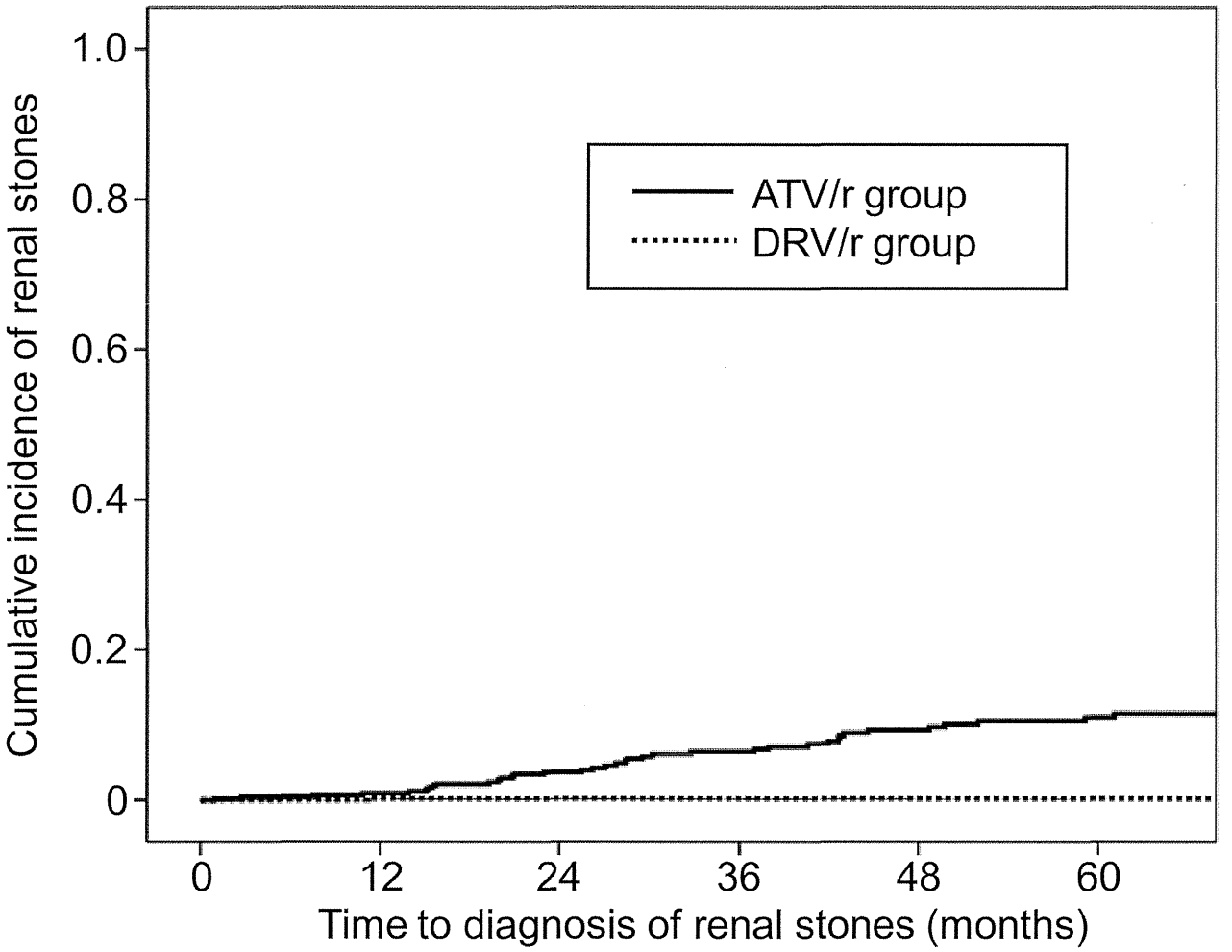

Figure 2. Kaplan-Meier curve showing time to the diagnosis of nephrolithiasis. ATV/r, ritonavir-boosted atazanavir; DRV/r, ritonavir-boosted darunavir.

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Table 2. Univariate analysis to estimate the risk of various factors for nephrolithiasis.

	Hazard ratio	95%CI	P value
ATV/r use over DRV/r	26.01	3.541-191.0	0.001
Age per 1 year	1.002	0.973-1.031	0.907
Male sex	1.665	0.401-6.919	0.483
Race (East Asian origin)	2.287	0.314-16.68	0.414
Weight per 1 kg increment	0.998	0.970-1.027	0.872
body mass index per 1 kg/m^2 increment	0.996	0.905-1.095	0.927
CD4 count per 10 /μl increment	0.999	0.983-1.016	0.901
HIV viral load per log10/ml	1.063	0.859-1.316	0.575
Treatment naïve	1.020	0.538-1.936	0.950
Tenofovir use	0.435	0.210-0.899	0.025
Baseline eGFR per 10 ml/min/1.73 m^2 decrement	1.103	0.980-1.242	0.105
Baseline serum uric acid per 1 mg/dl increment	1.415	1.173-1.705	<0.001
Hepatitis B or Hepatitis C	1.418	0.592-3.393	0.433
Past history of renal stone	2.658	1.111-6.359	0.028
Previous exposure to IDV	1.192	0.366-3.879	0.771

ATV/r, ritonavir-boosted atazanavir; DRV/r, ritonavir-boosted darunavir; HIV, human immunodeficiency virus; eGFR, estimated glomerular filtration rate; IDV, indinavir.

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Table 3. Multivariate analysis to estimate the risk of ATV/r- over DRV/r-containing antiretroviral therapies for nephrolithiasis.

	Model 1 crude (n=1,057)		Model 2 adjusted (n=1,056)		Model 3 adjusted (n=1,021)	
	HR	95%CI	HR	95%CI	HR	95%CI
ATV/r use over DRV/r	27.05	3.687-198.5	27.08	3.680-199.3	21.47	2.879-160.2
Age per 1 year			1.009	0.980-1.039	1.006	0.976-1.037
Male sex			1.939	0.441-8.528	1.202	0.262-5.512
Weight per 1 kg increment			0.988	0.956-1.021	0.979	0.947-1.012
Tenofovir use					0.678	0.313-1.470
Baseline serum uric acid per 1mg/dl increment					1.418	1.150-1.750
Past history of renal stone					1.661	0.520-5.307
Past exposure to IDV					0.491	0.100-2.403

HR, Hazard ratio; CI, confidence interval; ATV/r, ritonavir-boosted atazanavir; DRV/r, ritonavir-boosted darunavir; HIV, IDV, indinavir.

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Author Contributions

Conceived and designed the experiments: TN YH HG SO. Performed the experiments: TN YH KW K. Teruya. Analyzed the data: TN YH HK EK. Contributed reagents/materials/analysis tools: K. Tsukada YK. Wrote the manuscript: TN YH HG SO.

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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

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Background: The epidemiology of hepatitis C virus (HCV) infection among HIV-infected men who have sex with men (MSM) who do not inject drugs in Asia remains unknown.

Method: The incidence and risk factors for incident HCV infection among HIV-infected MSM at a large HIV clinic in Tokyo were elucidated. Poisson regression compared the incidence of HCV seroconversion at different observation periods.

Results: Of 753 HIV-1 infected MSM patients negative for HCV antibody (HCVAb) at baseline and available follow-up HCVAb test, 21 patients (2.8%) seroconverted to HCVAb positive over 2246 person-years (PY), for an incidence of 9.35 per 1000 PY. The incidence increased over time from 0 per 1000 PY in 2005–2006, 3.0 per 1000 PY in 2007–2008, 7.7 per 1000 PY in 2009–2010, to 24.9 per 1000 PY in 2011–2012 ($P = 0.012$). Of 21 incident cases, only 4 (19%) were injection drug users, and sensitivity analysis that excluded injection drug users yielded similar findings. Multivariate analysis identified illicit drug use to be an independent risk for HCV infection (hazard ratio = 3.006; 95% confidence interval: 1.092 to 8.275; $P = 0.033$).

Conclusions: Incident HCV infection is increasing among HIV-1-infected MSM noninjection drug users at resource-rich setting in Asia. Illicit drug use is an independent risk factor for incident HCV infection in this population.

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Key Words: incident HCV infection, illicit drug use, injection drug use, men who have sex with men, Tokyo, Asia

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INTRODUCTION

Coinfection with hepatitis C virus (HCV) in patients with HIV-1 infection is one of the major comorbidities and associates with worsened mortality.[1–6] Although HCV infection is especially common among injection drug users because of transparental infection,[7] noninjection illicit drug use and traumatic sexual behavior in the absence of injection drug use are reported in the United States and Europe to enhance noninjection drug-related transmission of HCV.[8–14]

In Japan, because of the low prevalence of injection drug use in the general population (lifetime prevalence of illicit drug use as a whole in 2009 was only 2.9%), coinfection with HCV in patients with HIV-1 infection is rare.[15–18] Recently, however, at our clinic, the AIDS Clinical Center, Tokyo, there has been a surge in the rate of acute HCV infection among men who have sex with men (MSM) with HIV-1 infection who deny using injection drugs. The AIDS Clinical Center is one of the largest referral centers for HIV care in Japan with patients mostly comprising MSM.[19]

This study was designed to determine the incidence and risk factors of incident HCV infection in MSM with HIV-1 infection at a resource-rich setting in Asia, with a special focus on noninjection illicit drug use.

