



## Trends of recent hepatitis C virus infection among HIV-positive men who have sex with men in Taiwan, 2011–2018

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### ARTICLE INFO

#### Article History:

Received 14 April 2020

Revised 8 June 2020

Accepted 11 June 2020

Available online xxx

#### Keywords:

Chemsex

Direct-acting antiviral

Genotyping

Seroconversion

Sexually transmitted infection

### ABSTRACT

**Background:** Increasing trends of HCV infection have been reported among HIV-positive men who have sex with men (MSM) in Europe, Australia and North America. The trends of recently acquired HCV infection among HIV-positive MSM are less clear in Asia–pacific region.

**Methods:** All HIV-positive patients seeking care at a university hospital in Taiwan tested for anti-HCV IgG at least once annually to estimate the incidence of HCV seroconversion during 2011–2018. HCV genotyping and sequencing were performed and multivariate logistic regression analysis was conducted to identify the factors associated with HCV seroconversion among MSM.

**Findings:** During the study period, 3495 HCV-seronegative patients (86.4% MSM) were included and 294 (8.4%) with recent HCV infection were identified, in whom 281 (95.6%) were MSM, during a total of 16,361.86 person-years of follow-up (PYFU), giving an overall incidence rate of 17.97 per 1000 PYFU, which increased from 14.28 per 1000 PYFU in 2011 to 25.38 per 1000 PYFU in 2018 ( $p < 0.001$ ). HCV seroconversion among MSM was associated with aspartate aminotransferase  $\geq 37$  U/L (adjusted odds ratio [AOR] 7.50, 95% CI 4.17–13.50), alanine aminotransferase  $\geq 41$  U/L (AOR 7.47, 95% CI 4.11–13.58), and syphilis acquisition (AOR 2.88, 95% CI 1.67–4.97). Among the 277 (94.2%) with HCV viremia, genotype 2a ( $n = 116$ ) was the leading genotype, followed by 1b ( $n = 85$ ), 6a ( $n = 34$ ), and 1a ( $n = 21$ ). Genotypes 3a and 6a increased from 0% and 5.2%, respectively, in 2011–2014 to 4.1% and 17.1% in 2015–2018. Phylogenetic analysis revealed increased clusters in genotypes 2a, 3a and 6a from 2011–2014 to 2015–2018.

**Interpretation:** An expanding HCV epidemic among HIV-positive MSM is occurring in Taiwan. Improving access to HCV testing and early linkage to treatment are needed to curb the expanding HCV epidemic.

**Funding:** This research was supported by a grant from National Taiwan University Hospital, Taipei, Taiwan (NTUH.106-003347 to Hsin-Yun Sun).

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### 1. Introduction

Hepatitis C virus (HCV) and HIV infections are two major global public health problems with overlapping modes of transmission [1]. UNAIDS estimated that, in 2018, about 37.9 million people were

living with HIV and 1.7 million were newly infected with HIV [2]. Moreover, WHO estimated that about 71 million people were infected with HCV and approximately 2.75 million people living with HIV were co-infected with HCV globally in 2017. The overall HCV seroprevalence is estimated 4.0% (interquartile range [IQR], 1.2–8.4) among HIV-positive heterosexuals and pregnant women, 6.4% (IQR, 3.2–10.0) among men who have sex with men (MSM), and 82.4% (IQR, 55.2–88.5) among injection drug users (IDUs) worldwide [1]. In Asia-Pacific region, the prevalence of HCV/HIV coinfection ranges from 3.8% in Singapore to 42.6% in Nepal [3,4].

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## Research in context

### *Evidence before this study*

We searched PubMed for cohort studies published up to December 31, 2019, with combinations of the search terms “HCV”, “seroconversion”, “HIV”, “chemsex”, and “sexually transmitted infections” with restrictions to English language and publication date. Sexually transmitted HCV infection remains a major public health concern globally, with crucially significant disease burden in HIV-positive men who have sex with men (MSM). In 2012, we first demonstrated the increasing incidence of recent HCV infections among HIV-positive MSM in Taiwan from 1994 to 2010. Moreover, we recently also observed a high incidence of HCV reinfection (8.2 per 100 person-years of follow-up) among HIV-positive Taiwanese. However, long-term data are sparse in the literature on the recent epidemic of sexually transmitted HCV infection in Asia-Pacific region.

### *Added value of this study*

Our study reveals that the epidemic of sexually acquired HCV had continued to expand among MSM in Taiwan between 2011 and 2018. The findings that the proportions and clusters of genotypes 1a, 3a, and 6a increased significantly in recent years suggest the network of HCV transmission might have involved HIV-positive MSM and IDUs.

### *Implications of all the available evidence*

In 2016, World Health Assembly (WHA) approved the Global Health Sector Strategy to eliminate HCV infection by 2030, and the government of Taiwan implemented fully-reimbursed direct-acting antiviral (DAA) program for people with HCV in 2017 and ambitiously aims to eliminate HCV infection by 2025. The findings of increasing trends of recently acquired HCV infection among HIV-positive patients in our current study implies that further improvement is needed in the access to HCV testing and expedited linkage to DAA program for people with HCV.

In this study, we aimed to investigate the trends of recently acquired HCV infection and distribution of HCV genotypes and to identify factors associated with incident HCV infections among HIV-positive patients in Taiwan between 2011 and 2018.

## 2. Methods

### 2.1. Study setting and patient population

HIV-positive patients are provided with free-of-charge HIV care, including plasma HIV RNA load (PVL) and CD4 lymphocyte count testing and combination antiretroviral therapy (cART), at designated hospitals around Taiwan. Testing for PVL and CD4 count was performed at baseline, one month after cART initiation, and subsequently every three months during the first year of follow-up, and subsequently every six months among those on stable cART with viral suppression. Serological assessment for HCV infection is recommended on an annual basis or when elevations of aminotransferases or sexually transmitted infections (STIs) are detected according to the national HIV treatment guidelines.

We retrospectively reviewed the medical and laboratory records of all HIV-positive patients seeking care at the National Taiwan University Hospital (NTUH) between 2011 and 2018. Patients testing anti-HCV-negative at baseline were included in this cohort study. Owing to the fact that not all patients were provided with anti-HCV testing at baseline or during the follow-up, we retrieved archived blood samples for HCV testing of HIV-positive patients who tested seronegative for HCV at baseline. Patients testing HCV-seropositive at baseline, those aged less than 18 years, and those without follow-up were excluded.

