Kaohsiung Journal of Medical Sciences (2016) 32, 526–530

Available online at www.sciencedirect.com

ScienceDirect

journal homepage: http://www.kjms-online.com

ORIGINAL ARTICLE

Hepatitis D virus infections among injecting drug users with and without human immunodeficiency virus infection in Taiwan

Meng-Hsuan Hsieh [a,b], Shu-Chi Wang [c], Ming-Yen Hsieh [d], Chung-Feng Huang [b,e], Ming-Lun Yeh [d], Jeng-Fu Yang [a], Ko Chang [b,f], Wei-Ru Lin [g], Chun-Yu Lin [b,g], Tun-Chieh Chen [b,d], Jee-Fu Huang [b,c], Chia-Yen Dai [a,b,c], Jih-Jin Tsai [b,g,h,*], Wan-Long Chuang [b,c], Ming-Lung Yu [b,c,i]

[a] *Health Management Center, Kaohsiung Medical University Chung-Ho Memorial Hospital, Kaohsiung City, Taiwan*
[b] *School of Medicine, College of Medicine, Kaohsiung Medical University, Kaohsiung City, Taiwan*
[c] *Hepatitis Center and Hepatobiliary Division, Department of Internal Medicine, Kaohsiung Medical University Chung-Ho Memorial Hospital, Kaohsiung City, Taiwan*
[d] *Department of Internal Medicine, Kaohsiung Municipal Ta-Tung Hospital, Kaohsiung Medical University, Kaohsiung City, Taiwan*
[e] *Department of Occupational Medicine, Kaohsiung Municipal Ta-Tung Hospital, Kaohsiung City, Taiwan*
[f] *Department of Internal Medicine, Kaohsiung Municipal Hsiao-Kang Hospital, Kaohsiung City, Taiwan*
[g] *Division of Infectious Diseases, Department of Internal Medicine, Kaohsiung Medical University Chung-Ho Memorial Hospital, Kaohsiung City, Taiwan*
[h] *Tropical Medicine Center, Kaohsiung Medical University Chung-Ho Memorial Hospital, Kaohsiung City, Taiwan*
[i] *Graduate Institute of Clinical Medicine, College of Medicine, Kaohsiung Medical University, Kaohsiung City, Taiwan*

Received 21 June 2016; accepted 27 July 2016
Available online 18 September 2016

KEYWORDS
Hepatitis B virus;
Hepatitis D virus;

Abstract In Taiwan, injecting drug use has been the main route of human immunodeficiency virus (HIV) transmission since 2005, with hepatitis B virus (HBV) and hepatitis D virus (HDV) also having similar transmission routes. This has now become an important public health issue. The

Conflicts of interest: All authors declare no conflicts of interest.

* Corresponding author. Division of Infectious Diseases, Department of Internal Medicine, Kaohsiung Medical University Chung-Ho Memorial Hospital, Number 100, Tzyou 1st Road, Kaohsiung 80756, Taiwan.
E-mail address: jijits@cc.kmu.edu.tw (J.-J. Tsai).

http://dx.doi.org/10.1016/j.kjms.2016.08.004

Human immunodeficiency virus;
Injecting drug users;
Taiwan

aim of this study is to explore the conditions of HDV infections between injecting drug users (IDUs) with and without HIV infection in Southern Taiwan. In this study, 87 IDUs were enrolled, including 27 anti-HDV seronegative IDUs and 60 anti-HDV seropositive IDUs, and the results of their liver function tests, CD4 cell counts, and anti-HIV and HIV RNA levels were analyzed. The prevalence of anti-HDV seropositivity among hepatitis B surface antigen (HBsAg) seropositive IDUs in this study was 68.9% (60/87). The prevalence rate of anti-HDV seropositive IDUs among anti-HIV seronegative and anti-HIV seropositive cases was 40.0% (12/30) and 84.2% (48/57), respectively. Anti-HIV seropositivity was related to anti-HDV seropositivity (odds ratio = 9.34, 95% confidence interval = 2.67–31.59, $p < 0.001$). Among IDUs with HIV infection, there was no significant difference in CD4 cell counts and HIV RNA viral load between HBsAg-positive patients with anti-HDV seronegativity and those with anti-HDV seropositivity. In conclusion, the prevalence of HDV infection among IDUs is higher among IDUs with HIV infection. Because anti-HIV seropositivity is significantly related to anti-HDV seropositivity, HDV infection among IDUs is still important. We suggest that for IDUs, HBsAg and anti-HDV should be monitored closely.

