Background

Opportunistic infections such as *Pneumocystis jirovecii* pneumonia (PCP), cytomegalovirus (CMV), non-tuber culous mycobacteria (NTM), and fungal infections are frequently found in patients with acquired immunodeficiency syndrome (AIDS) [1]. The most frequent opportunistic infection among patients with AIDS is CMV infection, which commonly causes retinitis, pneumonia, and gastrointestinal tract ulcers. PCP is also a frequent infectious disease in the lungs of patients with AIDS. Additionally, malignancies such as non-Hodgkin lymphoma (NHL) and Kaposi sarcoma (KS) are significant complications. NHL in particular is not easily controlled and is a frequent AIDS-associated cause of death. Interestingly, KS has only been reported in homosexual patients, and patients with multifocal KS lesions have a poor prognosis.

The introduction of antiretroviral therapy (ART) has drastically changed the incidence of opportunistic infections in patients infected with human immunodeficiency virus 1 (HIV-1), resulting in a decline in mortality rates [2-7]. ART has decreased the frequencies of CMV, PCP, and NTM infections in patients with AIDS [7]; however, the frequency of NHL has not changed dramatically [8]. Additionally, non-AIDS-defining malignancies such as liver, lung, and gastric cancers have been observed in patients with AIDS, regardless of ART [9]. A recent study demonstrated that low CD4 counts at ART initiation was associated with a greater risk of KS and lymphoma, whereas other cancers increased over time with ART, likely reflecting an increased risk of cancer with aging [10], low CD4 counts, and cigarette smoking [11-13].

Although mortality rates have decreased dramatically with the use of ART, its effect in many patients with AIDS is limited, and AIDS-associated complications remain a leading cause of death [14,15]. Additionally, untreated HIV-1-positive patients with severe AIDS-defining illnesses frequently visit hospitals and often rapidly succumb to suddenly aggressive progression of their illness [16,17]. Systematic pathological analysis of autopsy cases can provide useful information related to the cause of death and the distribution of pathogens in patients. However, there have been few reports describing the prevalence of infectious diseases and malignancies in autopsied patients with HIV infection [1,18]. A previous study using samples from autopsied patients with HIV infection during 1982–1998 demonstrated the prevalence of CMV, PCP, and NTM infections decreased during the study period [18]. The same study reported that, although the prevalence of KS was unchanged, the prevalence of NHL increased during the study period [18]. To the best of our knowledge, there are no reports demonstrating changes in the prevalence of opportunistic infections in autopsy cases of HIV infection following the introduction of ART after 2000.

In the present study, autopsy cases of HIV infection in Japan were retrospectively investigated to determine the prevalence of opportunistic infections and malignancies often found in patients with AIDS, including non-AIDS-defining malignancies. Additionally, the association of ART use with the prevalence of opportunistic infections and malignancies was investigated.

Patients and methods

Patients

The present study was approved by the Institutional Review Board of the National Institute of Infectious Diseases (Approval No. 356) and of four hospitals in Japan: Tokyo Metropolitan Komagome Hospital, National Center for Global Health and Medicine, Research Hospital, the Institute of Medical Science, the University of Tokyo, and Osaka National Hospital. Each hospital enrolled in the present study is a central hospital for AIDS treatment in Tokyo and Osaka, and has performed more than 15 autopsies of patients infected with HIV. According to a national autopsy survey by the Japan Pathology Society, 828 patients infected with HIV were autopsied in Japan from 1987–2009. During the period 1985–2009, 215 patients infected with HIV were autopsied at the 4 aforementioned hospitals. Thus, the number of cases in this study covered approximately 26% of all autopsied HIV cases. Ten cases autopsied in the period 2010–2012 were added to the 215 cases, making a total of 225 patients analyzed in this study (Table 1), of which 95.1% were male. The patients' ages at death ranged from 12 to 80 years, with a mean age of 44.4 years (median 44 years). Among them, 35.6% were homosexual, and 29.3% received ART (Table 1). The mean CD4 count at the last blood examination before death was 51.5 cells/μL (range: 0–560 cells/μL; median: 13.5 cells/μL). ART was introduced in Japan in 1997. In this study, ART was defined as any combination of therapy that included two nucleoside or nucleotide reverse transcriptase inhibitors plus a non-nucleoside reverse transcriptase inhibitor, protease inhibitor, or abacavir (another nucleotide reverse transcriptase inhibitor). Additionally, ART (+) patients were defined as patients who received any ART during their lifetime, whereas ART (−) patients were as patients who did not receive ART.