### 2.2. Data collection and laboratory investigations

A case record form was used to collect the information on demographics, risk group for HIV transmission, cART, and sequential laboratory data, which included CD4 count, hepatitis B surface antigen (HBsAg), aspartate aminotransferase (AST), alanine aminotransferase (ALT), total bilirubin (TBIL), and rapid plasma reagin (RPR) titre.

Antibodies to HCV were determined with the use of a fourth-generation enzyme immunoassay (Dia.Pro Diagnostic Bioprobes Srl, Italy). The seropositive specimens were further confirmed by detection of HCV RNA (COBAS® AmpliPrep HCV Test, v2.0, Roche, USA). HCV-seropositive patients with undetectable HCV RNA were confirmed to have HCV seroconversion using a recombinant immunoblot assay (RIBA) kit (Mikrogen Diagnostik, Neureid, Germany).

A 366-bp fragment covering partial HCV NS5B (nucleotides 8294 to 8629 relative to HCV reference strain H77) was amplified by polymerase-chain reaction (PCR). Phylogenetic analysis was performed to determine the HCV genotypes with the use of NS5B sequences amplified from HCV-infected patients in our cohort [18]. Phylogenetic analysis of genotypes and reference sequences were aligned using the Clustal W program with minor manual adjustment. The tree was constructed by the neighbor-joining method based on the Kimura 2 parameter distance matrix listed in MEGA software (version 6.0).

### 2.3. Matched cohort study to identify associated factors

A retrospective matched cohort study was performed to compare the clinical characteristics between HCV seroconverters and non-seroconverters. HCV seroconverters were identified as case patients, and four patients without HCV seroconversion who had a similar follow-up duration ( $\pm 3$  months) between the study entry and the time-point of HCV seroconversion to that of case patients were selected as controls. Because the majority of our included patients were MSM, the analysis was restricted to MSM. Clinical characteristics and laboratory data included for analysis were those determined within three

In early 1990s, parenteral exposure to contaminated blood or blood products was the main transmission route of HCV infection. With implementation of harm reduction programs, decreases of HCV infection among IDUs have been observed in several developed countries [5,6]. In contrast, an expanding epidemic of sexually transmitted HCV infection has been reported over the past two decades [7,8]. In several European countries and Australia, HCV continues to spread among HIV-positive MSM, with the interval from confirmed HIV infection to incident HCV infection significantly decreasing over time [7,9,10]. In the United States, new cases of HCV infection reported to the Centers for Disease Control and Prevention have increased since 2010, particularly among the individuals aged 20 to 39 years, after significant declines of HCV transmission through injection drug use, medical practices, and transfusion from 1990 to 2010 [11]. In France, the incidence of first HCV infection increased among HIV-positive MSM between 2012 and 2016 [12]; moreover, HCV appears to have spread from HIV-positive MSM to HIV-negative MSM because of shared risk factors such as injection drug use, nasal drug use, fisting, chemsex, and traumatic sexual practices [13]. The trends of HCV infection among HIV-positive patients, however, remain less investigated in Asia-Pacific region that bears one-fifth of the global HCV burden [1]. While a recent study from Jiangsu, China did not reveal an increasing trend of HCV infection among MSM [14], several studies have demonstrated increasing trends of HCV infection among HIV-positive MSM in Hong Kong [15], Tokyo [16], and Taipei [17,18].

months prior to or later than the estimated date of HCV seroconversion and during the last follow-up in matched HCV non-seroconverters with similar observation durations.

#### 2.4. Definitions

In this study, syphilis acquisition was defined as occurrence of a 4-fold or greater increase of RPR titers from baseline or presentation of consistent clinical symptoms, for which treatment of syphilis was administered within 90 days of incident HCV infection. HCV seroprevalence was defined as HCV seropositivity within three months of entry into HIV care. HIV-positive patients were included for follow-up of HCV seroconversion if they tested negative for anti-HCV antibodies within the three months of entry into care. Plasma HCV RNA was determined in HCV-seropositive patients, and patients with undetectable HCV RNA were further confirmed by RIBA in order to estimate the annual incidence rate of HCV seroconversion. Recent HCV seroconversion occurred at the first positive anti-HCV detected within one year after the last negative anti-HCV. The date of HCV seroconversion was defined as the midpoint between the last date of HCV seronegativity and the first date of seropositivity. The annual follow-up duration started from three months after entry into care until loss to follow-up, death, or HCV seroconversion with back-testing for anti-HCV and/or HCV RNA, whichever occurred first within the first three months of the next calendar year. The study started from 1 January, 2011 and ended on 31 March, 2019. Recently acquired HCV infection was defined as the first positive anti-HCV detected within 12 months after the last negative anti-HCV.

#### 2.5. Statistical analysis

Comparisons of categorical variables were performed by  $\chi^2$  analysis or Fisher's exact test and comparisons of continuous variables by Student's *T* test or Mann–Whitney *U* test and Wilcoxon signed-rank test for paired samples. The incidence rate of HCV infection was calculated as the number of HCV seroconversion per 1000 person-years of follow-up (PYFU). Variables with *P* value <0.05 in univariate analysis were entered into multivariate logistic regression analysis to identify the clinical characteristics associated with HCV seroconversion in the retrospective matched cohort study. The crude and odds ratios (OR) and their 95% confidence interval (95% CI) were calculated to express the magnitude of association. Statistical analyses were performed using SPSS software, version 21.0 (SPSS Inc., Chicago, IL) and Excel (Microsoft Office 2010). All analyses were two-tailed, and a *P* value <0.05 was considered statistically significant.

#### 2.6. Ethical oversight

The study was approved by the National Taiwan University Hospital Research Ethics Committee (registration number, 201605103RINC and 201605128RINC)

#### 2.7. Role of funding source

The funding source had no role in designing the trial, collecting data, conducting the analysis, or interpreting data. All authors had full access to the data and are responsible for the veracity and completeness of the reported data. The corresponding author had final responsibility for the decision to submit for publication.