Introduction

Hepatitis D virus (HDV) is a defective virus, which requires life-cycle function provided by the hepatitis B virus (HBV) [1]. HDV is reported globally, with more than 10 million people currently infected with the virus [2]. Estimates suggest that there are about 370 million individuals with chronic HBV infection [3], about 40 million individuals with human immunodeficiency virus (HIV) infections [4], and 3–6 million HIV-infected individuals coinfected with chronic HBV [5]. In addition, it was estimated that about 5% of HBV carriers are coinfected with HDV, and thus, there are about 15 million individuals infected with HDV worldwide [6]. HDV coinfection with HBV may lead to exacerbation and rapid progression of chronic liver disease among HBV monoinfected individuals [7], and HDV can increase the risk of decompensated liver disease and death of HBV/HIV coinfected individuals [8,9].

Because HBV and HIV share common routes of transmission, especially blood-borne transmission, such as injecting drug use [10], injecting drug use is a very important cause of HBV infection worldwide [11]. As a result, HDV transmission can also spread with HBV via a similar route.

One study in the United States reported that HDV prevalence among injection drug users was 15% in 1988–1989 and 11% in 2005–2006 [12]. In Taiwan, injecting drug use has been the main route of transmission of HIV since 2005 [13], with HDV also being prevalent among injecting drug users (IDUs; 75.4% among IDUs with HIV infection and 66.1% among IDUs without HIV infection in 2006) [14].

Because the prevalence of chronic HBV infection in Taiwan is about 15–20% among the general population born before the implementation of a nationwide HBV vaccination program in 1984 [15], HDV infection might be higher among those born before 1984, compared with those born after 1984. Therefore, the aim of this study was to explore the conditions of HDV infections among IDUs with and without HIV infection in Southern Taiwan.

Methods

Study participants

We enrolled IDUs from three prisons and Kaohsiung Medical University Chung-Ho Memorial Hospital (Kaohsiung City, Taiwan) from March 2008 to June 2010. We excluded individuals with severe liver, renal, or other systemic disease, such as decompensated hepatic failure, renal failure, any malignant disease, or severe heart failure, etc. This study was approved by the ethics committee of Kaohsiung Medical University Hospital. Signed informed consents were obtained from all participants.

Laboratory data

We measured the levels of aspartate aminotransferase (AST), alanine aminotransferase (ALT), hepatitis B surface antigen (HBsAg), anti-HIV antibody, HIV RNA viral load, CD4 cell count, and anti-HDV antibody. HBsAg was examined by enzyme immunoassay (EIA; Abbott Laboratories, North Chicago, IL, USA). Anti-HIV antibody assay (SERODIA-HIV; Fujirebio, Tokyo, Japan) was performed according to the manufacturer's instructions. HIV viral loads were quantified using reverse transcription-polymerase chain reaction (Roche Amplicor, version 1.5; Roche Diagnostics, Branchburg, NJ, USA), and CD4 counts were determined using FACFlow (BD FACSCalibur; Becton Dickinson, San Jose, CA, USA). Anti-HDV antibody was examined using an AUSAB EIA kit (Abbott Laboratories, Abbott Park, IL, USA).

Statistical analysis

Statistical tests used in this study were Student t test and the χ^2 square test. In addition, multivariate analysis was used for estimating the odds ratios (ORs) and 95% confidence intervals (CIs). All of these tests were two sided, and the significance levels were set at $\alpha = 0.05$. All statistical

analysis were performed using the SPSS statistical package (SPSS Inc., Chicago, IL, USA).