Methods

Pathological findings were collected from autopsy records. CMV infection was determined by the infiltration of large cells with typical inclusion bodies. Infections by other viral agents such as hepatitis B virus, herpes simplex virus, hepatitis C virus, JC virus (causing progressive multifocal leukoencephalopathy), and varicella zoster virus were confirmed by immunohistochemistry or polymerase chain reaction. HIV encephalopathy was defined by morphological features indicating the presence of syncytial

Table 1 Characteristics of the patients infected with HIV

Factors	Groupings	Total patients		ART (–) patients		ART (+) patients		*P* value
		n	%	n	%	n	%	
Total		225*	100%	136	100%	66	100%	
Sex	Male	214	95.1%	128	94.1%	63	95.5%	0.695**
	Female	11	4.9%	8	5.9%	3	4.5%	
Age at death	<10 years	0	0.0%	0	0.0%	0	0.0%	**0.028****
	11–20	2	0.9%	2	1.5%	0	0.0%	
	21–30	30	13.3%	22	16.2%	6	9.1%	
	31–40	60	26.7%	34	25.0%	19	28.8%	
	41–50	69	30.7%	48	35.3%	14	21.2%	
	51–60	34	15.1%	18	13.2%	14	21.2%	
	61–70	24	10.7%	10	7.4%	10	15.2%	
	71–80	5	2.2%	1	0.7%	3	4.5%	
	>81	0	0.0%	0	0.0%	0	0.0%	
	Unknown	1	0.4%	1	0.7%	0	0.0%	
Risk factor	Homosexual	80	35.6%	52	38.2%	24	36.4%	0.800**
	Heterosexual	38	16.9%	24	17.6%	10	15.2%	
	Blood product	37	16.4%	29	21.3%	7	10.6%	
	Other	9	4.0%	5	3.7%	4	6.1%	
	Unknown	61	27.1%	26	19.1%	21	31.8%	
CD4 count before death	<50 cells/μL	122	54.2%	80	58.8%	33	50.0%	0.639***
	51–100	25	11.1%	11	8.1%	14	21.2%	
	101–200	13	5.8%	4	2.9%	9	13.6%	
	201–300	5	2.2%	5	3.7%	0	0.0%	
	301–400	3	1.3%	1	0.7%	2	3.0%	
	>401	4	1.8%	1	0.7%	3	4.5%	
	Unknown	53	23.6%	34	25.0%	5	7.6%	

ART, antiretroviral therapy. ART (+) patients were defined as patients who received any ART during their lifetime, whereas ART (–) patients were as patients who did not receive any ART in their lifetime.
*Total number of patients = 225 and included 23 patients with unknown ART status.
**P values were calculated for the rates of male or homosexuals between ART (–) and (+) patients by Chi-square test.
***P values were calculated for age or CD4 counts between all ART (–) and (+) patients by Mann–Whitney *U*-test. Bold font indicates statistical significance.

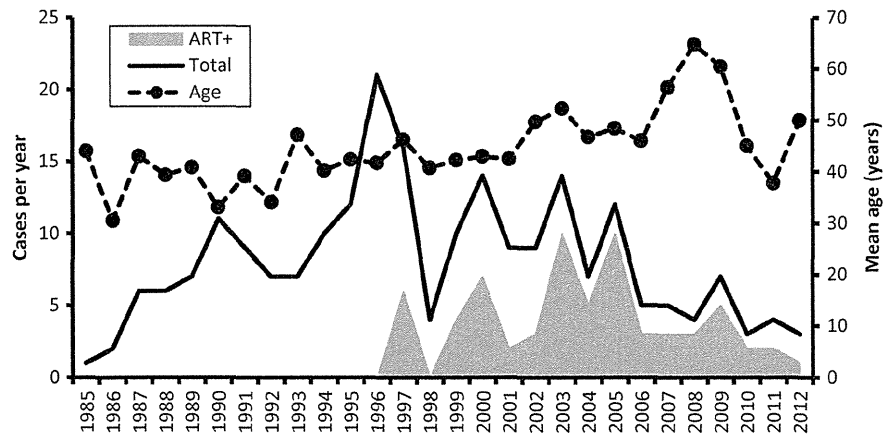

Figure 1 Annual number and mean age of AIDS-related autopsies. The solid line indicates total number of AIDS autopsies in each year. The gray area indicates the number of patients on ART in these autopsy cases. The broken bar indicates the mean age.