### 3. Results

#### 3.1. Clinical characteristics of the study population

During the 8-year study period, 3495 HIV-positive patients who tested seronegative for HCV at study entry were included for

investigation of the incidence of HCV seroconversion. Overall, 306 (8.8%) patients tested HCV-seropositive during the follow-up; and, among these patients, 29 (9.5%) had undetectable HCV RNA. After confirmation of the HCV antibody responses by RIBA, 17 cases of HCV seroconversion were considered resolved HCV infection and 12 cases were considered false-reactive response (Fig. 1); therefore, 294 (8.4%) patients developed recent HCV seroconversion with 277 (94.2%) having HCV viremia. The great majority of the included patients (96.6%) were male and the median age was 31.1 years (IQR, 26.2–38.1). Most of the patients (86.4%) were MSM, 295 (8.5%) were heterosexuals, and 22 (0.6%) were IDUs. The median last follow-up CD4 count was 570 cells/ $\mu$ L (IQR, 421–741). The percentage of syphilis acquisition was 45.0%. The prevalence of HBsAg seropositivity was 13.0% at study entry. At the last follow-up, AST  $\geq$ 37 U/L, ALT  $\geq$ 41 U/L, and TBil  $\geq$ 1.2 mg/dL occurred in 14.0%, 17.7% and 8.9% of the patients, respectively (sTable 1).

#### 3.2. Seroincidence of HCV

During a total observation duration of 16,361.86 PYFU, an overall incidence rate of recent HCV infection was 17.97 per 1000 PYFU (95% CI, 15.91–20.02), which increased significantly from 14.28 per 1000 PYFU (95% CI, 8.17–20.39) in 2011 to 25.38 per 1000 PYFU (95% CI, 19.06–31.70) in 2018 (*p*<0.001) (Fig. 2). Because the great majority of our included patients were male, we re-examined the trends of HCV incidence only among MSM. After exclusion of 474 non-MSM, the overall incidence rate of recent HCV infection was 19.20 per 1000 PYFU (95% CI, 16.95–21.44) among MSM, which increased significantly from 16.42 per 1000 PYFU (95% CI 9.22–23.61) in 2011 and 28.10 per 1000 PYFU (95% CI 20.99–35.20) in 2018 (*p*<0.001) (sFig. 1). Similarly, the HCV seroprevalence significantly increased from 11.5% (95% CI 10.1–12.9) in 2011 to 15.7% (95% CI 14.4–17.0) in 2018 (sFig. 2), with the overall seroprevalence being 12.5% (95% CI 11.6–13.4) in 2011–2018.

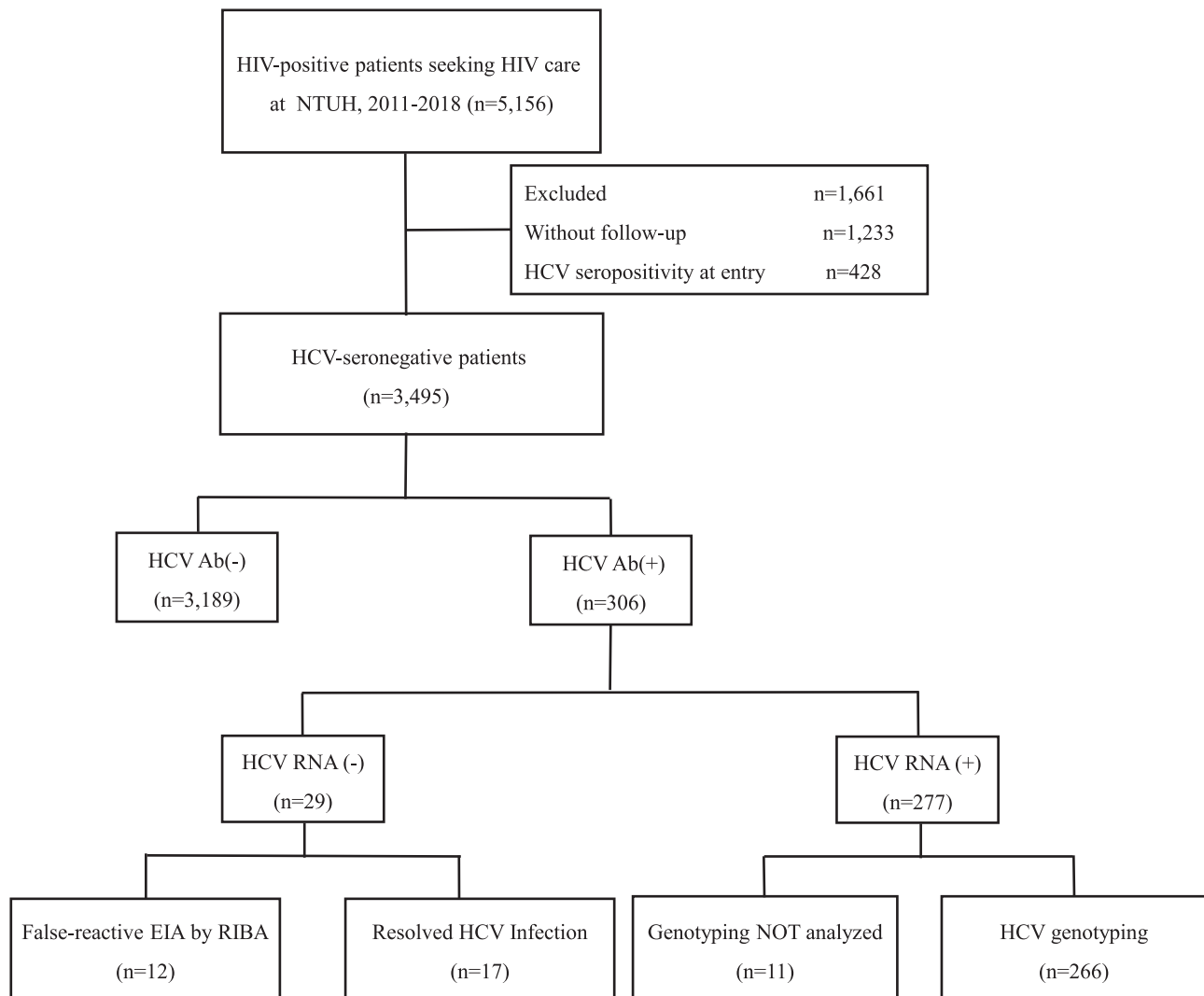
#### 3.3. Clinical characteristics associated with of HCV seroconversion among MSM

In the matched cohort study, 1124 non-seroconverters matched for observation durations were identified as controls for 281 case patients who had incident HCV seroconversion. Compared with non-seroconverters, 281 HCV seroconverters were significantly younger at the initiation for anti-HCV screening (median [IQR], 28.7 [25.2–33.9] vs 30.7 [26.2–36.9] years) and at the estimated time-point of HCV seroconversion (33.9 [29.2–39.7] vs 36.1 [30.2–43.3] years), and were more likely to have elevated RPR titers at the study entry (33.7% vs 21.0%), to acquire syphilis acquisition during the follow-up (34.9% vs 11.2%), and to have elevated aminotransferases (AST  $\geq$ 37 U/L, 75.0% vs 9.1% and ALT  $\geq$ 41 U/L, 81.7% vs. 14.4%) (all comparisons, *P* <0.001). There were no statistically significant differences between the two groups in terms of CD4 counts, HBsAg seropositivity, and TBil  $\geq$ 1.2 mg/dL at study entry or at the estimated time-point of HCV seroconversion (Table 1).