Results

In this study, 87 HBsAg seropositive IDUs were enrolled, including 60 anti-HDV seropositive IDUs and 27 anti-HDV seronegative IDUs. The prevalence of anti-HDV seropositivity among HBsAg seropositive IDUs was 68.9% (60/87) in this study.

Table 1 shows the basic characteristics of anti-HDV seropositive and seronegative IDUs among the 87 HBsAg seropositive IDUs studied. There was a male predominance among anti-HDV seronegative and seropositive IDUs. Univariate analysis showed a statistical significance ($p < 0.001$) in the association between anti-HIV and anti-HDV, and the prevalence rate of anti-HDV seropositive IDUs among anti-HIV seronegative and seropositive cases was 40.0% (12/30) and 84.2% (48/57), respectively. Lower HIV RNA viral load and higher CD4 cell count were noted in anti-HDV seropositive IDUs. Although AST and ALT ratios were also higher among anti-HDV seropositive IDUs, no significant association was noted (Table 1).

The factors related to seropositive anti-HDV among HBsAg seropositive IDUs were analyzed by multivariate analysis. Anti-HIV seropositivity was the factor of anti-HDV seropositivity among HBsAg seropositive IDUs in this study (OR = 9.34, 95% CI = 2.76–31.95; Table 2).

Table 1 Basic characteristics of anti-HDV seropositive and seronegative injecting drug users among the 87 HBsAg seropositive injecting drug users in this study.

	Univariate analysis		
	Anti-HDV seronegative	Anti-HDV seropositive	*p*
Total, *n* (%)	27 (100.0)	60 (100.0)	
Age (y)	33.04 ± 7.97	35.66 ± 6.26	0.69
Sex			
Male, *n* (%)	18 (67.7)	50 (83.3)	0.82
Female, *n* (%)	9 (33.3)	10 (16.7)	
Aspartate aminotransferase (ratio for UNL)	0.87 ± 0.47	0.97 ± 0.54	0.44
Alanine aminotransferase (ratio for UNL)	1.03 ± 0.92	1.23 ± 0.78	0.35
Anti-HIV			
Seronegative, *n* (%)	18 (67.7)	12 (20.0)	**<0.001**
Seropositive, *n* (%)	9 (33.3)	48 (80.0)	
HIV RNA, log(IU/mL)[a]	3.38 ± 0.42	3.15 ± 0.77	0.40
CD4 cell count[a]	385.33 ± 120.56	459.44 ± 159.52	0.19
>500 cells/μL	1 (11.1)	21 (43.8)	0.65
<500 cells/μL	8 (88.9)	27 (56.3)	

HBsAg = hepatitis B surface antigen; HDV = hepatitis D virus; HIV = human immunodeficiency virus; UNL = upper normal limit.
Bold value means $p < 0.05$.
[a] For 57 anti-HIV seropositive individuals.

Table 2 Multivariate analysis for evaluating factors of anti-HDV seropositivity among HBsAg seropositive injecting drug users.

Factors	Comparison	Odds ratio	95% Confidence interval	*p*
Anti-HIV	0 = negative; 1 = positive	9.34	2.76–31.59	<0.001
Sex	0 = male; 1 = female	1.36	0.36–5.24	0.65
Age	Increasing by 1 y	0.99	0.92–1.07	0.82

Factors used for analysis were anti-HIV, sex, and age.
CI = confidence interval; HBsAg = hepatitis B surface antigen; HDV = hepatitis D virus; HIV = human immunodeficiency virus.

We also performed a multivariate analysis for identifying factors responsible for abnormal levels of AST and ALT (Table 3). Unfortunately, no factor was identified. However, anti-HIV showed the highest OR value for abnormal AST (OR = 3.24) and ALT (OR = 2.99) levels among all factors evaluated.