METHODS

Study Subjects

The study population was HIV-1-infected MSM, aged older than 17 years, who visited our clinic for the first time from January 2005 to December 2010, and with negative HCV antibody (HCVAb) at baseline and at least 1 subsequent HCVAb test result. The following exclusion criteria were applied: (1) patients who visited the clinic for a second opinion, (2) patients referred to other facilities on their first or second visit. The study patients were followed up until December 31, 2012.

Measurements

At our clinic, HCVAb, CD4 cell count, HIV-1 viral load, hepatitis B surface antigen (HBsAg), and *Treponema pallidum* latex agglutination (TPHA) test are routinely conducted on the first visit. Patients positive for HCVAb subsequently undergo another HCV-RNA measurement. Patients visit our clinic at least every 3 months for monitoring CD4 cell count, HIV-1 viral load, and liver function tests, because the prescription period under the Japanese health care system is limited to 3 months. Repeat HCVAb tests were performed at the discretion of the treating physician. Laboratory data and baseline demographics on the first visit were collected from the medical records.

Social demographics including sexuality and history and type of illicit drug used (injection or noninjection) were collected through a structured interview conducted on the first visit.[20] Because the interview could underestimate the prevalence of illicit drug use, we also searched the medical records for information on illicit drug use and related variables covering the period from the first visit to December 2012.

At our clinic, written informed consent is obtained from each patient to store serum samples drawn at the first visit and subsequent visits at the discretion of the treating physician.[21] Either latest HCVAb test result conducted in clinical practice or from the latest stored serum sample was used to determine the status of HCV infection, whichever the follow-up time was longer. This study was approved by the Human Research Ethics Committee of the hospital.

Statistical Analysis

The incidence of HCV was calculated by dividing the number of HCVAb seroconversion by person-time at risk. Person-time represented the time from the first visit to the first positive HCVAb in patients diagnosed with incident HCV infection and the time from the first visit to the last negative HCVAb result in patients without incident infection. Poisson regression was used to compare the incidence of HCV seroconversion among 4 observation periods.

The uni- and multivariate Cox proportional hazards model was used to estimate the impact of illicit drug use as a whole over nonusers on the incidence of HCV infection as a primary exposure.

In multivariate analysis, age was added to the univariate analysis for illicit drug use over nonuse to form model 2. In model 3, injection drug use and insurance status were added because of their significant association with incident HCV infection in univariate analysis. Methamphetamine use and incarceration because of drugs were not added because of their multicollinearity with injection drug use (most injection drug users in Japan use methamphetamine injection).[18] Baseline CD4 count, HBsAg, and TPHA were added to the model because previous studies showed that low CD4 count and these infections are risk factors.[6,22]

Statistical significance was defined at 2-sided P values of <0.05. We used the hazard ratios (HRs) and 95% confidence intervals (95% CIs) to estimate the impact of each variable on the incidence of HCV infection. All statistical analyses were performed with the Statistical Package for Social Sciences version 20.0 (SPSS, Chicago, IL).

RESULTS

A total of 1182 MSM patients with HIV-1 infection visited the AIDS Clinical Center for the first time during the study inclusion period. Of these, 114 patients were excluded from the analysis (Fig. 1). The remaining 1068 patients had baseline HCVAb results, of whom 22 were positive for

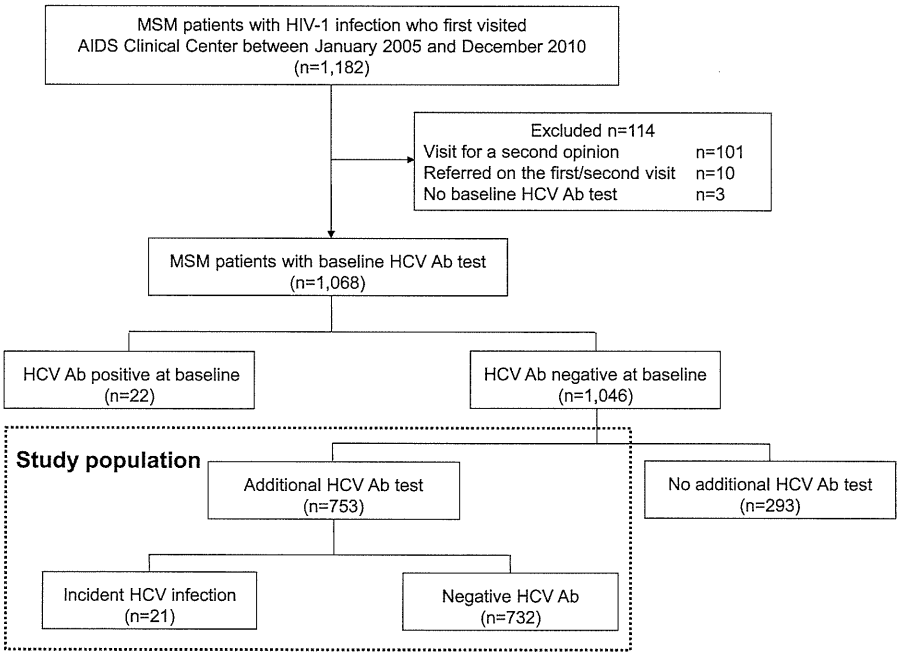

FIGURE 1. Patient enrollment process.

TABLE 1. Baseline Characteristics of Patients With and Without Incident HCV Infection

	All (n = 753)	Incident HCV (n = 21)	HCV-free (n = 732)	*P*
Median (IQR) age (yrs)	35 (29–41)	35 (32–42)	35 (29–41)	0.951
Illicit drug use, n (%)	298 (40)	15 (71)	283 (39)	0.003
Injection drug use, n (%)	37 (5)	4 (19)	33 (5)	0.016
Methamphetamine use, n (%)	48 (6)	4 (19)	44 (6)	0.039
Arrest due to illicit drug use, n (%)	24 (3)	3 (14)	21 (3)	0.026
Median (IQR) CD4 count (/μL)	244 (100–365)	311 (159–391)	244 (98–364)	0.539
Median (IQR) HIV-1 viral load (log_{10}/mL)	4.69 (4.00–5.20)	4.54 (3.51–4.97)	4.69 (4.00–5.22)	0.110
AIDS, n (%)	223 (30)	7 (33)	216 (30)	0.809
Positivity for hepatitis B surface antigen	48 (6)	2 (10)	46 (6)	0.638
Positivity for TPHA	236 (31)	8 (38)	228 (31)	0.483
On antiretroviral therapy, n (%)	66 (9)	3 (14)	63 (9)	0.419
Ethnicity, n (%)				0.537
Japanese	712 (95)	21 (100)	691 (94)	
Other Asian	20 (3)	0	20 (3)	
White	21 (3)	0	21 (3)	
Health insurance status, n (%)				0.003
With insurance	704 (94)	16 (76)	688 (94)	
No insurance	15 (2)	1 (5)	14 (2)	
On social benefits	34 (5)	4 (19)	30 (4)	
Median (IQR) follow-up days	984 (539–1557)	852 (324–1491)	997 (543–1562)	0.334

Patients' characteristics and social demographics were compared between those with incident HCV infection and those without such infection by the Student *t* test for continuous variables and by either the χ^2 test or Fisher exact test for categorical variables.

HCVAb. Of 1046 HIV-1 infected MSM patients with negative baseline HCVAb, the follow-up HCVAb test was available for 753 (72%) patients, either with clinical data (n = 295) or with stored blood samples (n = 458). The study population comprised the latter group of 753 patients.

Twenty-one patients (2.8%) incidentally seroconverted to HCVAb positive over 2246 person-years (PY) of total observation period, with an incidence of 9.35 per 1000 PY (95% CI: 6.12 to 14.2). The median time from the first visit to HCV seroconversion was 852 days [interquartile range (IQR), 324–1491 days]. The incidence increased over time from 0 per 1000 PY (0 case/270 PY) in 2005–2006, 3.0 per 1000 PY (2/672 PY) in 2007–2008, 7.7 per 1000 PY (6/779 PY) in 2009–2010, to 24.9 per 1000 PY (13/522 PY) in 2011–2012 ($P = 0.012$). As sensitivity analysis, the same calculations were conducted for the study population excluding injection drug users, and the results were similar; 17 patients became HCVAb positive over 2146 PY for an incidence of 7.92 per 1000 PY, and the incidence increased over time [0/1000 PY (0/258 PY) in 2005–2006, 1.5/1000 PY (1/650 PY) in 2007–2008, 8.0/1000 PY (6/747 PY) in 2009–2010, and 20.2/1000 PY (10/495 PY) in 2011–2012 ($P = 0.045$)].

The study patients were mostly Japanese men of relatively young age, covered with health insurance (Table 1); 40% of the study patients were illicit drug users based on the interview and medical records. Amyl nitrite, 5-methoxy-diisopropyltryptamine, 3,4-methylenedioxymethamphetamine, cannabis, heroin, cocaine, and opium were named by the patients. Patients with incident HCV infection were significantly more likely to be illicit drug users ($P = 0.003$). In addition, incident HCV cases were more likely to be injection drug users ($P = 0.016$), methamphetamine users ($P = 0.039$), and incarcerated owing to illicit drug use ($P = 0.026$) (Table 1). In contrast to the high prevalence of illicit drug use (40%), the prevalence of injection drug use was low (5%).

Among the 21 HCV incident cases, only 4 (19%) patients were injection drug users. Of the 21, 18 patients were screened for HCVAb because of high serum alanine aminotransferase levels, including all 17 positive for HCV RNA.

