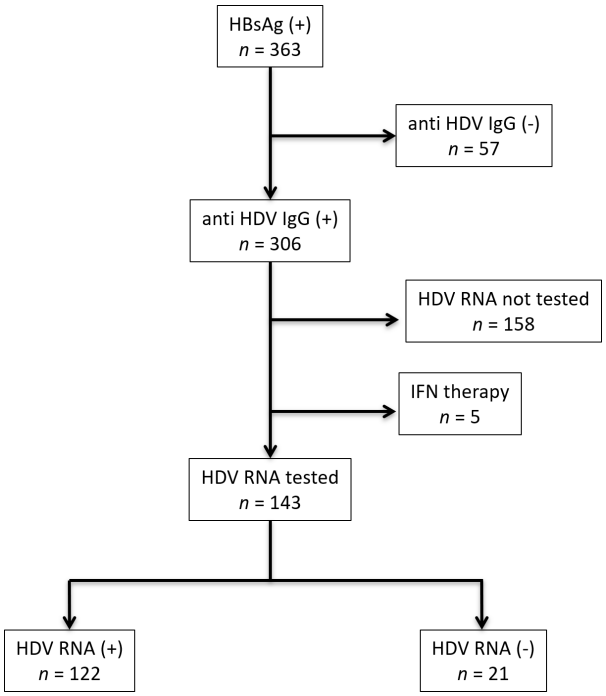

Figure 1. Subjects included in this study.

Clinical laboratory methods

Complete blood counts were determined using commercially available tests (XS-800i analyzer; Sysmex Co., Hyogo, Japan). An automatic biochemistry analyzer Chemix-180 (Sysmex Co.) was used for liver function tests, including aspartate aminotransferase (*10*), alanine aminotransferase (ALT), γ-glutamyl transpeptidase (GGT), alkaline phosphatase (ALP), albumin, and total bilirubin. Upper limits of normal (ULNs) were considered to be 30 U/L for ALT and 33 U/L for AST in women and 40 U/L for ALT and 41 U/L for AST in men.

Serologic methods

HBV markers, α-fetoprotein (AFP), and serum M2BPGi were measured by chemiluminescent enzyme immunoassay using an HISCL-5000 analyzer (Sysmex Co.). The diagnostic range for the quantitative determination of HBsAg titer was 0.03–2,500 IU/mL. Samples showing qHBsAg > 2,500 IU/mL were retested at a higher dilution. M2BPGi level was expressed as a cut-off index. Anti-HDV IgG level was determined by HDV-IgG enzyme immunoassay (Fortress Diagnostics, Antrim, UK). Anti-HDV IgM testing was performed using HDV IgM enzyme-linked immunosorbent assay (DRG Instruments GmbH, Marburg, Germany), according to the manufacturer's instructions. All samples with a signal-to-cutoff ratio (S/Co) > 1.1 were considered positive.

Detection of HBV DNA and HDV RNA

Detection of HDV RNA was performed using a RoboGen HDV RNA quantification kit 2.0 (Roboscreen GmbH, Marburg, Germany). This kit references the first WHO standard for HDV RNA quantification (*9*). Real-time PCR was performed using a Lightcycler 480 (Roche Diagnostics, Mannheim, Germany) in accordance with the manufacturer's instructions. All samples with a detection threshold for viral load >14 IU/mL were considered positive for HDV RNA.

Following DNA extraction from 500 μL of serum

using a High Pure System Viral Nucleic Acid Kit (Roche Diagnostics), quantitative detection of HBV DNA was performed using the Cobas TaqMan HBV test kit 2.0 (Roche Diagnostics) according to the recommendations from the manufacturer on using the Real-Time PCR Cobas TaqMan 48 instrument (Roche Diagnostics).

Statistical analysis

Statistical analyses were performed using SPSS version 22.0 software (SPSS Inc., Chicago, IL). All continuous variables are described as mean ± standard deviation or median and range, as appropriate. Categorical variables are expressed as frequency and percentage. Continuous variables were analyzed using Student's *t*-test or the Mann–Whitney *U*-test. A chi-square test was used to compare discrete variables. Logistic regression models were used to calculate odds ratio and 95% confidence intervals (CIs). Spearman's correlation test was used to determine the correlation between HDV RNA level and other continuous variables. Values of $p < 0.05$ were considered statistically significant.