Discussion

HDV is prevalent in the Amazon Basin, the Mediterranean Basin, and Central Africa. It is spread by close contact and is endemic to many Mediterranean countries. In the United States, HDV is confined to individuals exposed to contaminated blood [2]. Globally, about 5% of the HBsAg carriers

Table 3 Multivariate analysis for evaluating factors responsible for abnormal AST and ALT levels among HBsAg seropositive injecting drug users.

Factors	Comparison	Odds ratio	95% CI	*p*
AST				
Anti-HDV	0 = negative; 1 = positive	1.66	0.49–5.67	0.42
Anti-HIV	0 = negative; 1 = positive	3.24	0.78–13.45	0.11
Sex	0 = male; 1 = female	1.63	0.39–6.77	0.50
Age	Increasing by 1 y	0.99	0.92–1.06	0.74
ALT				
Anti-HDV	0 = negative; 1 = positive	0.64	0.21–1.94	0.42
Anti-HIV	0 = negative; 1 = positive	2.99	0.84–10.68	0.09
Sex	0 = male; 1 = female	1.01	0.28–3.62	0.99
Age	Increasing by 1 y	0.94	0.88–1.01	0.11

Factors for analyzing are anti-HDV, anti-HIV, sex, and age.
ALT = alanine aminotransferase; AST = aspartate aminotransferase; CI = confidence interval; HBsAg = hepatitis B surface antigen; HDV = hepatitis D virus; HIV = human immunodeficiency virus.

are also infected by HDV [16]; this rate is slightly higher in Europe (14.5%) [17]. However, after the implementation of HBV vaccination programs since the 1990s, the prevalence of HDV has consistently declined in the developed countries [16]. In Taiwan, 6.7% of chronic hepatitis B patients were positive for anti-HDV [18]. The prevalence of HDV infection among IDUs is 0% in Brazil [19] and 1.6% (2/122) in Spain [20]. However, from a previous study in Taiwan, 85% of HBsAg-positive carriers with drug abuse were positive for anti-delta [21]. In this study, the prevalence of anti-HDV seropositivity among HBsAg seropositive IDUs was 68.9%. Thus, in Taiwan, the prevalence of HDV infection is higher among IDUs; in fact, this HDV prevalence is also higher than that in Brazil and Spain. Thus, these data suggest that HDV infection among IDUs is severe in Taiwan. Although the underlying reason for this high prevalence rate is unclear, this issue should be addressed immediately.

HIV/HBV patients are at a potential risk of HDV infection; therefore, examining the presence of HDV infection is necessary in these patients. One study reported that of the 26 HDV seropositive general participants studied, 61.5% (16/26) exhibited HIV seropositivity [22]. In another study in Brazil, 1.2% (1/86) of HIV/HBV coinfected patients were found to be anti-HDV seropositive [23]. In addition, the prevalence of anti-HDV in chronic HBsAg-positive/HIV carriers in EuroSIDA was 14.5% (61/422), with hepatitis D being predominant among intravenous drug users, which explains why hepatitis delta predominated in southern and/or eastern Europe [17]. A previous study in Taiwan showed that HDV is also prevalent among IDUs (75.4% among IDUs with HIV infection and 66.1% among IDUs without HIV infection in 2006) [14], with an another study, also conducted in Taiwan, showing that the prevalence rates of HDV infection were 74.9% and 43.9% among HIV-infected IDUs and HIV-uninfected IDUs, respectively [24]. In our study, the prevalence of anti-HDV seropositivity among HBsAg seropositive individuals was 84.2% (48/57) among IDUs with HIV infection and 40.0% (12/30) among IDUs without HIV infection. Although the difference between these two studies could be attributed to differences in the backgrounds of the participants, our study result indicates a higher prevalence of anti-HDV seropositive IDUs among IDUs with HIV infection than previous studies in Taiwan [14,24]. The authors opine that HDV infection among IDUs has gradually become severe over the years in Taiwan. Furthermore, in this study, anti-HIV seropositivity was found to be related to anti-HDV seropositivity, so we suggest that for IDUs, including HBsAg seropositive individuals whose HBV DNA is undetectable, anti-HDV screening is necessary.