Among illicit drug users (n = 298) and nonusers (n = 455), 15 (5.0%) and 6 (1.3%) patients, respectively, were infected with HCV, with incidence of 16.6 and 4.48 per 1000 PY, respectively. Illicit drug users were significantly more likely to be infected with HCV, compared with nonusers ($P = 0.004$, Log-rank test). The total observation period was 906 PY (median, 1012 days; IQR, 543–1607 days) for illicit drug users and 1340 PY (median, 963 days; IQR, 538–1542 days) for nonusers.

Univariate analysis showed a significant relationship between illicit drug use and incident HCV infection (HR = 3.662; 95% CI: 1.420 to 9.439; $P = 0.007$) (see **Table S1, Supplemental Digital Content**, http://links.lww.com/QAI/A479). Furthermore, injection drug use (HR = 5.387; 95% CI: 1.804 to 16.09; $P = 0.003$), methamphetamine use (HR = 3.220; 95% CI: 1.083 to 9.573; $P = 0.035$), incarceration owing to illicit drugs (HR = 4.857; 95% CI: 1.429 to 16.51; $P = 0.011$), and on social benefits (government financial assistance) (HR = 6.982; 95% CI: 2.308 to 21.12; $P = 0.001$) were significantly associated with incident HCV infection. Conversely, age, low baseline CD4 count, positive HBsAg, and positive TPHA were not associated with incident HCV infection.

Multivariate analysis identified illicit drug use as a significant risk for incident HCV infection after adjustment for age (adjusted HR = 3.814; 95% CI: 1.447 to 10.05; P = 0.007) and for other variables (adjusted HR = 3.006; 95% CI: 1.092 to 8.275; P = 0.033) (see **Table S2, Supplemental Digital Content**, http://links.lww.com/QAI/A479). Injection drug users (HR = 4.672; 95% CI: 1.425 to 15.31; P = 0.011) and on social benefits (HR = 7.942; 95% CI: 2.370 to 26.62; P = 0.001) were also risk factors for incident HCV infection in multivariate analysis.

DISCUSSION

At our large urban HIV clinic in Tokyo, 21 (2.8%) HIV-infected MSM patients were newly infected with HCV during the study period, with an incidence of 9.35 per 1000 PY. The incidence of HCV infection showed statistically significant increase over the observation period, from 0 per 1000 PY in 2005–2006 to 24.9 per 1000 PY in 2011–2012. Most incident HCV cases were noninjection drug users, as 17 (81%) patients were not identified as using injection drugs. The incidence of new HCV infection was >3-folds higher in any illicit drug users than nonusers, with estimated incidence of 16.6 and 4.48 per 1000 PY, respectively, and illicit drug use was identified as a significant risk factor for incident HCV infection in multivariate analysis.

To our knowledge, this is the first study in Japan and is the second in Asia to report the incidence and risk factors for incident HCV infection among HIV-infected MSM.[23] The incidence of 9.35 per 1000 PY was similar to that reported in North America and Western Europe (8.7–16.3/1000 PY) and was also very close to that of Taiwan (9.25/1000 PY).[23–26] The results of this study demonstrated the emergence of noninjection drug-related HCV infection in HIV-1-infected MSM in resource-rich settings in Asia and support routine rescreening for HCVAb among HIV-infected MSM, in particular among drugs users, both injectors and noninjectors.

The reason for the increasing trend of incident HCV infection in HIV-infected MSM noninjection drug users remains to be elucidated. Both baseline positive HBsAg and TPHA, markers for sexual activity, were not associated with incident HCV in this study. This is probably because these variables were collected at the baseline and do not necessarily reflect the ongoing sexual activity during observation period. However, based on reports from Western Europe and North America, it is reasonable to assume that such change in the incidence is mainly because of increased practice of high-risk sexual behaviors, such as unprotected anal intercourse, group sex, and fisting (often practiced in the context of illicit drug use that can lower the pain threshold, leading to increased bleeding) among MSM with HIV-1 infection in Japan.[10] Sharing of drug paraphernalia and possible intranasal/intrarectal transmission can also contribute.[27,28] Because of the introduction of effective and tolerable antiretroviral therapy, the life expectancy and quality of life of people with HIV infection has improved substantially, which could associate with increased high-risk behavior in MSM with HIV-1 infection.[29,30]

Several limitations need to be acknowledged. First, because this study did not count the number of HCVAb tests for each patient during each observation period, it is possible that the increasing trend of incident HCV infection is because of increasing number of HCV testing in recent years. However, most patients with incident HCV infection were tested for HCVAb in clinical practice, because they were suspected to have acute HCV infection based on elevated liver enzymes. Thus, it is highly likely that the increasing trend of HCV infection shown in this study is true for this cohort. Second, we were not able to rescreen for HCVAb for 28% of the patients with negative baseline HCVAb result. Third, the structured interview designed for data collection and chart review do not necessarily prevent underreporting of illicit drug use and injection drug use. However, underreporting to a certain degree is unavoidable with regard to issues such as illicit drugs.[31]

In conclusion, this study showed that incident HCV is increasing among mostly noninjection drug use MSM with HIV-1 infection at resource-rich setting in Asia. Illicit drug use was identified as an independent risk factor for incident HCV infection. It is important to raise awareness of noninjection drug-related HCV infection and to take appropriate measures to prevent HCV infection in this population.

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PLOS ONE

Traditional but Not HIV-Related Factors Are Associated with Nonalcoholic Fatty Liver Disease in Asian Patients with HIV-1 Infection

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Abstract

Background: The prevalence and factors associated with nonalcoholic fatty liver disease (NAFLD) are largely unknown in HIV-1 monoinfected patients.

Methods: The present study elucidated the prevalence and factors associated with NAFLD among Asian patients with HIV-1 infection who underwent abdominal ultrasonography between January 2004 and March 2013. Diagnosis of NAFLD was based on the liver to kidney contrast and diffusion in hepatic echogenicity. Uni- and multi-variate logistic regression analyses were applied to estimate factors associated with NAFLD.

Results: 435 Asian patients free of chronic hepatitis B or C virus infection and without excessive alcohol intake were analyzed. NAFLD was diagnosed in 135 (31%) patients. Obesity (BMI >30 kg/m^2) was evident in 18 (4.1%) patients, and BMI was >25 kg/m^2 in 103 (24%). Multivariate analysis identified higher BMI (per 1 kg/m^2 increment, adjusted OR = 1.198; 95% CI, 1.112–1.290; $p<0.001$), dyslipidemia (adjusted OR = 2.045; 95% CI, 1.183–3.538; $p=0.010$), and higher ALT to AST ratio (per 1 increment, adjusted OR = 3.557; 95% CI, 2.129–5.941; $p<0.001$) as significant factors associated with NAFLD. No HIV-specific variables, including treatment with dideoxynucleoside analogues (didanosine, stavudine, and zalcitabine) and cumulative duration of antiretroviral therapy (ART), were associated with NAFLD.

Conclusions: The incidence of NALFD among Asian patients with HIV-1 infection is similar to that in Western countries. NAFLD was associated with high BMI, dyslipidemia, and high ALT/AST ratio, but not with HIV-related factors. The results highlight the importance of early recognition and management of NAFLD and traditional factors associated with NAFLD for Asian patients with HIV-1 infection.

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Introduction

Nonalcoholic fatty liver disease (NAFLD) is characterized by the presence of fat infiltration in the liver in the absence of excessive alcohol consumption or other causes of liver disease, such as viral hepatitis, and is considered the most common cause of fatty liver [1]. NAFLD is a major health issue since it can lead to fibrosis, cirrhosis, liver cancer, and mortality [2]. Although the prevalence of NAFLD seems increasing in parallel with the current epidemic of obesity, it varies among the general population according to the geographical area; for example, the prevalence of NAFLD in the US ranges from 10 to 46% [3,4], whereas in Asia it is 5–30% [5].

In the general population, obesity, type 2 diabetes mellitus, dyslipidemia, and metabolic syndrome are established conditions associated with NAFLD [6]. In addition to abovementioned environmental risk factors, genetic factors are also associated with the incidence of NAFLD [7]. However, only two studies (one from Italian metabolic clinic and the other from American naval hospital) have previously examined the prevalence and associated factors with NAFLD in patients infected with HIV-1 only (those without chronic hepatitis C virus (HCV) infection) [8,9]. At this stage, it is unknown whether variables specific to HIV-1 infection, such as HIV-1 viral load and cumulative years of antiretroviral therapy (ART) are associated with NAFLD. Although the use of so called "D drugs": dideoxynucleoside analogues [didanosine (ddI), stavudine (d4T), and zalcitabine (ddC)], a subgroup of antiretroviral agents nucleoside reverse transcriptase inhibitors (NRTI), is

reported to be associated with NAFLD, others have argued against such relation [8,9].

Liver diseases are important causes of morbidity and mortality among patients with HIV-1 infection [10–12], especially following the wide availability of ART and substantial improvement in prognosis of such patients [13]. Currently, there is no information on the prevalence and associated factors related to NAFLD among patients with HIV-1 infection in Asia, the region with the second largest number of patients with HIV-1 infection. The present study was designed to elucidate the prevalence and associated factors, including D drug use, with NAFLD in Asian patients with HIV-1 infection.

Methods

Ethics statement

This study was approved by the Human Research Ethics Committee of the National Center for Global Health and Medicine, Tokyo. Each participant provided a written informed consent for the clinical and laboratory data to be used and published for research purposes. The study was conducted according to the principles expressed in the Declaration of Helsinki.