Ethical considerations

The study design and protocols were reviewed and approved by the Ethics Review Committee of the Mongolian National University of Medical Sciences, in accordance with the 1975 Declaration of Helsinki (as revised in 2013). Written informed consent was obtained from each study subject prior to blood testing.

Results

Comparison of HDV-positive and HDV-negative groups

First, based on HDV RNA assay results, subjects were divided into the two main groups (Figure 1), namely, the HDV-positive and HDV-negative groups. Of the 143 patients, 122 patients (85.3%) tested positive and only 21 tested negative for HDV RNA. Comparisons of main laboratory and clinical characteristics were then performed between the two study groups (Table 1). Study groups showed a similar median age (40 years *vs.* 43 years) and sex distribution (53% *vs.* 52% in men). Compared with the HDV-negative group, the HDV-positive group showed more severe biochemical activity of liver disease, measured as the mean ratio of ALT to ULN (2.0 *vs.* 0.9, $p = 0.003$); mean ratio of AST to ULN (1.6 *vs.* 0.8, $p = 0.005$); GGT level (45 U/L *vs.* 21 U/L, $p = 0.002$); and albumin level (44 g/L *vs.* 48 g/L, $p = 0.025$). Platelet counts were lower in the HDV-positive group (189 $\times 10^9$/L *vs.* 235 $\times 10^9$/L, $p = 0.004$). On the other hand, mean levels of bilirubin and ALP did not differ between groups.

Table 2 shows a significantly higher mean qHBsAg level in the HDV-positive group (3.7 ± 0.7 log IU/mL *vs*. 2.9 ± 1.0 log IU/mL, $p = 0.001$). The HDV-positive group also showed a significantly higher frequency of patients testing positive for HDV IgM (68.1% *vs.* 14.3%, $p < 0.001$). The positivity rate for HBeAg did not differ significantly between groups (15.6% *vs.* 4.8%, $p = 0.31$). The rate of detectable HBV DNA did not differ significantly between the HDV-positive group (30.3%) and HDV-negative group (9.5%, $p = 0.06$). A higher proportion of patients with M2BPGi > 1 of the COI was observed in the HDV-positive group (84.4% *vs.* 38.1%, $p < 0.001$).

Subgroup analysis of HDV-positive patients with and without HBeAg

Since qHBsAg levels depend on HBV viral load and HBeAg positivity, we performed subgroup analyses of HDV-positive patients. Patients were divided into two subgroups: an HBeAg-positive HDV-positive; and an HBeAg-negative HDV-positive group. Table 3 shows the comparison between HBeAg-positive and HBeAg-negative patients with HDV RNA. Median age, sex distribution, and basic laboratory features did not differ

Table 1. Comparison of clinical and biochemical characteristics between HDV-positive and HDV-negative groups

Variables	HDV-positive ($n = 122$)	HDV-negative ($n = 21$)	*p* value
Age, y, median (range)	40 (21–70)	43 (26–65)	0.8
Sex, male, *n* (%)	65 (53.3%)	11 (52.4%)	0.5
WBC, $\times 10^{12}$/L, median (range)	4.8 (2.1–9.6)	5 (2.5–9.0)	0.48
PLT, $\times 10^9$/L, median (range)	189 (57–354)	235 (123–384)	0.004*
ALT, ratio to ULN, mean ± SD	2.0 ± 1.5	0.9 ± 0.7	0.003*
AST, ratio to ULN, mean ± SD	1.6 ± 1.1	0.8 ± 0.4	0.005*
Total bilirubin, μmol/L, median (range)	17 (4–49)	18 (8–39)	0.81
Alkaline phosphatase, U/L, median (range)	300 (128–982)	250 (154–541)	0.06
GGT, U/L, median (range)	45 (11–444)	21 (11–158)	0.002*
Albumin, g/L, median (range)	44 (28–52)	48 (39–54)	0.025*

*p value < 0.05. ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma-glutamyltransferase; HDV, hepatitis delta virus; PLT, platelet count; SD, standard deviation; ULN, upper limit of normal; WBC, white blood cell count; HDV-positive, HDV RNA-positive; HDV-negative, HDV RNA-negative.