giant cells and detection of HIV-1 antigen by immunohistochemistry in the brain. Bacterial infection was identified by Gram stain, and in some cases, species of bacteria were identified by bacterial cultures. Tuberculosis and NTM infection were determined by acid-fast stain and/or PCR. Fungal and protozoan infections such as PCP, toxoplasma, *Candida*, *Aspergillus*, and *Cryptococcus* infection, were determined morphologically using Grocott's methenamine silver stain, periodic acid-Schiff stain, or/and immunohistochemistry. The histological sub-typing of malignant lymphoma was based on the World Health Organization classification, fourth edition. KS was confirmed by immunohistochemistry for Kaposi sarcoma-associated herpesvirus-encoded latency-associated nuclear antigen 1. Causes of death were determined by pathologists at each hospital based on the severity, distribution, and type of illness in the pathological findings of autopsy. Clinical data, such as age at autopsy, sex, risk factors, CD4 cell counts at the last blood examination before death, and use of ART in their lifetime were collected from medical records. Analysis of statistical significance was carried out using Mann–Whitney *U*-test for non-parametric two-sample analysis and Chi-squared test for contingency table analysis.

Results

After the introduction of ART in Japan in 1997, the total number of autopsies conducted on patients with HIV infection has slowly decreased whereas the mean age at autopsy has increased slightly (Figure 1). After 1997, 66

Table 2 Infectious diseases and malignancies in AIDS-associated autopsies

	All patients		ART (−) patients		ART (+) patients			
	n	%	n	%	n	%	*P* values	
Total	225	100.0%	136	100.0%	66	100.0%		
Infectious diseases								
Cytomegalovirus	142	63.1%	97	71.3%	25	37.9%	**<0.001**	
Pneumocystis jirovecii pneumonia	66	29.3%	43	31.6%	11	16.7%	**0.024**	
Non-tuberculous mycobacterium	31	13.8%	20	14.7%	8	12.1%	0.618	
Candida	25	11.1%	17	12.5%	6	9.1%	0.474	
Aspergillus	24	10.7%	17	12.5%	4	6.1%	0.160	
Human immunodeficiency virus encephalopathy	21	9.3%	13	9.6%	6	9.1%	0.915	
Cryptococcus	16	7.1%	11	8.1%	3	4.5%	0.526	Y
Hepatitis B virus	12	5.3%	6	4.4%	5	7.6%	0.549	Y
Herpes simplex virus	12	5.3%	1	0.7%	1	1.5%	0.816	Y
Toxoplasmosis	11	4.9%	9	6.6%	3	4.5%	0.789	Y
Hepatitis C virus	9	4.0%	3	2.2%	5	7.6%	0.147	Y
Progressive multifocal leukoencephalopathy	8	3.6%	4	2.9%	2	3.0%	0.684	Y
Tuberculosis	6	2.7%	4	2.9%	0	0.0%	0.385	Y
Varicella zoster virus	4	1.8%	2	1.5%	2	3.0%	0.835	Y
Multicentric Castleman disease	2	0.9%	1	0.7%	1	1.5%	0.816	Y
Malignancies								
Non Hodgkin lymphoma	71	31.6%	41	30.1%	25	37.9%	0.272	
Kaposi sarcoma	38	16.9%	22	16.2%	10	15.2%	0.852	
Endocervical cancer	0	0.0%	0	0.0%	0	0.0%	-	
Non-AIDS defining malignancies	20	8.9%	10	7.4%	10	15.2%	0.082	
Hepatic cancer	8	3.6%	4	2.9%	4	6.1%	0.495	Y
Lung cancer	6	2.7%	2	1.5%	4	6.1%	0.174	Y
Leukemia	2	0.9%	0	0.0%	2	3.0%	0.200	Y
Hodgkin lymphoma	2	0.9%	1	0.7%	1	1.5%	0.816	Y
Gastric cancer	1	0.4%	1	0.7%	0	0.0%	0.711	Y
Other cancer	3	1.3%	3	2.2%	0	0.0%	0.551	Y

P values were calculated by Chi-square test. Y indicates the use of Chi-square test with Yates correction. Bold font indicates statistical significance. ART, antiretroviral therapy. Because more than one illness was detected in patients, the numbers of all illness are greater than the total number.

of 126 patients (52.6%) received ART during their lifetime. The mean age at death of patients on ART was 47.3 years, which was significantly higher than that of ART naïve patients (42.6 years; $P = 0.028$; Mann–Whitney *U*-test). Mean CD4 counts of ART (−) and (+) patients at the last blood examination before death were not significantly different (39.6 and 77.0 cells/μL, respectively, $P = 0.63$, Mann–Whitney *U*-test).

CMV was the most commonly identified pathogen among the autopsy cases (Table 2) and was detected in various organs, the most frequent being the adrenal gland (Figure 2A). PCP and NTM were also common pathogens found in the lungs of autopsied patients. *Candida albicans* was frequently detected in the gastrointestinal tract and oral cavity (Figure 2B). The prevalence of CMV and PCP was significantly lower in ART (+) patients than in ART (−) patients (Table 2). There was no significant difference in the prevalence of other opportunistic infections such as NTM and *Candida* or prevalence of HIV encephalopathy between ART (+) and (−) patients (Table 2).