Study design

We performed a single-center cross-sectional study of HIV-1-infected patients using the abdominal ultrasonography data and medical records at the National Center for Global Health and Medicine, Tokyo, Japan. Our facility is one of the largest clinics for patients with HIV infection in Japan with approximately 3,500 registered patients [14]. The study population was HIV-infected patients, aged >17 years, who underwent routine abdominal ultrasonography conducted by certified medical technologists at the Physiological Examination Unit of the hospital, between January 1, 2004 and March 31, 2013. The following exclusion criteria were employed in this study; 1) HCV or hepatitis B virus (HBV) infection defined by positive hepatitis C antibody or positive hepatitis B surface antigen, respectively, 2) use of injection drugs, 3) hemophilia, because all HIV-infected hemophiliacs in Japan were exposed to HCV through contaminated blood products [15], and 4) alcohol consumption >20 g of ethanol per day for males and >10 g/day for females. Fatty liver was diagnosed based on hyperechogenicity of the liver compared to renal cortex and diffusion in hepatic echogenicity [8,16,17]. The ultrasonographic images and diagnosis were double-checked and confirmed by radiologists, hepatologists, or gastroenterologists. If abdominal ultrasonography was conducted more than once during the study period, the latest data were used for the study.

Measurements

The potential risk factors for NAFLD were selected according to previous studies and collected from the medical records [18,19], together with the basic demographic data. These factors included age, sex, race, body weight, body mass index (BMI) = {bodyweight (kg)/[(height (m)]^2}, and presence or absence of other medical conditions [diabetes mellitus, defined by use of glucose-lowering agents or fasting plasma glucose ≥126 mg/dl or plasma glucose ≥ 200 mg/dl on two different days, dyslipidemia, defined by current treatment with lipid-lowering agents or two successive measurements of either low-density lipoprotein cholesterol (LDL-C) > 140 mg/dl, high-density lipoprotein cholesterol (HDL-C) < 40 mg/dl, total cholesterol (TC) >240 mg/dl, triglyceride (TG) >500 mg/dl, and hypertension defined by current treatment with antihypertensive agents or two successive measurements of systolic blood pressure ≥140 mmHg or diastolic blood pressure ≥ 90 mmHg at the clinic]. Data on smoking status and alcohol consumption were collected through a structured interview conducted at the first visit as part of routine clinical practice by the nurses specializing at the HIV outpatient care. Patients were divided into three groups according to the smoking status: non-smokers, low (<20 cigarettes/day) and heavy smokers (≥20 cigarettes/day). They were also divided according to alcohol consumption into two groups: non-drinkers and light drinkers (< 20 g ethanol/day for men and <10 g ethanol/day for women). The values of alanine aminotransferase (ALT), aspartate aminotransferase (AST), TC, LDL-C, HDL-C, and TG within three months and closest to the day ultrasonography was conducted were collected. HIV-specific variables, such as CD4 cell count, HIV viral load, ART-experienced or ART-naïve, ART regimen at ultrasonography, history of AIDS, and duration of ART were also collected. The duration of D drugs use, as a possible risk factor for NAFLD, was collected, regardless of continuation of these drugs at the time of abdominal ultrasonography [9,20]. Patients were divided into four groups according to duration of treatment with D drugs; no D drugs use, <1 year exposure, 1–3 years of use, and >3 years of use. In our clinic, it is customary for the patient to visit the clinic once a month before the initiation of ART and until the suppression of HIV-1 viral load, but the visit interval is extended up to every three months after viral load suppression.

Statistical analysis

Baseline characteristics were compared between patients with and without NAFLD, using the Student's *t*-test or χ^2 test (Fisher's exact test) for continuous or categorical variables, respectively. Univariate logistic regression analysis was used to identify factors associated with NAFLD. Basic demographics, such as age and sex, and variables with p values <0.05 in univariate analysis were entered into multivariate logistic regression models. ALT, and TG and LDL-C were not added to the model, based on their multicollinearity with ALT to AST ratio and dyslipidemia, respectively. Statistical significance was defined as two-sided p value <0.05. We used the odds ratio (OR) and 95% confidence interval (95%CI) to estimate the association of each variable with NAFLD. All statistical analyses were performed with The Statistical Package for Social Sciences ver. 20.0 (SPSS, Chicago, IL).

Results

Of the total of 895 patients with HIV-1 infection who underwent abdominal ultrasonography during the study period, 435 were included in the analysis (Figure 1). NAFLD was diagnosed by abdominal ultrasonography in 135 cases, with a prevalence of 31%. None of these patients had any ultrasonographic finding compatible with cirrhosis. Table 1 shows the characteristics of the study population, patients with NAFLD, and those without NAFLD. The study patients were mostly East Asian males with maintained CD4 count [median 349/μl, interquartile range (IQR) 203–512], and approximately half of the patients had suppressed viral load. Obesity (BMI >30 kg/m^2) was noted in 18 (4.1%) patients, and BMI was >25 kg/m^2 in 103 (24%). Body weight was significantly heavier in patients with NAFLD (median 71 kg, IQR 61–78 kg), compared with non-NAFLD (median 61 kg, IQR 55–68 kg, p<0.001), as was BMI (median 25, IQR 21.7–27.5 versus median 21.5, IQR 20–23.3, p<0.001). Dyslipidemia (p<0.001), hypertension (p = 0.019), high ALT (p = 0.017), high LDL-C (p = 0.041), hypertriglyceridemia (p = 0.008), and high CD4 count (p = 0.001) were significantly more common in

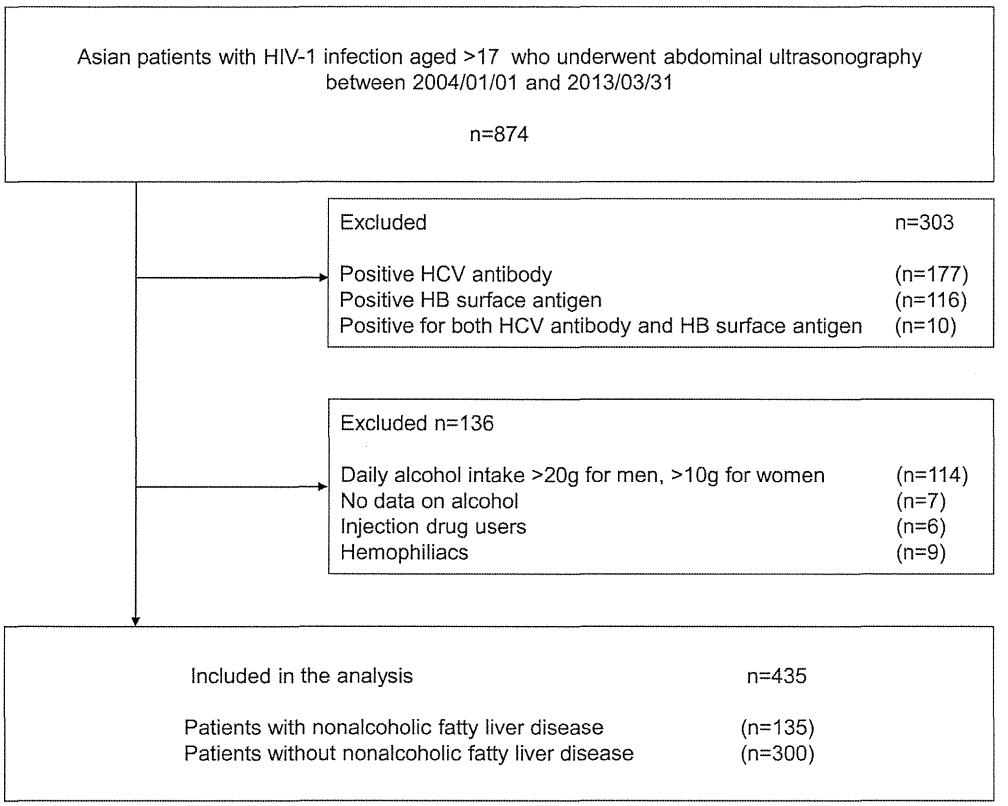

Figure 1. Patient enrollment process.
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patients with NAFLD than those without (Table 1). On the other hand, history of D drug use and cumulative years of ART were not significantly different between the two groups.

Univariate analysis showed a significant association between NAFLD and the following non-HIV specific variables (Table 2): higher BMI (per 1 kg/m^2 increment, OR = 1.282; 95% CI, 1.197–1.373; p<0.001), dyslipidemia (OR = 2.475; 95% CI, 1.594–3.842; p<0.001), hypertension (OR = 1.818; 95% CI, 1.117–2.961; p = 0.016), ALT to AST ratio (per 1 increment, OR = 4.831; 95% CI, 3.073–7.594; p<0.001), higher ALT (per 10 IU/l increment, OR = 1.027; 95% CI, 1.002–1.053; p = 0.034), higher triglyceride (per 10 mg/dl increment, OR = 1.021; 95% CI, 1.005–1.038; p = 0.010), and higher LDL-C (per 10 mg/dl increment, OR = 1.096; 95% CI, 1.003–1.196; p = 0.042). Among HIV-specific variables, only higher CD4 count was associated with NAFLD (per 1/μl increment, OR = 1.001; 95% CI, 1.001–1.002; p = 0.002)(Table 3). On the other hand, older age (per 1 year increment, OR = 0.996; 95% CI, 0.980–1.013; p = 0.668) and diabetes mellitus (OR = 1.577; 95% CI, 0.657–3.784; p = 0.308) were not associated with NAFLD. Compared to no D drug use, history of D drug use was not associated with NAFLD (Any to <1 year of D drug use, n = 42, OR = 0.956; 95% CI, 0.476–1.919; p = 0.899)(1 to 3 years of D drug use, n = 46, OR = 1.137; 95% CI, 0.592–2.184; p = 0.699)(> 3 years of D drug use, n = 40, OR = 0.533; 95% CI, 0.237–1.200; p = 0.129)(Table 3).