Table 2. Comparison of serologic and virologic features between HDV-positive and HDV-negative groups

Variables	HDV-positive (*n* = 122)	HDV-negative (*n* = 21)	OR 95% CI	*p* value
qHBsAg log IU/mL, mean ± SD	3.7 ± 0.7	2.9 ± 1.0	-	0.001*
qHBsAg > 1,000 IU/mL, *n* (%)	108 (88.5%)	10 (47.6%)	8.5 (3.0–23.5)	< 0.001*
HBeAg-positive, *n* (%)	19 (15.6%)	1 (4.8%)	3.7 (0.4–29.1)	0.31
Anti-HBe-positive, *n* (%)	107 (87.7%)	20 (95.2)	2.8 (0.5–22.4)	0.47
Anti-HDV IgM-positive, *n* (%)	83 (68.1%)	3 (14.3%)	12.7 (3.5–45.9)	< 0.001*
HBV DNA (positive PCR), *n* (%)	37 (30.3%)	2 (9.5%)	4.1 (0.9–18.7)	0.06
AFP, ng/mL, median (range)	4.3 (0.6–78.8)	2.3 (1.0–7.5)	-	0.024*
M2BPGi, COI, median (range)	1.90 (0.1–12.4)	0.77 (0.4–2.7)	-	0.001*
M2BPGi > 1 COI, *n* (%)	103 (84.4%)	8 (38.1%)	8.8 (3.2–24.1)	< 0.001*

*p value < 0.05. AFP, α-fetoprotein; anti-HBe, antibody against hepatitis B e antigen; anti-HDV IgM, immunoglobulin M against hepatitis delta virus; CI, confidence interval; COI, cut-off index; HBeAg, hepatitis B e antigen; HBV DNA, hepatitis B virus deoxyribonucleic acid; HDV, hepatitis delta virus; M2BPGi, Mac-2 binding protein glycosylation isomer; PCR, polymerase chain reaction; OR, odds ratio; qHBsAg, quantitative hepatitis B surface antigen; HDV-positive, HDV RNA-positive; HDV-negative, HDV RNA-negative.

Table 3. Comparison of clinical and biochemical characteristics between HBeAg-positive and HBeAg-negative subgroups in HDV-positive patients

Variables	HBeAg-positive (*n* = 19)	HBeAg-negative (*n* = 103)	*p* value
Age, y, median (range)	42 (26–65)	40 (21–70)	0.8
Sex, male (%)	11 (57.9%)	54 (52.4%)	0.8
WBC, $\times 10^{12}$/L, median (range)	4.4 (2.1–9.6)	4.8 (2.1–8.9)	0.8
PLT, $\times 10^{9}$/L, median, (range)	189 (76–306)	188 (57–345)	0.9
ALT, ratio to ULN, mean ± SD	1.8 ± 1.3	2.0 ± 1.5	0.6
AST, ratio to ULN, mean ± SD	1.6 ± 1.0	1.6 ± 1.1	0.9
Total bilirubin, μmol/L, median (range)	20 (4–41)	17 (4–49)	0.1
Alkaline phosphatase, U/L, median (range)	303 (223–634)	297 (128–982)	0.6
GGT, U/L, median (range)	45 (11–413)	44 (11–444)	0.7
Albumin, g/L, median (range)	48 (39–54)	44 (28–52)	0.2

ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma-glutamyltransferase; HBeAg, hepatitis B e antigen; PLT, platelet count; SD, standard deviation; ULN, upper limit of normal; WBC, white blood cell count.

Table 4. Comparison of serologic and virologic features between HBeAg-positive and HBeAg-negative subgroups among HDV-positive patients

Variables	HBeAg-positive (*n* = 19)	HBeAg-negative (*n* = 103)	*p* value
qHBsAg, log IU/mL, mean ± SD	4.2 ± 0.3	3.6 ± 0.7	0.006*
Anti-HDV IgM-positive, *n* (%)	13 (68.4%)	70 (67.9%)	0.6
HBV DNA, log IU/mL	2.5 ± 1.8	1.8 ± 0.8	0.023*
HDV RNA, log IU/mL	5.2 ± 1.1	5.2 ± 1.2	0.9

*p value < 0.05. Anti-HDV IgM, immunoglobulin M against hepatitis delta virus; HBeAg, hepatitis B e antigen; HBV DNA, hepatitis B virus deoxyribonucleic acid; HDV RNA, hepatitis delta virus ribonucleic acid; qHBsAg, quantitative hepatitis B surface antigen.

between groups. As predicted, mean qHBsAg level was significantly higher in HBeAg-positive patients (4.2 log IU/mL) than in HBeAg-negative patients (3.6 log IU/mL, $p = 0.006$) (Table 4). The proportion of patients with anti-HDV IgM positivity was around 68% in both groups (Table 4). HBV DNA was detectable in 52.6% of HBeAg-positive patients and 26.2% of HBeAg-negative patients. Mean HBV DNA level was significantly higher in HBeAg-positive patients (2.5 ± 1.8 log IU/mL) than in HBeAg-negative patients (1.8 ± 0.8 log IU/mL, $p = 0.023$). On the other hand, HDV RNA level did not differ between groups (5.2 ± 1.1 and 5.2 ± 1.2 log IU/mL, $p = 0.9$).