Malignancies were identified in 50.2% (113/225) of all cases (Table 2). NHL was the most frequent malignancy with a lower prevalence in ART (−) patients (30.1%) than ART (+) patients (37.9%); however, the difference was not significant (Table 2). Diffuse large B-cell lymphoma was the most frequent histological subtype of NHL followed by Burkitt lymphoma, primary effusion lymphoma, and plasmablastic lymphoma (Table 3). Epstein–Barr virus positivity in lymphoma cases was significantly lower in ART (+) patients compared with ART (−) patients ($P = 0.001$, Chi-square test). KS was frequently found in the skin as well as other sites such as the gastrointestinal tract upon autopsy. In addition to NHL and KS, non-AIDS-defining malignancies such as Hodgkin lymphoma (HL), hepatic cancer, lung cancer, and leukemia were also observed in 20 patients. The prevalence of non-AIDS-defining malignancies was higher in ART (+) patients compared with ART (−) patients (Table 2).

The lung was the most frequent target for pathogens in patients with AIDS and 173 (76.9%) autopsy cases demonstrated the presence of lung-related illnesses (Table 4), which were significantly more frequent in ART (−) patients (112/136, 82.4%) than ART (+) patients (42/66, 63.6%) ($P = 0.003$, Chi-square test). CMV then PCP was the most frequently observed lung-related illnesses. The brain was the second most frequently affected organ in the autopsy cases. Although the brain was not investigated in 53 autopsies, 85 of the remaining 172 cases (49.4%) had brain-related illnesses, with CMV infection the most common, followed by lymphoma and HIV encephalopathy (Table 5). However, there was no significant difference in the rate of brain-related illnesses in ART (+) (37.8%, 17 of 45) or ART (−) patients (52.7%, 58/110) ($P = 0.091$, Chi-square test).

We also investigated the direct causes of death in the autopsied patients (Table 6). Lymphoma was the most frequent cause of death, followed by CMV infection. Non AIDS-defining cancers as a cause of death were significantly different between ART (−) (2, 1.5%) and ART (+) patients (6, 9.1%) ($P = 0.026$; Chi-square test with Yates correction). The prevalence of CMV, pneumonia, PCP, and NTM as a cause of death were lower in ART (+) patients compared with ART (−) patients, but no significant differences were observed between the groups.

Discussion

In the present study, we measured the prevalence of infectious disease and malignancy in autopsy cases of

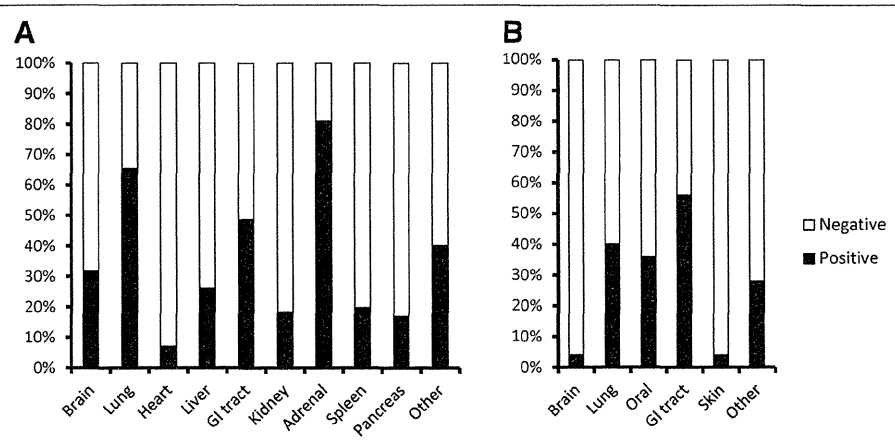

Figure 2 Distribution of cytomegalovirus and *Candida albicans*. **(A)** CMV positive rate in each organ. Black bar indicates the CMV positive rate in each organ from 142 CMV-positive patients. Because CMV was detected in more than one organ per patient, the sum of the black bars is over 100%. **(B)** The positive rate of *Candida albicans* in each organ. Black bar indicates the positive rate of *Candida albicans* per organ from 25 *Candida albicans*-positive patients.