Among patients treated with D drugs (n = 128), the median time period since withdrawal was 3.46 years (IQR 1.03–6.29). Compared to treatment-naïve patients, ART use was not associated with NAFLD as well (<2 year of ART exposure, n = 80, OR = 1.110; 95% CI, 0.620–1.985; p = 0.726) (2 to 6 years of ART exposure, n = 100, OR = 0.941; 95% CI, 0.541–1.637; p = 0.830) (>6 year of ART exposure, n = 103, OR = 1.135; 95% CI, 0.664–1.943; p = 0.643)(Table 3).

Multivariate analyses identified the following variables as independently associated with NAFLD: BMI (per 1 kg/m^2 increment, adjusted OR = 1.198; 95% CI, 1.112–1.290; p< 0.001), dyslipidemia (adjusted OR = 2.045; 95% CI, 1.183–3.538; p = 0.010), ALT to AST ratio (per 1 increment, adjusted OR = 3.557; 95% CI, 2.129–5.941; p<0.001)(Table 4).

Discussion

To our knowledge, this is the first study that investigated the prevalence and associated factors of NAFLD in Asian patients with HIV-1 infection, and is the largest study that focused on NAFLD in patients with HIV-1 monoinfection (without chronic hepatitis C infection). The prevalence of NAFLD in this study was 31%, which is comparable to 31% at the Naval hospital in San Diego, US, and 36.9% at the metabolic clinic in Modena, Italy [8,9]. Multivariate analysis indicated that traditional predictors for NAFLD in the general population, such as higher BMI, dyslipidemia, and ALT to AST ratio [6], were significantly associated with NAFLD, whereas HIV-specific variables, including history of D drug use and cumulative years of ART, were not associated with NAFLD.

Our result of nearly one third of Asian patients with HIV-1 monoinfection have NAFLD highlights the importance of screening for NAFLD among this patient population, due to the potential progression of NAFLD to liver fibrosis, cirrhosis, and liver cancer [2,21]. In addition, the finding that higher BMI,

Table 1. Basic demographics of the entire study population, patients with NAFLD and without NAFLD.

	Total (n = 435)	NAFLD (n = 135)	No NAFLD (n = 300)	P[a]
Age (years)[¶]	40 (35–50)	41 (36–48)	40 (34–55)	0.669
Male sex, n (%)	406 (93)	129 (96)	277 (92)	0.299
Body weight (kg)[¶]	63 (57–73)	71 (61–78)	61 (55–68)	<0.001
Body mass index, (kg/m^2)[¶]	22.1 (20.2–24.9)	25 (21.7–27.5)	21.5 (20–23.3)	<0.001
Body mass index >25 kg/m^2, n (%)	103 (24)	64 (49)	39 (13)	<0.001
Body mass index >30 kg/m^2, n (%)	18 (4.1)	16 (12)	2 (1)	<0.001
East Asian origin, n (%)	424 (98)	133 (99)	291 (97)	0.515
Diabetes mellitus, n (%)	22 (5)	9 (7)	13 (4)	0.345
Dyslipidemia, n (%)	120 (28)	55 (41)	65 (22)	<0.001
Hypertension, n (%)	86 (20)	36 (27)	50 (17)	0.019
ALT (IU/l)[¶]	26 (17–47)	47 (25–80)	22 (16–33)	0.017
AST (IU/l)[¶]	25 (19–37)	31 (21–50)	23 (18–31)	0.152
ALT to AST ratio[¶]	1.05 (0.8–1.42)	1.42 (1.02–1.76)	1 (0.74–1.21)	<0.001
Low-density lipoprotein cholesterol (mg/dl)[¶]	102 (85–126)	111 (90–129)	101 (83–125)	0.041
High-density lipoprotein cholesterol (mg/dl)[¶]	44 (35–52)	43 (34–52)	44 (35–54)	0.701
Triglyceride (mg/dl)[¶]	162 (104–233)	189 (125–254)	149 (96–226)	0.008
Total cholesterol (mg/dl)[¶]	175 (150–205)	179 (151–208)	177 (149–226)	0.202
Smoking status, by no. of cigarettes per day				0.244
None, n (%)	247 (57)	84 (62)	163 (55)	
<20, n (%)	82 (19)	20 (15)	62 (21)	
≥20, n (%)	105 (24)	31 (23)	74 (25)	
Alcohol consumption				1.000
None, n (%)	209 (48)	65 (48)	144 (48)	
Moderate (<20 g/day for men, <10 g/day for women), n (%)	226 (52)	70 (52)	156 (52)	
HIV-specific variables				
CD4 cell count (cells/μL)[¶]	349 (203–512)	377 (230–591)	338 (172–480)	0.001
HIV load (log_{10} copies/mL)[¶]	1.70 (1.70–4.45)	1.70 (1.70–4.36)	1.70 (1.70–4.52)	0.508
HIV load <50 copies/mL, n (%)	227 (52)	73 (55)	154 (52)	0.602
Homosexual contact, n (%)	377 (87)	120 (89)	257 (86)	0.446
History of ddI/ddC/d4T exposure, n (%)	128 (29)	37 (27)	91 (30)	0.571
ART duration (years)[¶]	1.4 (0–5.6)	1.4 (0–6.1)	1.6 (0–5.4)	0.844
Current antiretroviral therapeutic regimen				
Ritonavir-boosted PI plus 2NRTIs, n (%)	186 (43)	58 (43)	128 (43)	1.000
NNRTI plus 2NRTIs, n (%)	44 (10)	14 (10)	30 (10)	1.000
Treatment naïve, n (%)	152 (35)	46 (34)	106 (35)	0.829
History of AIDS, n (%)	156 (36)	51 (38)	105 (35)	0.590

[¶]Data are median (interquartile range). Four missing values in variable HIV load<50 copies/mL.
[a]χ^2 test or Fisher's exact test was used for categorical data, and Student's *t* test for continuous variables.
NAFLD, nonalcoholic fatty liver disease; ALT, alanine aminotransferase; AST, aspartate aminotransferase; ddI, didanosine; ddC, zalcitabine; d4T, stavudine; ART, antiretroviral therapy; PI, protease inhibitor; NRTI, nucleoside reverse transcriptase inhibitor; NNRTI, non-nucleoside reverse transcriptase inhibitor; AIDS, acquired immunodeficiency syndrome.
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dyslipidemia, and ALT to AST ratio were associated with NAFLD warrants aggressive approach to life-style changes and keeping optimal body weight, as well as the management of dyslipidemia. This is particularly important because the metabolic syndrome, obesity, type 2 diabetes mellitus, and dyslipidemia are widely prevalent and are increasing among the general population in Asia [5]. Our study identified obesity in 4.1% of the study population (BMI >30 kg/m^2), the number that is similar to that reported from the Italian metabolic clinic (4.9%), although much lower than that reported in US (14.8%) [8,9]. Our results showed that the prevalence of NAFLD in Asian patients with HIV-1 infection is as high as that reported in the above two studies, and warrants the need for paying attention to this disease in Asian patients with HIV-1 infection.

Interestingly, the present study did not identify HIV-specific variables, especially treatment with D drugs, to be associated with NAFLD. D drugs (dideoxynucleoside analogues; ddI, d4T, and ddC), a subgroup of NRTIs, inhibit mitochondrial DNA (mDNA)

Table 2. Univariate analysis to estimate the associations of non HIV-specific variables with nonalcoholic fatty liver disease.

	Odds ratio	95%CI	P value
Male sex	1.785	0.710–4.491	0.218
Age per 1 year increment	0.996	0.980–1.013	0.668
Body mass index per 1 kg/m^2 increment	1.282	1.197–1.373	<0.001
Alcohol consumption			
No drinking	Reference	Reference	Reference
Ethanol <20 g/day for men, <10 g/day for women	0.994	0.662–1.493	0.977
Smoking status			
Non smoker	Reference	Reference	Reference
<20 cigarettes/day	0.626	0.354–1.105	0.106
≥20 cigarettes/day	0.813	0.495–1.334	0.412
Diabetes mellitus	1.577	0.657–3.784	0.308
Dyslipidemia	2.475	1.594–3.842	<0.001
Hypertension	1.818	1.117–2.961	0.016
ALT to AST ratio per 1 increment	4.831	3.073–7.594	<0.001
ALT per 10 IU/l increment	1.027	1.002–1.053	0.034
AST per 10 IU/l increment	1.034	0.986–1.084	0.169
Triglyceride per 10 mg/dl increment	1.021	1.005–1.038	0.010
Low-density lipoprotein cholesterol per 10 mg/dl increment	1.096	1.003–1.196	0.042
Total cholesterol per 10 mg/dl increment	1.037	0.981–1.096	0.202
High-density lipoprotein cholesterol per 10 mg/dl increment	1.032	0.878–1.215	0.700

CI, confidence interval; ALT, alanine aminotransferase; AST, aspartate aminotransferase.
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Table 3. Univariate analysis to estimate the association of HIV-specific variables with nonalcoholic fatty liver disease.