On correlation analyses, serum HDV RNA level correlated negatively with qHBsAg levels in HBeAg-positive HDV patients ($r = -0.49$, $p = 0.034$) and positively in HBeAg-negative HDV patients ($r = 0.35$, $p < 0.001$) (Table 5). In HBeAg-positive subjects, serum GGT level was significantly associated with HDV RNA level ($r = -0.52$, $p = 0.028$). In the HBeAg-negative group, ALT level was associated with HDV RNA level ($r = 0.35$, $p < 0.001$). No association was found between HBV DNA and HDV RNA levels in this study. M2BPGi level also did not correlate with HDV RNA level.

Table 5. Correlation of quantitative HDV RNA level with other variables in HBeAg-positive and HBeAg-negative HDV-positive patients

Variables	HBeAg-positive (n = 19)		HBeAg-negative (n = 103)	
	r	p value	r	p value
Age	0.11	0.64	-0.07	0.4
PLT	-0.14	0.56	0.02	0.78
ALT	-0.23	0.33	0.35	< 0.001*
GGT	-0.52	0.028*	0.09	0.32
ALP	-0.35	0.13	0.09	0.34
M2BPGi	-0.13	0.58	-0.53	0.59
qHBsAg log IU/mL	-0.49	0.034*	0.35	< 0.001*
HBV DNA	-0.25	0.40	-0.09	0.47

*p value < 0.05. ALP, alkaline phosphatase; AFP, α-fetoprotein; GGT, gamma-glutamyltransferase; HBeAg, hepatitis B e antigen; HBV DNA, hepatitis B virus deoxyribonucleic acid; HDV RNA, hepatitis delta virus ribonucleic acid; M2BPGi, Mac-2 binding protein glycosylation isomer; PLT, platelet count; qHBsAg, quantitative hepatitis B surface antigen; r, correlation coefficient.

Discussion

Interest in HDV infection has been re-emerging during recent years. Several reports on topics including HDV virology, immune control of HDV infection, and defining the clinical benefits of interferon therapy have been published (*15-18*). Reliable global epidemiologic data on the prevalence of HDV and clinical data are required in light of these new findings.

Mongolia stands out with one of the highest recorded prevalence rates of HDV globally (*2,3*). This unique epidemiological scenario may contribute to distinctive drivers identified in Mongolian HCC, including noteworthy genomic factors associated with hepatitis D viral infection (*19*). Recently published data have revealed that 65% of HBsAg carriers in the country were co-infected with HDV, with 74% of these cases detectable for HDV RNA (*3*).

In our study, we found that 84.3% of HBsAg-positive patients were positive for anti-HDV IgG. Moreover, 85.3% of patients positive for the anti-HDV IgG antibody also tested positive for HDV RNA. This observation may be related to the specific characteristics of the study population in our research.

The HDV RNA assay used in this study was validated by the WHO in recent years. This highly sensitive assay is the standard for HDV quantification (*9, 10*).

Hepatitis severity was higher in the HDV-positive group, supporting previous findings from Western countries (*20,21*). However, a report from Taiwan found no relationship between severity of hepatitis and anti-HDV status (*22*). The less severe course of HDV infection in Taiwan compared to Western countries may be explained by differences in the distribution of HDV genotypes between geographic regions. HDV genotype 2 has been shown to be predominant in Taiwan and is associated with a milder disease course. In contrast, HDV genotype 1 is predominant in Western countries and Mongolia, with a more pronounced severity of disease compared to genotype 2 (*3,23*).