Table 3 Non-Hodgkin lymphoma and Kaposi sarcoma in AIDS-associated autopsies

		Total		ART (−) patients		ART (+) patients		*P* values	
		n	%	n	%	n	%		
All NHL cases		71	100.0%	41	100.0%	25	100.0%		
Histology	DLBCL	53	74.6%	30	73.2%	18	72.0%	0.917	
	BL	4	5.6%	3	7.3%	1	4.0%	0.987	Y
	PEL	5	7.0%	4	9.8%	1	4.0%	0.706	Y
	PBL	1	1.4%	1	2.4%	0	0.0%	0.801	Y
	Other	6	8.5%	2	4.9%	4	16.0%	0.279	Y
	Unknown	2	2.8%	1	2.4%	1	4.0%	0.703	Y
Site	Nodular	1	1.4%	0	0.0%	1	4.0%	0.801	Y
	Extranodular	45	63.4%	28	68.3%	12	48.0%	0.102	
	Both	21	29.6%	11	26.8%	10	40.0%	0.265	
	Unknown	4	5.6%	2	4.9%	2	8.0%	0.987	Y
PCNS	Yes	27	38.0%	18	43.9%	6	24.0%	0.103	
EBV	Positive	52	73.2%	35	85.4%	12	48.0%	**0.001**	
KSHV	Positive	6	8.5%	5	12.2%	1	4.0%	0.495	Y
Cause of death	Yes	50	70.4%	33	80.5%	16	64.0%	0.137	
All KS cases		38		22		10			
Site	Skin	32	84.2%	19	86.4%	9	90.0%	0.410	
	GI tract	27	71.1%	15	68.2%	8	80.0%	0.705	
	Lung	21	55.3%	11	50.0%	6	60.0%	0.799	
	Lymph node	20	52.6%	13	59.1%	6	60.0%	0.502	
	Other	16	42.1%	0	0.0%	0	0.0%	0.787	Y
Cause of death	Yes	11	29.0%	7	31.8%	2	20.0%	0.791	Y

NHL, Non-Hodgkin lymphoma; DLBCL, diffuse large B-cell lymphoma; BL, Burkitt lymphoma; PEL, primary effusion lymphoma; PBL, plasmablastic lymphoma; PCNS, primary central nervous system lymphoma; EBV, Epstein–Barr virus; KSHV, Kaposi sarcoma-associated herpes virus; KS, Kaposi sarcoma; GI, gastrointestinal. *P* values were calculated by Chi-square test. Y indicates the use of Chi-square test with Yates correction. Bold font indicates statistical significance.

Table 4 Lung disease in patients infected with HIV

Illness	n	% of total patients (n = 225)
Any illness	173	76.9%
Cytomegalovirus infection	93	41.3%
Pneumocystis jirovecii pneumonia	66	29.3%
Any bacterial pneumonia	31	13.8%
Aspergillus infection	23	10.2%
Kaposi sarcoma	21	9.3%
Non-tuberculous mycobacterium infection	14	6.2%
Cryptococcosis	11	4.9%
Candida infection	10	4.4%
Tuberculosis	4	1.8%

Because more than one illness was detected in patients, the numbers of all illness are greater than the total number.

HIV-infected patients identified from 1985–2012 at four central hospitals in Japan. CMV infection, PCP, NTM infection, NHL, and KS were frequently observed in the autopsy cases. The prevalence of CMV and PCP was lower in ART (−) patients compared with ART (+) patients. The prevalence of non-AIDS defining malignancies was higher among ART (+) patients than ART (−) patients, suggesting that the onset of various opportunistic infections and malignancies should be carefully monitored regardless of whether the patient is receiving ART.

The autopsy cases in the present study were predominantly male (95.1%, Table 1). Additionally, more than 70% of the autopsy cases in the present study had a CD4 count < 200 cells/μL at the last blood examination before death (Table 1). A recent clinical study demonstrated the incidence of AIDS-defining illnesses in patients with HIV infection was decreased by the introduction of ART, especially in patients with CD4 counts >200 cells/μL [2]. Thus, our findings at autopsy cannot be compared with previous clinical studies because many clinical study patients had a high range of CD4 counts and ART responses. Interestingly, there was no significant difference in the

Table 5 Brain disease in patients infected with HIV

Illness	n	% in total autopsied brains (n = 172)
Any illness	85	49.4%
Cytomegalovirus infection	45	26.1%
Malignant lymphoma	26	15.1%
HIV encephalopathy	21	12.2%
Progressive multifocal leukoencephalopathy	8	4.7%
Toxoplasmosis	8	4.7%
Non-tuberculous mycobacterium infection	4	2.3%
Aspergillus infection	2	1.2%
Varicella zoster virus infection	2	1.2%
Herpes simplex virus infection	1	0.6%
Glioblastoma	1	0.6%
Candida infection	1	0.6%

Because more than one illness was detected in patients, the numbers of all illness are greater than the total number.

cause of death between ART (+) and (−) patients, with the exception of those with cancer (Table 6), indicating the prevalence of lethal illness did not differ between ART (+) and (−) patients.