	Odds ratio	95%CI	P value
ddI/ddC/d4T use	0.867	0.552–1.362	0.536
No ddI/ddC/d4T use (n = 307)	Reference	Reference	
<1 year of ddI/ddC/d4T use (n = 42)	0.956	0.476–1.919	0.899
1–3 years of ddI/ddC/d4T use (n = 46)	1.137	0.592–2.184	0.699
>3 years of ddI/ddC/d4T use (n = 40)	0.533	0.237–1.200	0.129
ART exposure			
Treatment naïve (n = 152)	Reference	Reference	
<2 years of ART exposure (n = 80)	1.110	0.620–1.985	0.726
2–6 years of ART exposure n = 100)	0.941	0.541–1.637	0.830
>6 years of ART exposure (n = 103)	1.135	0.664–1.943	0.643
CD4 count per 1/μl increment	1.001	1.001–1.002	0.002
HIV viral load per $\log_{10}$/ml increment	0.955	0.833–1.094	0.507
HIV viral load <50 copies/ml	1.138	0.755–1.715	0.538
History of AIDS	1.128	0.740–1.718	0.576
Treatment naive	0.946	0.617–1.450	0.799

OR, odds ratio; CI, confidence interval; ddI, didanosine; ddC, zalcitabine; d4T, stavudine; ART, antiretroviral therapy; AIDS, acquired immunodeficiency syndrome.
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polymerase γ, resulting in depletion of mDNA in the liver [22], and causes mitochondria toxicity with potential fatal lactic acidosis and hepatic steatosis [23–25]. However, previous studies on patients with HIV monoinfection (without chronic hepatitis C infection) showed conflicting results with regard to the relation between NAFLD and D drug use [8,9]. The present study also did not find significant association between D drug use and NAFLD. Considering that D drugs are rarely used in resource-rich settings and their use is also rapidly decreasing in resource-limited settings, especially after 2010 revision of WHO guidelines, which eliminated d4T from the first line therapy (http://whqlibdoc.who.int/publications/2010/9789241599764_eng.pdf), it is probably plausible to say that more focus needs to be put on traditional

Table 4. Multivariate analysis of independent variables associated with nonalcoholic fatty liver disease (n = 408).

	Adjusted OR	95%CI	P value
Male sex	1.953	0.640–5.966	0.240
Age 1 year increment	1.005	0.983–1.027	0.672
Body mass index per 1 kg/m^2 increment	1.198	1.112–1.290	<0.001
Dyslipidemia	2.045	1.183–3.538	0.010
ALT to AST ratio per 1 increment	3.557	2.129–5.941	<0.001
Hypertension	0.959	0.510–1.805	0.897
CD4 count per 1/μl increment	1.001	0.999–1.002	0.336

OR, odds ratio; CI, confidence interval; ALT, alanine aminotransferase; AST, aspartate aminotransferase.
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predictors for NAFLD, such as obesity and dyslipidemia, rather than D drug use when screening and managing NAFLD in patients with HIV-1 infection.

There are several limitations to our study. First, the diagnosis of NAFLD was achieved by use of ultrasonography, although histological confirmation of NAFLD by liver biopsy is considered the gold standard [16]. Because it is also difficult to grade the severity of fat infiltration in the liver by ultrasonography, the present study could not distinguish nonalcoholic steatohepatitis (NASH), the more severe form of NAFLD [6,16,19]. However, liver biopsy is an invasive and costly procedure. Compared to histopathology and other imaging devices, such as computed tomography (CT) and magnetic resonance imaging (MRI), the reliability and accuracy of ultrasonography in the diagnosis of fatty liver has been well-established [16]. Other advantages of ultrasonography includes low cost, safety, and availability, compared with liver biopsy, CT, and MRI [16]. Second, because the study population comprised of mostly males, the results of the present study might not apply to female patients. Third, we cannot exclude possible overestimation of the prevalence of NAFLD in this study since the study population included patients who underwent abdominal ultrasonography in clinical practice. However, considering that the two previous reports on NAFLD in HIV-monoinfected patients included only patients with dyslipidemia and hyperglycemia at the metabolic clinic [9], and almost exclusively military personnel at the naval hospital [8], respectively, the present study confers clinically useful information derived from routine clinical practice with comparatively unrestricted patient population at a large urban HIV clinic.

In conclusion, the present study demonstrated that the prevalence of NAFLD in Asian patients with HIV-1 infection was 31%, which is comparable to the studies from Western Europe and US. NAFLD was significantly associated with traditional predictors for NAFLD, such as higher BMI, dyslipidemia, and ALT to AST ratio, but not with any HIV-specific variable, including history of D drug use and cumulative years of ART. The results highlight the importance of early recognition and management of NAFLD and its traditional predictors, in order to prevent further progression of NAFLD in Asian patients with HIV-1 infection.

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Author Contributions

Conceived and designed the experiments: TN TS HK. Performed the experiments: TN YN MY. Analyzed the data: TN HG TS HK SO. Contributed reagents/materials/analysis tools: YN NN YK MY SO. Wrote the paper: TN HG TS HK SO.

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Impact of HIV Infection on Colorectal Tumors: A Prospective Colonoscopic Study of Asian Patients

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Background: Non-AIDS defining cancer has recently become a major problem in HIV-infected patients. Little has been reported on whether HIV infection is a risk factor for colorectal adenoma, especially in Asians.

Methods: The study was conducted under a prospective cross-sectional design and included all adults who underwent colonoscopy. Subjects were matched by age and sex to compare the prevalence of colorectal adenoma, adenocarcinoma, polyps, and other tumors. Detailed risk factors were assessed, including lifestyle habits, medications, comorbidities, gastrointestinal symptom rating scale, HIV-associated factors, and human papillomavirus infection. To evaluate the effects of HIV infection on adenoma, the odds ratio (OR) was estimated by multivariate logistic regression.

Results: A total of 177 HIV-infected patients and 177 controls were selected for analysis. No significant difference was noted in the prevalence of adenoma (n = 29 vs. 40, $P = 0.14$). Multivariate analysis adjusted by baseline demographics and risk factors showed that HIV is not associated with increased risk of adenoma (adjusted OR = 0.66, $P = 0.16$). Kaposi's sarcoma was more common in HIV-infected patients (n = 6 vs. 0, $P = 0.03$). Among HIV-infected patients, advanced age was an independent and significant risk factor for adenoma (adjusted OR = 2.28, $P < 0.01$). CD4 count, HIV-RNA, history of antiretroviral treatment, and oncogenic human papillomavirus infection were not risk factors for adenoma.

Conclusions: HIV infection was not identified as risk for adenoma in Asian patients. However, advanced age was independently associated with increased risk of adenoma. HIV-infected patients should not miss screening opportunity for colorectal adenoma and other gastrointestinal malignancies.

Key Words: colorectal cancer, colorectal adenoma, oncogenic HPV infection, Japan, gastrointestinal malignancy

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INTRODUCTION

The introduction of highly active antiretroviral therapy (HAART) has significantly improved the morbidity and mortality of HIV-infected patients.[1,2] However, the incidence of non-AIDS defining cancer has increased with prolongation of life expectancy of HIV-infected patients.[3–6]

Colorectal cancer is the third most commonly diagnosed cancer in male patients and the second in female patients, and a major cause of death worldwide.[7] To prevent the development of colorectal cancer and death, removal of premalignant lesion, adenoma, is effective, and screening is recommended in patients aged 50 years and older.[8–11] Recent studies from western countries have suggested that higher incidence of colonic adenoma in patients with HIV infection compared with the general population.[12–14] Furthermore, HIV-infected patients are at high risk of oncogenic human papillomavirus (HPV) infection, and the potential role of HPV infection in the development of colorectal cancer has been suggested.[15–17] However, little is known about the risk of adenoma in HIV-infected patients compared with the general population.

In the past, the incidence of colorectal cancer was lower in Asia compared with Western countries.[18] However, the incidence has increased lately in Asian countries, including Japan, and is currently comparable with that in western countries.[18,19] Nevertheless, all previous studies on colorectal adenoma in HIV-infected patients were only from the United States, and there are no available data in Asia.[12–14] This study reports the findings of a prospective cross-sectional colonoscopic study that compared the prevalence of colorectal

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adenoma in HIV-infected patients with HIV-negative patients in Japan.

METHODS

Study Design, Setting, and Participants

We conducted a prospective cross-sectional single-center study in adults who underwent colonoscopy between September 2009 and July 2012 at the endoscopy unit of the National Center for Global Health and Medicine (NCGM). NCGM has one of the largest clinics for patients with HIV infection in Japan with more than 3500 registered patients as of May 2013. The institutional review board at NCGM approved this study. The study was conducted according to the principles expressed in the Declaration of Helsinki.

The following inclusion criteria were used in this study: (1) aged 18 years or older, (2) independent in activities of daily living, (3) able to understand written documents and to write, and (4) asymptomatic but desired screening for colorectal cancer, or presented with continuous or intermittent lower gastrointestinal (GI) symptoms. The following exclusion criteria were used in this study: (1) contraindication or patient refusal of total colonoscopy, (2) colonoscopy for follow-up evaluation during the study period, and (3) previous diagnosis of either adenoma or adenocarcinoma. All inclusion and exclusion criteria were fulfilled before patients were enrolled in the study. Each HIV-infected patient was matched with 1 HIV-negative patient based on age in 5-year age-bands and sex.