In this study, a positive correlation was found between the HBsAg levels and the HDV RNA load in HBeAg-negative group. This was similar to a previous report, and was attributed to the ease with which HDV proliferates (*24*). Liver inflammation and fibrosis were also more pronounced in the HDV-positive group. These findings were also similar to previous reports (*7,24*). M2BPGi was higher in the HDV-positive group and was considered to reflect fibrosis and inflammation. The HDV-positive group had a high anti-HDV IgM-positivity rate indicating active hepatitis delta infection (*7*). The HDV-negative group may have included cases in which only HDV had been eliminated after co-infection with HBV.

HDV has been reported to suppress HBV replication (*25-27*).However, no correlation between levels of HDV RNA and HBV DNA was shown in this study. This may be due to differences in the combinations of HDV and HBV genotypes between reports (*7*). In Mongolia, HBV genotype D and HDV genotype 1 are reportedly dominant (*3,23,28,29*). Another report found that HBV genotype B and HDV genotype 2 were unrelated in Taiwan (*30*). Unfortunately, the present study could not consider the genotypes of HBV and HDV because it was not possible to examine the virus genotypes this time.

Comparing the HBeAg-positive and HBe-negative groups revealed that severity of hepatitis delta was not significantly related to HBeAg status. Similar findings have been reported previously (*31*). Consistent with previous reports, no significant difference in HDV RNA levels was evident between HBeAg-positive and -negative groups (*30*). Likewise, no difference was found in anti-HDV IgM prevalence, probably because HDV RNA levels were comparable (*7*). Notably, our study explores separate examinations for correlations between HDV RNA levels and liver disease markers in both HBeAg-positive and HBeAg-negative cases. ALT levels showed a weak positive correlation with HDV RNA levels in the HBeAg-negative group. GGT showed an inverse correlation with HDV RNA levels in the HBeAg-positive group. The clinical implications of these findings require further investigation.

In our study, we measured M2BPGi in Mongolian patients with HDV. M2BPGi serves as a marker for inflammation and fibrosis in liver diseases of various origins (*14*). Previous assessments included measurements in healthy individuals (*32*), patients with HBV and HCV (*33*), and HDV-positive patients (*34*). The latter demonstrated significantly elevated M2BPGi levels in HDV patients compared to healthy individuals (6.04 ± 5.8 COI *vs.* 0.8 ± 0.49 COI), particularly in those with high HDV viral loads as opposed to low viral loads.

A noteworthy discovery in our study was the lack of correlation between M2BPGi and HDV RNA levels in HDV RNA-positive patients. Nonetheless, M2BPGi levels were higher in the HDV-positive group compared to the HDV-negative group. Taken together, these findings suggest that M2BPGi could serve as a potential predictor of disease progression in HDV infection. Further investigation is necessary to fully understand the significance of M2BPGi in HDV infection, and this remains an avenue for exploration in future research.

The routes of infection are thought to include mother-to-child transmission, iatrogenic incidents, folk remedies, tattoos, intravenous drug use (IVDU), and sexual contact including that among men who have sex with men. IVDU is well recognized as a risk factor for HDV co-infection (*35*), but medical records in the present study did not consistently contain such information.

Taiwan is considered to be endemic for hepatitis B, but HBV prevalence has decreased markedly following the implementation of effective immunization schedules (*36*). In Mongolia, however, HDV prevalence still appears high. This continues to be a public problem of medical importance.

The data in this study were obtained from among HBV patients who visit hospitals in Mongolia and will be important in the future. A common standard measurement is ideal for obtaining epidemiologic data. Use of a common measurement kit would thus be indispensable for measuring HDV RNA in anti-HDV-positive individuals around the world.

This study aims to compare HDV-positive and HDV-negative groups among anti-HDV-positive subjects in Mongolia using a high-sensitive HDV RNA assay to diagnose active HDV infection. However, several limitations should be considered. Firstly, the study is constrained by a small sample size, which may affect the generalizability of the findings. Additionally, the availability of data on liver biopsies and genotyping of HBV and HDV is limited, limiting a comprehensive understanding of the disease dynamics. Future studies should address these limitations and delve into investigating the risk factors, clinical and virologic characteristics, as well as host factors influencing HDV viral clearance in Mongolian patients with chronic hepatitis D.

In conclusion, we measured sera of HDV patients in Mongolia and confirmed a high positivity rate using a highly sensitive HDV RNA assay. The anti-HDV IgM-positivity rate was high among HDV RNA-positive patients. HDV RNA-positive patients showed more severe hepatitis and higher M2BPGi levels, but no correlation was seen between HDV RNA levels and M2BPGi levels.

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