In this study, the ratios of AST and ALT were higher among IDUs with HDV infection, although no significant difference was noted. In addition, none of the factors studied was related to abnormal ratios of AST and ALT in this study. We thought these might be due to small number of cases studied, and this issue will be addressed in the future study.

The CD4 cell count was higher in the anti-HDV seropositive group, the percentage of CD4 cell count < 500 cells/μL was higher in the anti-HDV seronegative group, and HIV RNA viral load was higher among those in the anti-HDV seronegative group. Besides, no significant difference between CD4 cell count, HIV RNA viral load, and anti-HDV was noted in this study. We think that HIV RNA viral load and CD4 cell count are not related to anti-HDV infection in HBsAg seropositive IDUs, and it is also possible that anti-HDV has an influence on HIV infection among IDUs.

The limitation of this study is that the IDUs in this study were prisoners, so visiting and following them up was very difficult, and thus data collected, such as serial changes in the disease course and therapy given, could not be assessed completely. In addition, the number of cases studied is small, and therefore some analysis did not show significant difference, although we think that the difference is reasonable.

Conclusion

The prevalence of anti-HDV seropositivity among HBsAg seropositive IDUs was 68.9%, and this rate was higher among IDUs with HIV infection than among those without HIV infection; additionally, anti-HIV seropositivity was related to anti-HDV seropositivity. Abnormal ratios of AST/ALT and CD4 cell count were not related to anti-HDV among HBsAg seropositive IDUs. In Taiwan, because HDV infection among IDUs is becoming severe gradually, we suggest that for IDUs, HBsAg, and anti-HDV should be closely monitored. We also recommend that abstinence from drug abuse is a very important method that can prevent HBV and HDV infection in these individuals.

Acknowledgments

This study was supported by grants from Kaohsiung Medical University Chung-Ho Memorial Hospital (KMUH104-4R067), Taiwan

References

[1] Rizzetto M, Canese MG, Gerin JL, London WT, Sly DL, Purcell RH. Transmission of the hepatitis B virus-associated delta antigen to chimpanzees. J Infect Dis 1980;141:590—602.
[2] Centers for Disease Control and Prevention (CDC). A CDC map of HDV prevalence across the globe. Atlanta, GA: CDC; 2002. Available at: http://web.stanford.edu/group/virus/delta/2002blejer/Epidemiology.htm. [accessed 18.08.16].
[3] World Health Organization (WHO). Hepatitis B fact sheet. Geneva, Switzerland: WHO; 2005. Available at: http://www.who.int/mediacentre/factsheets/fs204/en/ [accessed 18.08.16].
[4] Joint United Nations Programme on HIV/AIDS (UNAIDS). AIDS epidemic update 2004. Geneva, Switzerland: UNAIDS; 2005. Available at: http://www.unaids.org/wad2004/report_pdf.html [accessed 18.08.16].
[5] Kourtis AP, Bulterys M, Hu DJ, Jamieson DJ. HIV-HBV coinfection—a global challenge. N Engl J Med 2012;366:1749—52.
[6] Farci P. Delta hepatitis: an update. J Hepatol 2003;39:S212—9.
[7] Stroffolini T, Ferrigno L, Cialdea L, Catapano R, Palumbo F, Novaco F, et al. Incidence and risk factors of acute delta hepatitis in Italy: results from a national surveillance system. SEIEVA collaborating group. J Hepatol 1994;21:1123—6.
[8] Fernández-Montero JV, Vispo E, Barreiro P, Sierra-Enguita R, de Mendoza C, Labarga P, et al. Hepatitis delta is a major