Clinical Factors

A detailed questionnaire was completed at the endoscopy unit on the same day of colonoscopy. Patients were asked about their (1) lifestyle habits (smoking history and alcohol consumption), (2) medications [nonsteroidal anti-inflammatory drugs (NSAIDs) and low-dose aspirin], and (3) comorbidities (hypertension, diabetes mellitus, and coronary heart disease) in a face-to-face interview with the medical staff. With regard to medication history, prescriptions and medical records were reviewed in addition to information provided by the patients to avoid omissions. The survey form included photographs of all these oral drugs, which are approved in Japan. Regular use of medication was defined as oral administration starting at least 1 year before the interview. The smoking index was evaluated among ever and daily smokers and was defined as the number of cigarettes per day multiplied by the number of smoking years. Then, smoking index was categorized into nil, <400, 400–799, and >800. Alcohol consumption was calculated and categorized into nondrinker, light (1–180 g/wk), moderate (181–360 g/wk), and heavy drinker (>360 g/wk). To evaluate lower GI symptoms, the GI symptom rating scale rating on a 7-graded Likert scale was used.[20,21] The GI symptom rating scale consists of 15 questions covering lower GI symptoms: increased flatus, decreased passage of stools, increased passage of stools, loose stools, hard stools, urgent need for defecation, and feeling of incomplete evacuation. Positive symptoms were defined as score ≥ 3.

For HIV-infected patients, CD4 cell count, HIV viral load, history of HAART, and sexual behavior were also obtained. CD4 cell counts within 1 week and HIV-RNA viral load within 1 month were used in the analysis, and positive result for real-time HIV-RNA was defined as $\geq$40 copies per milliliter. Sexual behavior was defined as men who have sex with men or heterosexuality. Furthermore, immediately following colonoscopy, rectal swabs (DNAPAP cervical sampler; Qiagen, Gaithersburg, MD) were obtained. Rectal samples were analyzed for HPV-DNA and genotyping by means of polymerase chain reaction-invader assay as described previously.[22] HPV types 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, and 59 were defined as oncogenic HPV.[23]

Diagnosis of Colorectal Adenoma, Adenocarcinoma, and Non-Neoplastic Polyps

After intestinal lavage with 2 L of solution containing polyethylene glycol, colonoscopy was performed by experienced gastroenterologists by using an electronic high-resolution video endoscope (model CFH260; Olympus Optical, Tokyo, Japan). The location of all lesions was recorded in electronic endoscopic database (Olympus Medical Systems; Solemio Endo). All visualized lesions were biopsied and histologically assessed by experienced pathologists.

Statistical Analysis

Baseline characteristics were compared using the unpaired Student t test or χ^2 test (Fisher exact test) for quantitative or qualitative variables, respectively. To estimate the effect of HIV infection on adenoma, multivariate logistic regression analysis was performed adjusted for age, sex, and possible risk factors for adenoma (these included smoking and alcohol consumption, diabetes mellitus, coronary artery diseases, and NSAIDs and aspirin use). In addition, we conducted uni- and multivariate logistic regression analysis in HIV-infected patients to elucidate the impact of other factors on adenoma related to HIV-infected patients (CD4 count, HIV-RNA, history of HAART, sexual behavior, and oncogenic HPV infection).

Statistical significance was defined at 2-sided P values < 0.05. We estimated the odds ratios (ORs) and 95% confidence intervals (CIs). All statistical analyses were performed using the Statistical Package for Social Sciences version 17.0 (SPSS, Chicago, IL).

RESULTS

Participants

A total of 177 HIV-infected patients and 177-HIV-negative controls were selected for analysis after the application of the aforementioned exclusion criteria and age matching (Fig. 1). The baseline characteristics are listed in Table 1. The study subjects were mostly men, Asians, and comparatively young. HIV-infected patients were more likely to be smokers

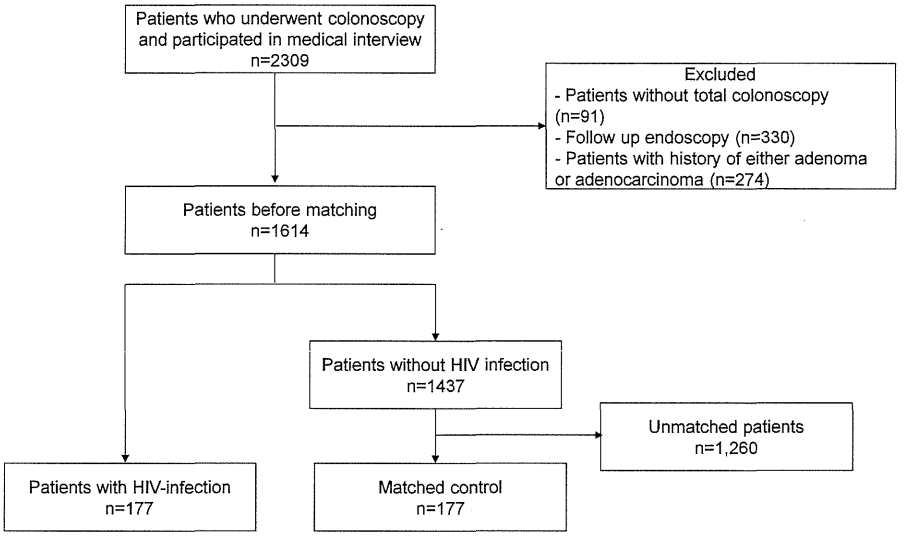

FIGURE 1. Flow diagram of patient selection.

and on treatment with NSAIDs. In contrast, aspirin was mostly used by the control subjects. All other major background parameters were similar in the 2 groups. With regard to the clinical symptoms, there was no difference in GI symptom scores other than increased passage of stools but there was no difference in the proportion of asymptomatic patients between the 2 groups. In patients with HIV infection, the median CD4 count was 371/μL (interquartile range, 121–579), 29.4% of the patients were treatment naive, 75.4% had HPV infection, and 71.5% were infected with oncogenic HPV. The most frequently identified HPV types were type 16 (41%), followed by type 58 (35%), 59 (33%), 52 (27%), 31 (25%), 33 (25%), 51 (19%), 18 (18%), 35 (14%), 39 (13%), 56 (11%), and type 45 (7%).

Prevalence of Colorectal Adenoma, Adenocarcinoma, Non-Neoplastic Polyps, and Other Tumors

Adenomas were identified in 29 (16.4%) patients with HIV infection and in 40 (22.6%) control subjects, and the incidence was not significantly different between the 2 groups (Table 2). Classification of the adenoma according to size (<5, 5–9, and ≥10 mm) showed that HIV-negative subjects tended to have mainly adenomas measuring <5 mm (P = 0.08) although this difference did not reach statistical significance. The incidences of adenocarcinoma and hyperplastic polyps were higher in patients without HIV infection, although the differences in the rates were not statistically significant ($P > 0.05$). In contrast, Kaposi's sarcoma was diagnosed only in HIV-infected patients ($P = 0.03$).

Uni- and multivariate analyses showed that HIV infection did not correlate with higher prevalence of adenoma (Table 3, adjusted OR = 0.66; 95% CI: 0.37 to 1.18; P = 0.16). Multivariate analysis identified age as an independent and significant factor associated with increased risk of adenoma (adjusted OR = 1.72; 95% CI: 1.29 to 2.29; $P < 0.01$). All other factors did not correlate with adenoma by multivariate analysis.

Factors Associated With Colorectal Adenoma in Patients With HIV Infection

Age was an independent factor associated with increased risk of adenoma by uni- and multivariate analysis (adjusted OR = 2.28; 95% CI: 1.37 to 3.80; $P < 0.01$; Table 4). High CD4 count, low HIV-RNA, and history of HAART were associated with prevalence of adenoma by univariate analysis, although these factors were not significant on multivariate analysis. Oncogenic HPV infection was not associated with adenoma.

DISCUSSION

This study demonstrated that HIV infection was not an independent risk for colorectal adenoma after adjustment for variables known to be related to adenoma. In HIV-infected patients, only age was associated with increased risk of colorectal adenoma, whereas CD4 count, HIV-RNA, and HPV infection were not associated with adenoma by multivariate analysis. To our knowledge, this is the first study that compared the prevalence of colorectal adenoma between patients with and without HIV infection in Asia.

Previous reports suggested possible relation between HIV infection and increased risk of colorectal adenoma.[12–14] Bini et al[12] investigated the prevalence of adenoma in 2382 patients (165 HIV-infected patients and 2217 controls) who underwent screening sigmoidoscopy. Their study identified a high incidence of adenoma in HIV-infected patients and that the risk of such lesion was higher in patients with low CD4 count and long-term HIV infection. The same group also conducted a prospective study of 408 patients who underwent total colonoscopy in the United States.[13] They included only