Compared with non-HCV seroconverters, HCV seroconverters were younger at initiation of anti-HCV screening (per 1-year increase, adjusted odds ratio [AOR] 0.99, 95% CI 0.93–1.06) and at the estimated time-point of HCV seroconversion (AOR 0.96, 95% CI 0.90–1.01). In contrast, HCV seroconverters were more likely to present with AST  $\geq$ 37 U/L (AOR 7.50, 95% CI 4.17–13.50), ALT  $\geq$ 41 U/L (AOR 7.47, 95% CI 4.11–13.58), and to acquire syphilis during the follow-up (AOR 2.88, 95% CI 1.67–4.97) than non-seroconverters (all comparison *P*<0.001) (Table 2).

#### 3.4. HCV genotype distribution and phylogenetic analysis

Of the 277 HCV viremic specimens subjected to PCR for genotyping, 11 were not analyzed mainly due to low HCV RNA loads. The



**Fig. 1. Flow chart.** NTUH, National Taiwan University Hospital; RIBA, recombinant immunoblot assay.

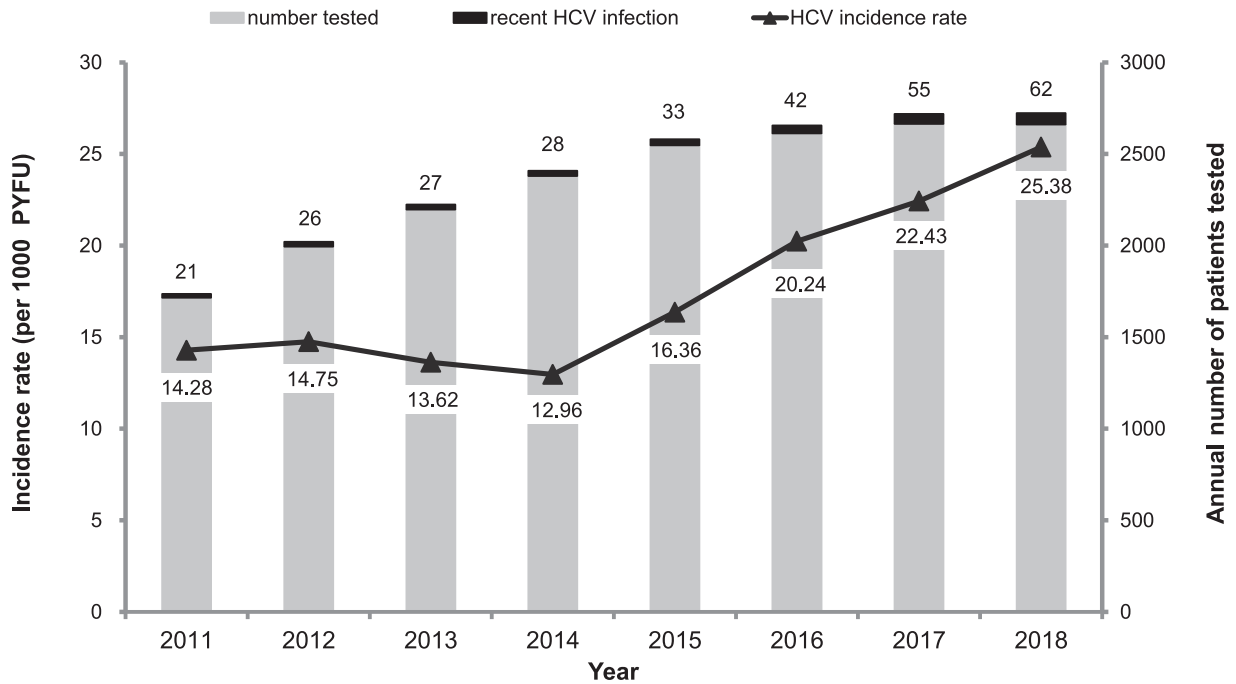
most common HCV genotype was 2a ( $n = 116$ , 41.9%), followed by 1b ( $n = 85$ , 31.7%), 6a ( $n = 34$ , 12.3%), 1a ( $n = 21$ , 7.6%), 3a ( $n = 7$ , 2.5%), and 6n ( $n = 3$ , 1.0%) (sFig. 3). With two study periods being defined (2011–2014 vs. 2015–2018), we found that the proportion of genotypes 1a, 3a, and 6a increased significantly from 3.1%, 0%, and 5.2%, respectively, in 2011–2014 to 10.6%, 4.1%, and 17.1% in 2015–2018 (all comparisons,  $P < 0.05$ ) (sFig. 4). In the phylogenetic analysis, large clusters were mainly observed in the most prevalent HCV genotypes 1b and 2a. During 2011–2018, nine independent clusters and one pair belonging to four genotypes (genotypes 1, 2, 3, and 6) were identified, including four clusters within genotype 2a, two clusters within genotype 1b and 3a, one cluster within genotype 6a. In addition, only one transmission pair was observed within genotype 6a (Table 3). Almost all clusters occurred among MSM, except for three patients with unknown sexual orientation and one heterosexual within genotype 2a. Furthermore, the changes of cluster size between 2011 and 2018 were also observed. In genotype 1b, the number of sequences observed in each cluster was four and nine sequences in 2011–2014, respectively, which increased to seven and 16 sequences, respectively, in 2015–2018 (Fig. 3a). In genotype 3a, no cluster of sequences was identified in 2011–2014 and two clusters were observed in 2015–2018 containing three and four sequences, respectively (Fig. 3b). In genotype 6a, only one transmission pair of sequences was identified in 2011–2014, which evolved to a cluster containing ten sequences in total. Moreover, a new transmission pair was

observed within genotype 6a in 2015–2018 (Fig. 3c). In genotype 2a, one pair and one cluster containing two and 18 sequences, respectively, were observed during 2011–2014, and which increased to four clusters totally with 24, four, three, and ten sequences, respectively (Fig. 3d and Table 3).