determinant of liver decompensation events and death in HIV-infected patients. Clin Infect Dis 2014;58:1549—53.
[9] Sheng WH, Hung CC, Kao JH, Chang SY, Chen MY, Hsieh SM, et al. Impact of hepatitis D virus infection on the long-term outcomes of patients with hepatitis B virus and HIV coinfection in the era of highly active antiretroviral therapy: a matched cohort study. Clin Infect Dis 2007;44:988—95.
[10] Koziel MJ, Peters MG. Viral hepatitis in HIV infection. N Engl J Med 2007;356:1445—54.
[11] Nelson PK, Mathers BM, Cowie B, Hagan H, Des Jarlais D, Horyniak D, et al. Global epidemiology of hepatitis B and hepatitis C in people who inject drugs: results of systematic reviews. Lancet 2011;378:571—83.
[12] Kucirka LM, Farzadegan H, Feld JJ, Mehta SH, Winters M, Glenn JS, et al. Prevalence, correlates, and viral dynamics of hepatitis delta among injection drug users. J Infect Dis 2010; 202:845—52.
[13] Department of Health, Taiwan (DOH). Review data of monitoring for infectious disease. Taipei, Taiwan: DOH, Taiwan; 2013. Available at: http://www.cdc.gov.tw/uploads/files/201308/9590e86d-875f-4d65-8435-dbcc95d9fe6d.pdf.
[14] Chang SY, Yang CL, Ko WS, Liu WC, Lin CY, Wu CH, et al. Molecular epidemiology of hepatitis D virus infection among injecting drug users with and without human immunodeficiency virus infection in Taiwan. J Clin Microbiol 2011;49:1083—9.
[15] Sun HY, Ko WC, Tsai JJ, Lee HC, Liu CE, Wong WW, et al. Seroprevalence of chronic hepatitis B virus infection among taiwanese human immunodeficiency virus type 1-positive persons in the era of nationwide hepatitis B vaccination. Am J Gastroenterol 2009;104:877—84.
[16] Rizzetto M, Ciancio A. Epidemiology of hepatitis D. Semin Liver Dis 2012;32:211—9.
[17] Soriano V, Grint D, d'Arminio Monforte A, Horban A, Leen C, Poveda E, et al. Hepatitis delta in HIV-infected individuals in Europe. AIDS 2011;25:1987—92.
[18] Chen DS, Lai MY, Sung JL. Delta agent infection in patients with chronic liver diseases and hepatocellular carcinoma—an infrequent finding in Taiwan. Hepatology 1984;4:502—3.
[19] Oliveira ML, Bastos FI, Telles PR, Yoshida CF, Schatzmayr HG, Paetzold U, et al. Prevalence and risk factors for HBV, HCV and HDV infections among injecting drug users from Rio de Janeiro, Brazil. Braz J Med Biol Res 1999;32:1107—14.
[20] Santana Rodríguez OE, Malé Gil ML, HernándezSantana JF, Limiñana Cañal JM, Martín Sánchez AM. Prevalence of serologic markers of HBV, HDV, HCV and HIV in non-injection drug users compared to injection drug users in Gran Canaria, Spain. Eur J Epidemiol 1998;14:555—61.
[21] Chen PJ, Chen DS, Chen CR, Chen YY, Chen HM, Lai MY, et al. Delta infection in asymptomatic carriers of hepatitis B surface antigen: low prevalence of delta activity and effective suppression of hepatitis B virus replication. Hepatology 1988;8:1121—4.
[22] Nicolini LA, Taramasso L, Schiavetti I, Giannini EG, Beltrame A, Feasi M, et al. Epidemiological and clinical features of hepatitis delta in HBsAg-positive patients by HIV status. Antivir Ther 2015;20:193—7.
[23] Mendes-Correa MC, Gomes-Gouvêa MS, Alvarado-Mora MV, Da Silva MH, Lázari C, Cavalcanti NC, et al. Hepatitis delta in HIV/HBV co-infected patients in Brazil: is it important? Int J Infect Dis 2011;15:e828—32.
[24] Lin HH, Lee SS, Yu ML, Chang TT, Su CW, Hu BS, et al. Changing hepatitis D virus epidemiology in a hepatitis B virus endemic area with a national vaccination program. Hepatology 2015; 61:1870—9.