TABLE 1. Clinical Characteristics of Patients With and Without HIV Infection

	HIV-Positive Patients (n = 177)	HIV-Negative Patients (n = 177)	*P*
Age, yr (IQR)	42 (37–50)	42 (37–50)	0.99
Male gender (%)	167 (94.4)	167 (94.4)	1.00
Asian (%)	171 (96.6)	176 (99.4)	0.12
Cigarette smoking (%)			
Never smoker	58 (32.8)	78 (44.1)	
Smoking index			
<400	89 (50.3)	60 (33.9)	
400–799	22 (12.4)	25 (14.1)	
>800	8 (4.5)	14 (7.9)	0.02*
Alcohol consumption (%)			
Nondrinker	77 (43.5)	59 (33.3)	
Light drinker	82 (46.3)	86 (48.6)	
Moderate drinker	13 (7.9)	24 (13.6)	
Heavy drinker	5 (2.8)	8 (4.5)	0.09
Current NSAIDs use (%)†	27 (15.3)	13 (7.3)	0.02*
Current aspirin use (%)	3 (1.7)	11 (6.2)	0.03*
Diabetes mellitus (%)	9 (5.1)	17 (9.6)	0.10
Coronary vascular disease (%)	6 (3.4)	5 (2.8)	0.76
Asymptomatic, %	33.5‡	36.6‡	0.55
GI symptoms score			
Increased flatus (SD)	1.9 (1.1)	2.0 (1.4)	0.80
Decreased passage of stools (SD)	1.8 (1.3)	1.8 (1.3)	0.76
Increased passage of stools (SD)	2.7 (2.0)	2.2 (1.6)	0.03*
Loose stools (SD)	2.4 (1.6)	2.1 (1.4)	0.13
Hard stools (SD)	1.7 (1.2)	1.6 (1.0)	0.84
Urgent need for defecation (SD)	2.3 (1.7)	2.1 (1.6)	0.14
Feeling of incomplete evacuation (SD)	2.2 (1.3)	2.2 (1.4)	0.61
CD4 count (IQR)	371 (121–579)	NA	NA
HIV-RNA $\log_{10}$/mL (IQR)	1.6 (1.6–3.8)	NA	NA
Treatment naive (%)	52 (29.4)	NA	NA
MSM (%)	135 (76.3)	NA	NA
HPV infection (%)	98/130 (75.4)	NA	NA
Oncogenic HPV (%)	93/130 (71.5)	NA	NA

*$P < 0.05$.

†None of the patients was on selective cox-2 inhibitor.

‡There were 1 missing data in HIV-positive group and 2 in HIV-negative group, thus comparisons were made between 59/176 (33.5%) of HIV-positive and 64/175 (36.6%) of HIV-negative patients.

IQR, interquartile range; SD, standard deviation; MSM, men who have sex with men; NA, not applicable.

TABLE 2. Prevalence of Colorectal Adenoma, Adenocarcinoma, Non-Neoplastic Polyps, and Other Tumors

	HIV-Positive Patients (n = 177)	HIV-Negative Patients (n = 177)	*P*
Any adenoma	29 (16.4%)	40 (22.6%)	0.14
Adenoma, <5 mm	21 (11.9%)	33 (18.6%)	0.08
Adenoma, 5–9 mm	12 (6.8%)	10 (5.6%)	0.66
Adenoma, ≥10 mm	0	4 (2.3%)	0.12
Adenocarcinoma	0	5 (2.8%)	0.06
Hyperplastic polyp	17 (9.6%)	28 (15.8%)	0.08
Other tumors	6 (33.9%)	3 (17.0%)	0.502
Kaposi's sarcoma	6 (33.9%)	0	0.03*
Malignant lymphoma	0	0	1.00
Carcinoid tumor	0	1 (0.6%)	1.000
Lipoma	0	2 (1.1%)	0.499

*$P < 0.05$.

asymptomatic patients aged 50 years or older and found a high rate of colonic neoplasm, including adenoma, in HIV-infected patients. They also reported that patients with HIV infection who were not on treatment with HAART and those with a positive family history of colorectal cancer were at higher risk for colonic neoplasm. In contrast, the study of Kothari et al,[14] which included 130 HIV-infected patients and 779 controls who underwent screening colonoscopy, did not find significant difference in the prevalence of adenoma between the 2 groups. Similarly, our study showed similar prevalence of adenoma in patients with and without HIV infection. These differences may be explained by differences in sample size, populations, and different inclusion criteria. The abovementioned previous studies included only asymptomatic patients whereas this study included many patients with GI symptoms. Taken together, these results suggest lack of consensus on this issue. Thus, it is still unclear whether HIV infection is truly associated with increased risk of colorectal adenoma. Bini et al[12] suggested that the low immune status associated with HIV infection may enhance the development of adenoma; however, CD4 count did not correlate with adenoma in our study. Furthermore, HIV itself is also suggested to play a role in oncogenesis.[24] There is limited information on this issue, and further studies are needed to clarify the association between HIV infection and colorectal adenoma.

In this study, advanced age correlated with increased risk of adenoma in HIV-infected patients. Excision of adenoma prevents colon cancer and screening colonoscopy is recommended for individuals aged 50 years or older.[8,10,11] However, it has been suggested that colorectal cancer screening is underused in HIV-infected patients.[25] In addition, patients with HIV infection are at higher risk for other GI malignancies such as Kaposi's sarcoma, anal cancer, and GI lymphoma than general population,[26–28] and these patients are sometimes asymptomatic.[28–31] Therefore, we believe that screening colonoscopy is important in HIV-infected patients, especially those aged 50 years or older.

The association between HPV infection and colorectal cancer is controversial.[32] Although 2 recent studies argued against such association, a recent meta-analysis study demonstrated increased risk of colorectal cancer with HPV infection.[17,33,34] Because previous reports suggested increased prevalence of colorectal adenoma in HIV-infected patients, in whom the prevalence of HPV infection is known to be higher than that in the general population,[15] we hypothesized

TABLE 3. Uni- and Multivariate Analysis to Estimate the Risk for Adenoma

	Unadjusted OR (95% CI)	P	Adjusted OR (95%CI)	P
HIV infection	0.67 (0.39 to 1.14)	0.14	0.66 (0.37 to 1.18)	0.16
Age per 10 yrs	1.96 (1.53 to 2.53)	<0.01*	1.72 (1.29 to 2.29)	<0.01*
Male gender	0.71 (0.25 to 2.03)	0.52	0.92 (0.28 to 3.05)	0.89
Smoking	1.60 (1.19 to 2.13)	<0.01*	1.35 (0.98 to 1.86)	0.06
Alcohol consumption	0.89 (0.63 to 1.26)	0.51	0.83 (0.56 to 1.22)	0.34
Current NSAIDs use	0.70 (0.28 to 1.75)	0.45	0.94 (0.35 to 2.53)	0.91
Current aspirin use	4.48 (1.52 to 13.3)	<0.01*	11.8 (0.52 to 6.44)	0.35
Diabetes mellitus	2.37 (1.00 to 5.56)	0.05	1.39 (0.54 to 3.60)	0.49
Coronary heart disease	3.63 (1.08 to 12.3)	0.04	1.30 (0.30 to 5.54)	0.72

*$P < 0.05$.

that oncogenic HPV infection may be a risk factor for adenoma in patients with HIV. However, our results did not find such association.

Fecal occult blood test is a useful screening tool for the detection of colorectal cancers.[10] However, fecal blood test is also positive in various GI diseases such as asymptomatic colitis and Kaposi's sarcoma.[35,36] Thus, the diagnostic accuracy of fecal occult blood test may be less than ideal in HIV-infected patients and accordingly was not used in all subjects in this study. Instead, we assessed the clinical symptoms because we hypothesized that differences in GI symptoms might affect the prevalence of colorectal adenoma. Nevertheless, the proportion of asymptomatic patients was not different between the 2 groups.

Important strengths of this study includes its prospective study design, detailed assessment of GI symptoms and other GI tumors, first study in Asia, and conducting total colonoscopy in all subjects. However, there are several limitations to our study. First, because our study population was younger than those in previous studies, the prevalence might have been underestimated compared with other studies. It is well known that the risk of colorectal cancer increases with age.[37] Thus, the young age of our study subjects and the small sample size of our study could have masked any association between HIV infection and colorectal adenoma. Similar to the study by Bini et al,[13] which examined the relation between HIV infection and colorectal adenoma, larger studies on patients aged 50 years or older will be needed in Asia. Second, because we included both symptomatic and asymptomatic patients who underwent diagnostic colonoscopy, a selection bias could not be ruled out in our study. As a result, it is possible that the control group could have included patients suspected to have colon cancer, whereas HIV-infected patients tended to include those who were referred for colonoscopy based on the suspicion of opportunistic infections, which might have led to the higher prevalence of adenoma in the control group. However, the background characteristics and proportion of asymptomatic patients were similar between the 2 groups. Third, although we collected detailed information on risk factors of adenoma, we could not collect data on factors such as obesity and family history of colon cancer as reported previously,[38,39] and these might have influenced the results.

In conclusion, the incidence of adenoma was not significantly different between patients with and without HIV infection. However, it should be noted that 16.4% HIV-infected patients had adenoma and its risk increased with age. As the issue of aging in patients with HIV infection is growing, the results of this study carry certain significance. Thus, HIV-infected patients should not miss screening opportunities for colorectal adenoma and other HIV-related malignancies.

TABLE 4. Uni- and Multivariate Analyses to Estimate the Risk for Adenoma in HIV-Infected Patients

	Unadjusted OR (95% CI)	P	Adjusted OR (95% CI)	P
Age	2.49 (1.66 to 3.79)	<0.01*	2.28 (1.37 to 3.80)	<0.01*
CD4 count per 10/μL	1.02 (1.00 to 1.03)	0.02*	1.01 (0.99 to 1.03)	0.54
HIV-RNA log10/mL	0.40 (0.21 to 0.76)	<0.01*	0.50 (0.18 to 1.37)	0.18
Treatment naive	0.15 (0.03 to 0.64)	0.01*	1.31 (0.12 to 14.49)	0.83
MSM	0.52 (0.22 to 1.24)	0.14	0.66 (0.19 to 2.26)	0.51
Oncogenic HPV	0.25 (0.10 to 0.65)	<0.01*	0.50 (0.17 to 1.47)	0.21

*$P < 0.05$.
MSM, men who have sex with men.

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