Malignancies were frequent causes of death in the present study regardless of ART status (Table 6). Several studies demonstrated that the introduction of ART reduced the incidence of NHL in patients with HIV infection [13,15,19-23]. The use of ART has also been associated with a decrease in the incidence of KS [15,24,25]. However, an association between the incidence of non-AIDS-defining cancers and ART remains controversial. An increase of non-AIDS-defining cancers in patients receiving ART was shown in previous clinical reports [26,27], but a separate study showed that, with the exception of long-term protease inhibitor usage, ART exposure was generally not associated with a risk of non-AIDS-defining cancers [28]. The reasons for increased risk of non-AIDS-defining cancers in patients on ART are unclear, but might reflect the concomitant increase of the mean age at autopsy during the study period. This suggests that life extension of HIV-infected patients by ART results in the increased chance of developing non-AIDS events and malignancies. It was also

Table 6 Cause of death in AIDS-associated autopsies

	All		ART (−) patients		ART (+) patients			
	n	%	n	%	n	%	*P* values	
Total*	225	100.0%	136	100.0%	66	100.0%		
Malignant lymphoma	50	22.2%	33	24.3%	16	24.2%	0.997	
Cytomegalovirus	44	19.6%	27	19.9%	9	13.6%	0.279	
Pneumonia	31	13.8%	19	14.0%	9	13.6%	0.949	
Pneumocystis jirovecii pneumonia	30	13.3%	21	15.4%	4	6.1%	0.058	
Non-tuberculous mycobacterium	12	5.3%	10	7.4%	2	3.0%	0.367	Y
Kaposi sarcoma	11	4.9%	7	5.1%	2	3.0%	0.749	Y
Progressive multifocal leukoencephalopathy	8	3.6%	4	2.9%	2	3.0%	0.684	Y
Cancer	8	3.6%	2	1.5%	6	9.1%	**0.026**	Y
Hepatitis	8	3.6%	3	2.2%	4	6.1%	0.320	Y
Cryptococcus	7	3.1%	6	4.4%	0	0.0%	0.197	Y
Kidney failure	7	3.1%	4	2.9%	3	4.5%	0.861	Y
HIV encephalopathy	7	3.1%	5	3.7%	2	3.0%	0.861	Y
Aspergillus	6	2.7%	5	3.7%	0	0.0%	0.274	Y
Toxoplasmosis	4	1.8%	3	2.2%	1	1.5%	0.835	Y
Tuberculosis	3	0.9%	2	1.5%	0	0.0%	0.816	Y
Sepsis	3	1.3%	2	1.5%	1	1.5%	0.551	Y
Candida	3	1.3%	2	1.5%	0	0.0%	0.816	Y
Varicella zoster virus	2	1.3%	1	0.7%	1	1.5%	0.816	Y
Nocardia	1	0.4%	1	0.7%	0	0.0%	0.711	Y
Histoplasma	1	0.4%	1	0.7%	0	0.0%	0.711	Y

*Because more than one illness was detected in patients, the numbers of all illness are greater than the total number.
HIV, human immunodeficiency virus. *P* values were calculated with the Chi-square test. Y indicates the use of Chi-square test with Yates correction. Bold font indicates statistical significance.

demonstrated that ART introduction changed the pathological features of lymphoma; for example, a decrease of Epstein–Barr virus-positive lymphoma in Japanese patients with AIDS was reported [29]. Although HL was rare in the general Japanese population compared with European countries and the United States [30], the incidence of HL increased in Japanese patients on ART [17]. Thus, the increased risk of malignancies during the clinical course of HIV infection in patients receiving ART was reflected as a cause of death in the autopsy cases used in our study.

The prevalence of opportunistic infections differs among various regions and countries. In sub-Saharan African countries, more than 80% of HIV-positive patients die of infectious diseases, with disseminated tuberculosis being the most common (36%) [31]. Furthermore, there was no difference in the type of disease HIV patients succumbed to, regardless of ART status. In the USA and European countries, tuberculosis/NTM represented <10% of mortality in autopsy cases after 1996 [18]. In this study, tuberculosis was detected in only 2.7% of Japanese autopsy cases, but was the cause of death for 50% of afflicted patients. Mortality by PCP has decreased worldwide in patients with AIDS owing to prophylactic administration of an anti-PCP drug [16]. PCP was found in 36.4% (36/99 cases) of patients with AIDS before 1997, but was significantly reduced after 1997 (23.8%; 30/126 cases; $P = 0.04$; Chi-square test). This suggests that the decrease in PCP cases is associated with ART and anti-PCP prophylaxis.