#### 4. Discussion

In this retrospective cohort study consisting mainly of HIV-positive MSM in Taiwan, we demonstrate significantly increasing trends of recent HCV seroconversion during an 8-year study period. With an overall incidence rate of 17.97 per 1000 PYFU of the cohort, the annual incidence rate had increased from 14.28 per 1000 PYFU in 2011 to 25.38 per 1000 PYFU in 2018. While the association between HCV seroconversion within the past 12 months with syphilis acquisition suggests HCV transmission was sexually transmitted, the increasing number and size of clusters highlight the epidemic of HCV was expanding among HIV-positive MSM in our cohort.

The findings of increasing trends of recently acquired HCV infection in our current study extend those of our previous study, in which the incidence rate of recent HCV infection significantly increased from 0 in 1994–2000 and 2.29 per 1000 PYFU in 2001–2005 to 10.13 per 1000 PY in 2006–2010 [17]. Moreover, the overall HCV seroprevalence in this study almost doubled, 12.5% (631/5065; 95% CI 11.6–13.4) in 2011–2018 (sFig. 2) compared with 6.7% (138/



**Fig. 2.** Incidence rates of HCV infection among HIV-positive patients from 2011 to 2018. Gray bar indicates the annual number of tests performed; black square indicates the annual number of HCV seroconversion; and black line and triangle indicate the trend of annual incidence rates of HCV seroconversion (per 1000 person-years of follow-up).

2507; 95% CI 5.7–7.7) in 1994–2010 [17]. The great majority of our patients with recent acquisition of HCV infection were MSM, which is similar to the situation of HCV infection observed among HIV-positive MSM in Western Europe, Australia and North America since 2000 [9]. Recent studies of HIV-positive MSM in San Diego and France

also indicate an increase of HCV incidence in 2000–2015 and 2012–2015, respectively [12,19]. A continuing HCV epidemic is also occurring among HIV-positive MSM in the United Kingdom [20]. On the contrary, with the unrestricted access to DAA treatments in the Netherlands, a 51% decrease in acute HCV infections was observed

**Table 1**

Comparisons of clinical characteristics between HCV seroconverters and non-seroconverters among HIV-positive MSM in matched cohort study.

Characteristics	HCV seroconverters	HCV non-seroconverters	P value
Patient number, N=	281	1124	
Age at the initiation for anti-HCV screening, median (IQR), years	28.7 (25.2–33.9)	30.7 (26.2–36.9)	<0.001
Age at the estimated time-point of HCV seroconversion, median (IQR), years	33.9 (29.2–39.7)	36.1 (30.2–43.3)	<0.001
CD4 at baseline, median (IQR), cells/ $\mu$ L, N	416 (238–576) (276)	404 (236–563) (1107)	0.232
CD4 at baseline, cells/ $\mu$ L, n (%)			
<200	55 (19.9)	231 (20.9)	0.793
200–500	126 (45.7)	493 (44.5)	0.790
>500	95 (34.4)	383 (34.6)	>0.99
CD4 at the estimated time-point of HCV seroconversion, median (IQR), cells/ $\mu$ L, N	587 (431–731) (263)	561 (410–747) (1103)	0.470
CD4 at the estimated time-point of HCV seroconversion, cells/ $\mu$ L, n (%)			
<200	10 (3.8)	65 (5.9)	0.235
200–500	82 (31.2)	363 (32.9)	0.642
>500	171 (65.0)	675 (61.2)	0.282
Baseline HBsAg-positivity, n/N (%)	34/271 (12.5)	141/1085 (13.0)	0.923
Latest HBsAg-positivity, n/N (%)	28/262 (10.7)	62/448 (13.8)	0.271
Baseline RPR $\geq$ 1:4, n/N (%)	90/267 (33.7)	214/1017 (21.0)	<0.001
Syphilis acquisition, n/N (%)	95/272 (34.9)	121/1081 (11.2)	<0.001
<b>Liver function tests at baseline</b>			
AST, median (IQR), U/L, (N)	23 (18–29) (221)	23 (19–31) (849)	0.348
AST $\geq$ 37 U/L, n (%)	35 (15.8)	140 (16.5)	0.895
ALT, median (IQR), U/L, (N)	21 (15–35) (210)	23 (16–35) (797)	0.162
ALT $\geq$ 41 U/L, n (%)	36 (17.1)	161 (20.2)	0.370
TBil, median (IQR), mg/dL, (N)	0.58 (0.41–0.79) (193)	0.59 (0.45–0.85) (728)	0.215
TBil $\geq$ 1.2 mg/dL, n (%)	17 (5.8)	91 (12.5)	0.197
<b>Liver function tests the estimated time-point of HCV seroconversion</b>			
AST, median (IQR), U/L, (N)	69.5 (37–167) (228)	21 (18–27) (839)	<0.001
AST $\geq$ 37 U/L, n (%)	171 (75.0)	76 (9.1)	<0.001
ALT, median (IQR), U/L, (N)	131 (56–355) (251)	22 (15–33) (882)	<0.001
ALT $\geq$ 41 U/L, n (%)	205 (81.7)	127 (14.4)	<0.001
TBil, median (IQR), mg/dL, (N)	0.75 (0.6–1.2) (184)	0.63 (0.47–0.97) (588)	0.164
TBil $\geq$ 1.2 mg/dL, n (%)	45 (24.3)	111 (18.9)	0.124

N, total number of patients; n, number of patients with indicated result.

Syphilis acquisition was defined as occurrence of a 4-fold or greater increase of rapid plasma reagin (RPR) titer within 90 days of an incident HCV seroconversion.

**Abbreviations:** ALT, alanine aminotransferase; AST, aspartate aminotransferase; HBsAg, hepatitis B virus surface antigen; IQR, interquartile range; MSM, men who have sex with men; TBil, total bilirubin.

**Table 2**  
Multivariate analysis for the clinical characteristics associated with HCV seroconversion in matched cohort study.

Variables	Reference	Adjusted odds ratio (95% CI)	P value
Age at the initiation of anti-HCV screening	Per 1-year increase	0.99 (0.93–1.06)	0.850
Age at the estimated time-point of HCV seroconversion	Per 1-year increase	0.96 (0.90–1.01)	0.140
AST $\geq$ 37 U/L	AST <37 U/L	6.13 (3.26–11.51)	<0.001
ALT $\geq$ 41 U/L	ALT <41 U/L	6.85 (3.72–12.61)	<0.001
Syphilis acquisition	No syphilis acquisition	2.83 (1.63–4.90)	<0.001

**Abbreviations:** ALT, alanine aminotransferase; AST, aspartate aminotransferase.

among HIV-positive MSM [21], suggesting that, similar to treatment as prevention in the control of HIV epidemic, expanding harm reduction services to incorporate HCV screening and early diagnosis of HCV infection in combination with making DAA available to all HIV/HCV-coinfected patients to decrease the community HCV viral load will be of vital importance in the micro-elimination of HCV among HIV-positive subpopulation [22–24].

HCV seroconversion of HIV-positive patients in our study almost exclusively occurred among MSM, and syphilis acquisition was significantly associated with HCV seroconversion, suggesting that HCV transmission was facilitated by concurrent sexually transmitted infections (STIs) or the behaviors that might increase the risk of acquiring STIs, such as unprotected anal sex and chemsex [8,25]. HCV can be detected in the rectal fluid, and traumatic sex and prolonged sexual contact with HCV-infected rectal fluid may increase the risk for HCV transmission [26]. The findings of our matched cohort study, similar to those of our previous investigation of HCV reinfections after viral clearance among HIV-positive patients [27], have important implications in terms of prevention and testing strategies for HCV transmission. DAA treatments had not been reimbursed by the National Health Insurance, Taiwan, until 2017, before which time patients with chronic HCV infection had to be referred to hepatologists for treatment with pegylated interferon and ribavirin. In January, 2019, HIV-positive patients with chronic HCV infection could be treated with DAAs by their HIV-treating physicians. The restriction on access to reimbursed DAA treatments for patients confirmed to have HCV viremia for less than six months was lifted in June 2019. Therefore, other than promotion of safe sex, and counseling and support to quit using illicit drugs, early detection and treatment of STIs should be incorporated with HCV testing and linkage to DAA treatments to reduce the risk of onward transmission of HCV [28].

Recent molecular investigations of HCV infection have shown that there were MSM-specific HCV lineages and clusters [8] and the most prevalent HCV genotype was 3a, 1b, and 2a in Hong Kong, Tokyo, and

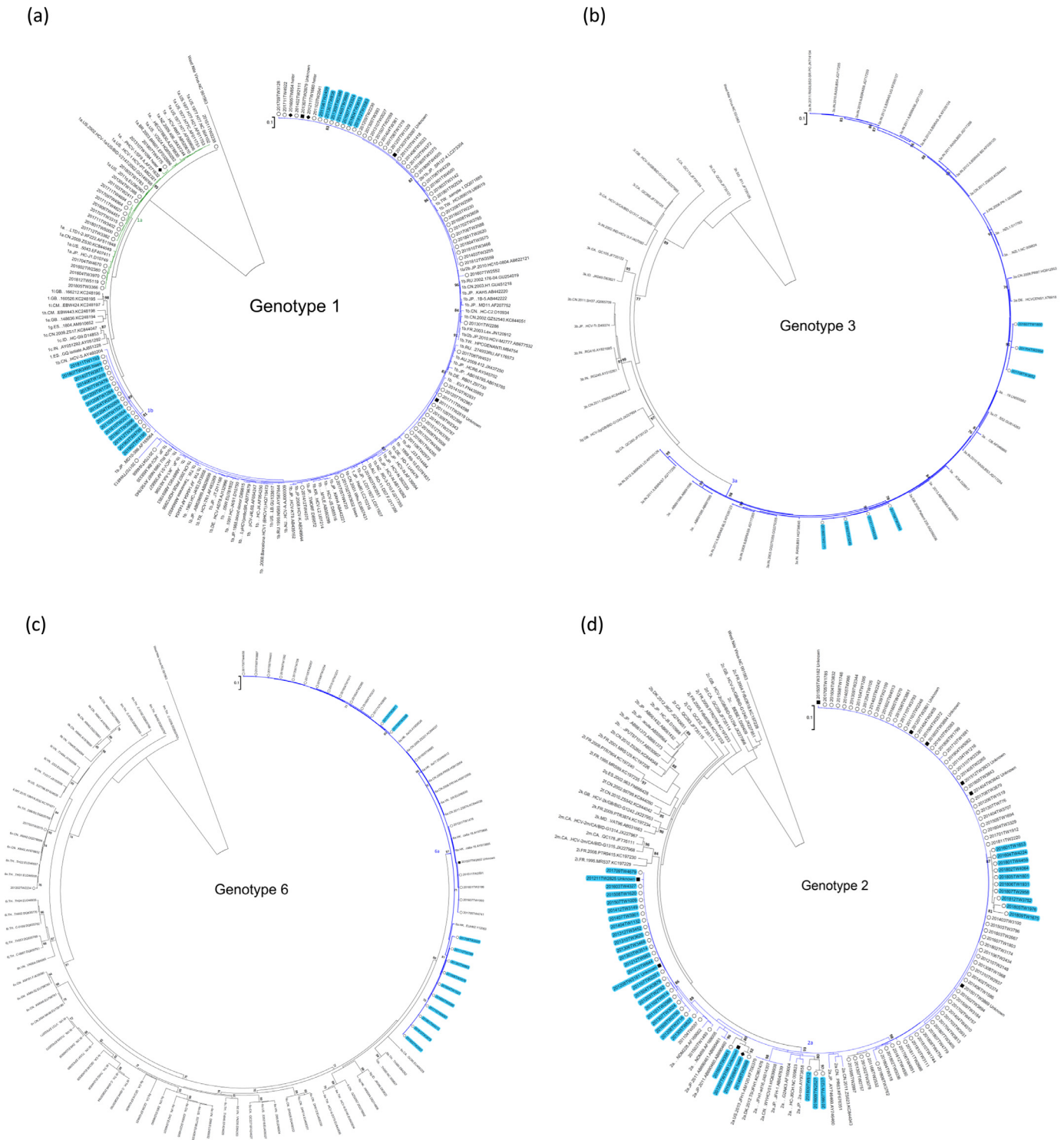
Taipei, respectively [18]. In this study between 2011 and 2018, we found that genotype 2a (39.5%) continued to predominate among our HIV-positive MSM with recent HCV infection, followed by 1b (28.9%), 6a (11.6%), 1a (7.1%), 3a (2.4%), and 6n (1.0%). This contrasts with the studies in Hong Kong and Tokyo, where genotype 3a and 1b was predominant in Hong Kong and Tokyo, respectively [15,16]. Of note, compared with our previous study, we found that genotypes 1a, 1b, 2a, and 6a were increasing dramatically in recent years [17]. Genotype 1a increased from 3 cases (3.1%, 3/96) in 2011–2014 to 18 cases (10.6%, 18/170) in 2015–2018, genotype 3a from zero during 2011–2014 to 7 cases during 2015–2018 and genotype 6a from 5 during 2011–2014 to 29 cases during 2015–2018 (sFig. 4). In addition, almost all clusters occurred among MSM (Fig. 3). In Australia, HCV genotypes 1a and 3a from HIV-positive MSM showed a strong link with those circulating among local IDUs [29]. While previous studies showed genotype 6a was found mainly among IDUs in southern China and Taiwan [30,31], and genotype 6n is one of most common subtypes among IDUs in Thailand [32]. In Japan, the phylogenetic analysis of the epidemiological status of HCV transmission indicated it is possible that MSM and IDU with high risk of infection change their sexual partners and drug-using groups, resulting in the spread of the viruses to a wider range of high-risk groups [33]. Hence, we postulate that the clusters in our study might present the possibility of cross-transmission between IDUs and MSM through sharing the injection devices or sex contacts and these factors probably play a role in the circulation of the virus within this population.