Our study had several limitations. Bacterial culture was not available in this study owing to the use of formalin-fixed paraffin-embedded samples, and it was therefore difficult to identify the bacterial species responsible for many cases of pneumonia. Additionally, clinical information was limited. Information on HIV-RNA, an important indicator of ART effects, was not available for these patients. In addition, information regarding CD4 counts and the type, duration and possible interruption of ART were not available for a subset of patients. Therefore, we could not identify cases of immune reconstitution syndrome. Age at seroconversion and time living with HIV are also major predictors of HIV disease progression, however information of these parameters was limited. Thus, it should be noted that the conclusions in this study cannot be generally applied to the current HIV positive population in Japan. Furthermore, all findings in this study were obtained from autopsies.

Conclusions

Although further studies are required to demonstrate the association between ART and illness identified at autopsy, the present study demonstrates the prevalence of infectious diseases and malignancies in autopsy cases of HIV infection in Japan. While the prevalence of CMV infection and PCP at autopsy were lower in ART (+) patients than ART (−) patients, non-AIDS–defining malignancies were observed as a cause of death more frequently in ART (+) patients than ART (−) patients.

Abbreviations

HIV: Human immunodeficiency virus; ART: Antiretroviral therapy; AIDS: Acquired immunodeficiency syndrome; PCP: *Pneumocystis jirovecii* pneumonia; CMV: Cytomegalovirus; NTM: Non-tuberculous mycobacteria; NHL: Non-Hodgkin lymphoma; KS: Kaposi sarcoma.

Competing interests

The authors declare no conflicts of interests.

Authors' contributions

HK, S Okada and AY conceived this study; TH, MM, YKod, NO, YO, SM, TI, HH, and HK performed the autopsies, pathological analyses and reviews; AA, KT, JT, YKi, TU, TS, TK, AI, and S Oka collected clinical data; HK analyzed the data, performed statistical analyses, and drafted the manuscript. All authors read and approved the final manuscript.

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Original Article

Clinical Outcomes of AIDS-related Burkitt Lymphoma: A Multi-institution in Japan

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Objective: Acquired immunodeficiency syndrome-related non-Hodgkin lymphoma is treated similarly to non-acquired immunodeficiency syndrome lymphoma, but it is not clear whether highly intensive regimens are beneficial for acquired immunodeficiency syndrome-related Burkitt lymphoma. We conducted a multicenter retrospective survey to clarify the clinical outcomes of acquired immunodeficiency syndrome-related Burkitt lymphoma in the combined antiretroviral therapy era in Japan.

Methods: We retrospectively analyzed the outcome of 33 patients with acquired immunodeficiency syndrome-related Burkitt lymphoma, who were diagnosed at five regional hospitals for human immunodeficiency virus/acquired immunodeficiency syndrome in Japan between January 2002 and December 2010.

Results: The median follow-up period was 20.0 months (range 0.5–92.7 months). Six (18.2%) patients were treated with cyclophosphamide, vincristine, doxorubicin, high-dose methotrexate, ifosphamide, etoposide and high-dose cytarabine, and 23 (69.7%) patients were treated with hyperfractionated cyclophosphamide, vincristine, doxorubicin, dexamethasone, high-dose methotrexate and high-dose cytarabine. The overall response rate for all patients was 78.8%, with a complete response rate of 72.7%. The two-year overall survival rate was 68.1%. There was no significant difference in overall survival between chemotherapeutic regimens with rituximab ($n = 20$) and without rituximab ($n = 13$) ($P = 0.49$). The two-year overall survival rate was 66.7% for patients receiving cyclophosphamide, vincristine, doxorubicin, dexamethasone, etoposide, ifosfamide and cytarabine, and was 72.6% for patients receiving cyclophosphamide, vincristine, doxorubicin, dexamethasone, methotrexate and cytarabine ($P = 0.72$). There was one treatment-related death.

Conclusions: Highly intensive chemotherapy would bring a high remission rate and prolonged overall survival for patients with acquired immunodeficiency syndrome-related Burkitt lymphoma.