There are several limitations to our study. First, the study was conducted at one single institution and the risk behavioral data of patients included in the study, though predominantly MSM, were extremely scanty. Our findings may not be generalized to other settings that mainly included heterosexuals or IDUs. Second, our estimates of incidence rate of HCV seroconversion could be biased by the frequency and targeting of the HCV testing. Adherence to the national guidelines for HCV screening may vary, and HIV-treating clinicians may be prone to adopting symptom-driven testing strategies, which might not be sensitive enough to identify HCV transmission in the key populations [27]. Despite a high association between syphilis acquisition and HCV seroconversion, the testing strategy that was prompted by recent STIs might still miss a substantial proportion of HCV infection [27]. More studies are warranted to identify cost-effective testing strategies among the high-risk groups for HCV transmission. While all included patients were assumed to have acquired HCV sexually, we cannot exclude that some cases of HCV infection might have occurred through other routes than sex contacts. Further epidemiologic investigations are needed to provide insights into specific behavioral factors that may increase the risk for HCV transmission among MSM in Taiwan.

In conclusion, the increasing trends of recent HCV seroconversion and the number and size of clusters in the phylogenetic analyses suggest that epidemic of recently acquired HCV infection among HIV-

**Table 3**  
The number of HCV sequences in each cluster and pair among HIV-positive patients included in phylogenetic analysis.

Genotypes	2011–2014		2015–2018		2011–2018	
	Cluster (no. of sequences in each cluster)	Pair	Cluster (no. of sequences in each cluster)	Pair	Cluster (no. of sequences in each cluster)	Pair
1b	2 (4;9)	0	2 (3;7)	0	2 (7;16)	0
3a	0	0	2 (3;4)	0	2 (3;4)	0
6a	0	1 (2)	1 (8)	1 (2)	1 (10)	1 (2)
2a	1 (18)	1 (2)	3 (6;3;10)	1 (2)	4 (24;4;3;10)	0



**Fig. 3. Phylogenetic analysis of HCV identified among HIV-positive patients with recent HCV infection.** Phylogenetic tree analysis was conducted using HCV sequences derived from HCV seroconverters detected in genotypes 1 (a), 3 (b), 6 (c), and 2 (d). Nine HCV transmission clusters and one transmission pair were identified. The circles represent men who have sex with men patients, the filled diamonds represent heterosexual patients and the filled squares represent patients with unknown risk behaviors. The partial NS5B sequences were PCR-amplified from a total of 266 plasma specimens. The derived NS5B partial sequences were used to construct the phylogenetic trees. The clusters/pairs observed in genotypes, 1b, 2a, 3a, and 6a were labeled in blue. The horizontal branch was drawn in accordance with their relative genetic distances. Bootstrap values greater than 700 of 1000 replicates are considered significant and indicated at the nodes of the corresponding branches. Outlier is West Nile virus.

positive MSM is expanding in Taiwan. Our findings highlight that HCV control strategies consisting of efficient detection of incident HCV infections and linkage to DAA care are urgently needed among HIV-positive MSM.

**Declaration of Competing Interest**

Chien-Ching Hung received research support from Gilead Sciences, Merck, and ViiV Healthcare and speaker honoraria from Gilead

Sciences and ViiV Healthcare and served on the advisory boards for Gilead Sciences and ViiV Healthcare. Other authors have no conflicts of interest to report.

## Funding

This research was supported by a grant from National Taiwan University Hospital, Taipei, Taiwan ([NTUH.106-003347](https://doi.org/10.1016/j.eclinm.2020.100441) to Hsin-Yun Sun).

## Supplementary materials

Supplementary material associated with this article can be found in the online version at doi:[10.1016/j.eclinm.2020.100441](https://doi.org/10.1016/j.eclinm.2020.100441).

## References

- [1] Platt L, Easterbrook P, Gower E, et al. Prevalence and burden of HCV co-infection in people living with HIV: a global systematic review and meta-analysis. *Lancet Infect Dis* 2016;16(7):797–808.
- [2] <http://www.unaids.org/en/resources/fact-sheet>. UNAIDS Global Statistics. 2018.
- [3] Li CW, Yang CJ, Sun HY, et al. Changing seroprevalence of hepatitis C virus infection among HIV-positive patients in Taiwan. *PLoS One* 2018;13(3):e0194149.
- [4] Martinello M, Amin J, Matthews GV, Dore GJ. Prevalence and disease burden of HCV coinfection in HIV cohorts in the asia pacific region: a systematic review and meta-analysis. *AIDS Rev* 2016;18(2):68–80.
- [5] de Vos AS, van der Helm JJ, Matser A, Prins M, Kretzschmar ME. Decline in incidence of HIV and hepatitis C virus infection among injecting drug users in Amsterdam; evidence for harm reduction. *Addiction* 2013;108(6):1070–81.
- [6] Serrano-Villar S, Sobrino-Vegas P, Monge S, et al. Decreasing prevalence of HCV coinfection in all risk groups for HIV infection between 2004 and 2011 in Spain. *J Viral Hepat* 2015;22(5):496–503.
- [7] Boesecke C, Grnt D, Soriano V, et al. Hepatitis C seroconversions in HIV infection across Europe: which regions and patient groups are affected. *Liver Int* 2015;35(11):2384–91.
- [8] Chan DP, Sun HY, Wong HT, Lee SS, Hung CC. Sexually acquired hepatitis C virus infection: a review. *Int J Infect Dis* 2016;49:47–58.
- [9] van de Laar T, Pybus O, Bruisten S, et al. Evidence of a large, international network of HCV transmission in HIV-positive men who have sex with men. *Gastroenterology* 2009;136(5):1609–17.
- [10] van Santen DK, van der Helm JJ, Del Amo J, et al. Lack of decline in hepatitis C virus incidence among HIV-positive men who have sex with men during 1990–2014. *J Hepatol* 2017;67(2):255–62.
- [11] Shiffman ML. The next wave of hepatitis C virus: the epidemic of intravenous drug use. *Liver Int* 2018;38(Suppl 1):34–9.
- [12] Pradat P, Huleux T, Raffi F, et al. Incidence of new hepatitis C virus infection is still increasing in French MSM living with HIV. *AIDS* 2018;32(8):1077–82.
- [13] Charre C, Cotte L, Kramer R, et al. Hepatitis C virus spread from HIV-positive to HIV-negative men who have sex with men. *PLoS One* 2018;13(1):e0190340.
- [14] Shen L, Liu X, Fu G, et al. The epidemic of human immunodeficiency virus, hepatitis C virus, and syphilis infection, and the correlates of sexually transmitted infections among men who have sex with men in Zhenjiang, Jiangsu, China. *Jpn J Infect Dis* 2017;70(2):171–6.
- [15] Lin AW, Wong KH, Chan K. More safer sex intervention needed for HIV-positive MSM with higher education level for prevention of sexually transmitted hepatitis C. *J Int AIDS Soc* 2014;17(4 Suppl 3):19663.
- [16] Nishijima T, Shimbo T, Komatsu H, Hamada Y, Gatanaga H, Oka S. Incidence and risk factors for incident Hepatitis C infection among men who have sex with men with HIV-1 infection in a large Urban HIV clinic in Tokyo. *J Acquir Immune Defic Syndr* 2014;65(2):213–7.
- [17] Sun HY, Chang SY, Yang ZY, et al. Recent hepatitis C virus infections in HIV-infected patients in Taiwan: incidence and risk factors. *J Clin Microbiol* 2012;50(3):781–7.
- [18] Sun HY, Uemura H, Wong NS, et al. Molecular epidemiology of acute HCV infection in HIV-positive patients from Hong Kong, Taipei, Tokyo. *Liver Int* 2019;39(6):1044–51.
- [19] Chaillon A, Sun X, Cachay ER, et al. Primary incidence of hepatitis C virus infection among HIV-infected men who have sex with men in San Diego, 2000–2015. *Open Forum Infect Dis* 2019;6(4):ofz160.
- [20] Martin NK, Thornton A, Hickman M, et al. Can Hepatitis C Virus (HCV) direct-acting antiviral treatment as prevention reverse the HCV epidemic among men who have sex with men in the United Kingdom? Epidemiological and modeling insights. *Clin Infect Dis* 2016;62(9):1072–80.
- [21] Boerekamps A, van den Berk GE, Lauw FN, et al. Declining Hepatitis C Virus (HCV) incidence in Dutch human immunodeficiency virus-positive men who have sex with men after unrestricted access to HCV therapy. *Clin Infect Dis* 2018;66(9):1360–5.
- [22] Iversen J, Dore GJ, Catlett B, Cunningham P, Grebely J, Maher L. Association between rapid utilisation of direct hepatitis C antivirals and decline in the prevalence of viremia among people who inject drugs in Australia. *J Hepatol* 2019;70(1):33–9.
- [23] Lee SS, Crofts N, Hung CC. Macro-efforts for the micro-elimination of hepatitis C targeting people who inject drugs. *Int J Infect Dis* 2019;85:141–2.
- [24] Rockstroh J, Boesecke C. Treatment of acute hepatitis C in HIV coinfection: is this a chance for achieving microelimination? *United Eur Gastroenterol J* 2019;7(4):465–6.
- [25] Breskin A, Drobnik A, Pathela P, et al. Factors associated with hepatitis C infection among HIV-infected men who have sex with men with no reported injection drug use in New York City, 2000–2010. *Sex Transm Dis* 2015;42(7):382–6.
- [26] Foster AL, Gaisa MM, Hijdra RM, et al. Shedding of hepatitis C virus into the rectum of HIV-infected men who have sex with men. *Clin Infect Dis* 2017;64(3):284–8.
- [27] Huang MH, Chang SY, Liu CH, et al. HCV reinfections after viral clearance among HIV-positive patients with recent HCV infection in Taiwan. *Liver Int* 2019.
- [28] Bhagani S. HCV infection in HIV-negative MSM using HIV pre-exposure prophylaxis (PrEP)—Another piece of the jigsaw. *Liver Int* 2018;38(10):1733–5.
- [29] Matthews GV, Pham ST, Hellard M, et al. Patterns and characteristics of hepatitis C transmission clusters among HIV-positive and HIV-negative individuals in the Australian trial in acute hepatitis C. *Clin Infect Dis* 2011;52(6):803–11.
- [30] Liu JY, Lin HH, Liu YC, et al. Extremely high prevalence and genetic diversity of hepatitis C virus infection among HIV-infected injection drug users in Taiwan. *Clin Infect Dis* 2008;46(11):1761–8.
- [31] Yuan G, Liu J, Hu C, et al. Genotype distribution and molecular epidemiology of hepatitis C virus in Guangzhou, China: predominance of genotype 1b and increasing incidence of genotype 6a. *Cell Physiol Biochem* 2017;43(2):775–87.
- [32] Martin M, Vanichseni S, Leelawiwat W, et al. Hepatitis C virus infection among people who inject drugs in Bangkok, Thailand, 2005–2010. *WHO South East Asia J Public Health* 2019;8(1):50–5.
- [33] Ishida Y, Hayashida T, Sugiyama M, et al. Full-genome analysis of hepatitis C virus in Japanese and non-Japanese patients coinfecting with HIV-1 in Tokyo. *J Acquir Immune Defic Syndr* 2019;80(3):350–7.