Key words: Burkitt lymphoma – AIDS-related – chemotherapy – rituximab – survival

INTRODUCTION

Combination antiretroviral therapy (cART) has dramatically reduced the occurrence of opportunistic infections such as acquired immunodeficiency syndrome (AIDS)-defining illnesses (ADIs). Decreases in the incidence of AIDS-related lymphoma (ARL) are not as evident compared with other ADIs; thus, lymphoma is now a more common cause of AIDS-related mortality. Adult Burkitt lymphoma (BL) comprises 1–5% of cases of non-Hodgkin lymphoma (1). However, BL in human immunodeficiency virus (HIV) patients is a typical AIDS-defining cancer (ADC) and accounts for 25–40% of ARLs (2–4). AIDS-related BL (AIDS-BL) occurs even in subjects with higher CD4 + lymphocyte counts (3, 5–10), and is therefore important even in patients who are receiving effective therapy for HIV. AIDS-BL progresses very rapidly and is associated with a poor prognosis. In spite of the widespread use of cART, the survival of patients with AIDS-related diffuse large B-cell lymphoma (DLBCL) and that of patients with AIDS-BL remain poor (11, 12). Recent studies have demonstrated that highly intensive chemotherapeutic regimens, such as cyclophosphamide, vincristine, doxorubicin, dexamethasone, etoposide, ifosfamide and cytarabine (CODOX-M/IVAC) or cyclophosphamide, vincristine, doxorubicin, dexamethasone, methotrexate and cytarabine (hyper-CVAD/MA), have improved the survival of patients with non-HIV BL. Thus, the poor outcomes associated with AIDS-BL in previous reports might have resulted from the use of less intensive treatment strategies, such as cyclophosphamide, doxorubicin, vincristine and prednisone (CHOP)-based chemotherapy (13–15). The introduction of cART has resulted in improved general health for patients with HIV; thus, the eligibility of these patients to receive cytotoxic agents has increased to the point where it is nearly similar of that of the non-HIV population, and these patients now would be candidates for highly intensive chemotherapy regimens. The CD20-directed monoclonal antibody, rituximab can improve survival when added to standard chemotherapy in patients with low-grade B-cell lymphomas and DLBCL (16–18). Also the addition of rituximab in non-HIV BL has been examined by randomized clinical trials. However, the impact of rituximab for patients with AIDS-BL patients is unknown. Therefore, we performed a nationwide retrospective survey to elucidate the clinical outcomes and to identify significant prognostic variables for AIDS-BL in the cART era.

PATIENTS AND METHODS

Patients

This retrospective study examined the clinical outcomes of all untreated patients with AIDS-BL who visited one of five regional hospitals for HIV/AIDS in Japan, from January 2002 to December 2010. The pathological diagnosis of each institution was accepted. The pathological diagnosis of BL was based on the 2008 World Health Organization (WHO) classification. AIDS-BL was defined as immunodeficiency-associated BL of the 2008 WHO classification. Examination for *myc* gene alterations was not mandatory.

Chart reviews were performed for all identified patients to obtain the following information: age, sex, performance status (PS) at diagnosis, number of CD4 + cells at diagnosis, prior AIDS, concurrent opportunistic diseases, prior cART, clinical stage, lactate dehydrogenase (LDH), the International Prognostic Index (IPI), disease site, presence of central neurological system (CNS) involvement, presence of bone marrow (BM) involvement, chemotherapy regimen, tumor response and clinical outcomes. This study received approval from the appropriate ethics committees.

Definitions of Terms and Treatment

The PS was evaluated according to the Eastern Cooperative Oncology Group scale (19). Clinical stage was assessed using the Ann Arbor staging system (20). The IPI was assessed according to the International Non-Hodgkin Lymphoma Prognostic Factors project (21). The response was assessed according to the International Workshop Criteria for non-Hodgkin lymphoma (22). Patients were classified according to tumor response: complete response (CR), CR unconfirmed (CRu), partial response (PR), stable disease (SD) or disease progression (PD). The overall response rate (ORR) was calculated as the proportion of patients who achieved a CR, CRu or PR.

The CODOX-M/IVAC regimen consisted of cyclophosphamide, vincristine, doxorubicin and high-dose methotrexate, alternating with etoposide, ifosfamide and high-dose cytarabine (23). The hyper-CVAD/MA regimen consisted of hyperfractionated cyclophosphamide, vincristine, doxorubicin and dexamethasone, alternating with high-dose methotrexate and cytarabine (24). The EPOCH regimen consisted of etoposide, vincristine, doxorubicin, cyclophosphamide and prednisone (25). These regimens and rituximab combinations were chosen according to each institutional decision.

Statistical Analyses

Progression-free survival (PFS) was defined as the time between the date of initial chemotherapy until the date of PD or death from any cause. The absence of PD was treated as a censored observation. Overall survival (OS) was defined as the time from the date of initial chemotherapy to the date of death due to any cause. Patients without such events were treated as censored observations. PFS and OS were estimated using the Kaplan–Meier method, and survival curves were compared using the log-rank test. Univariate Cox regression analyses were used to estimate the hazard ratios and 95% confidence intervals. A two-sided $P < 0.05$ was considered to indicate statistical significance. Subgroups stratified by clinical variables were compared using the log-rank test. All statistical