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Prolonged survival without neurological improvement in patients with AIDS-related progressive multifocal leukoencephalopathy on potent combined antiretroviral therapy

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To evaluate the benefit of combined antiretroviral therapy including protease inhibitors (CART) on survival time and neurological progression in patients with AIDS-related progressive multifocal leukoencephalopathy (PML), 81 consecutive PML cases, collected between January 1990 and June 1998, were reviewed. Fifteen patients were neuropathologically proven. JC virus detection in CSF was positive in 59 patients. At PML diagnosis, median CD4 cell count was low (median, 35 cells/μL) and plasma HIV load, determined in 41 patients, was high (median, 4.8 $\log_{10}$ copies/ml). Following PML diagnosis, there was a significant difference ($P<10^{-4}$) in survival between patients who were untreated or treated with nucleoside analogs (n=50, median: 80 days) and patients who were started early on CART (n=23, median: 246 days). A third group of eight patients who received CART late during the course of PML was considered separately. At the study endpoint, 18 of all the CART-treated patients (n=31) were still alive. Plasma HIV load was undetectable in 67% of them. The median increase in CD4 cell count was 112 cells/μL from CART onset. In contrast, no significant improvement in neurological status was observed. Our results demonstrate a benefit of CART on survival of AIDS-related PML patients and suggest the need for an early, specific anti-JC virus treatment to limit the neurological deterioration.

Keywords: HIV; PML; JCV; demyelination; cidofovir; antiHIV agents

Introduction

Progressive multifocal leukoencephalopathy (PML) is a demyelinating disease of the central nervous system caused by the JC virus (JCV). The first description of PML as an opportunistic disease in association with acquired immunodeficiency syndrome (AIDS) was reported in 1982 (Miller *et al*, 1982). Consecutive to the human immunodeficiency virus (HIV) epidemic, the occurrence of PML has greatly increased during the past 15 years. PML currently affects 1–4% of AIDS patients (Krupp *et al*, 1985; Berger *et al*, 1987; Gillepsie *et al*, 1991; Holman *et al*, 1991; Major and Ault 1995).

Median survival time after PML diagnosis is about 4 months in AIDS patients (Gillepsie *et al*, 1991; Berger and Concha, 1995; Fong *et al*, 1995a). Recent reports have indicated that patients on combined antiretroviral therapy (CART) including protease inhibitors (PI) have prolonged survival (Power *et al*, 1997; Elliot *et al*, 1997; Domingo *et al*, 1997; Baqi *et al*, 1997; Albrecht *et al*, 1998; Cinque *et al*, 1998). Here we report a large retrospective series of 81 patients with AIDS-related PML. The aim of this observational study was to compare survival time and neurological progression according to the different antiretroviral therapy (ART) regimens used.

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Results

Clinical and laboratory features of the study population

Among the 113 HIV patients diagnosed with PML during the study period, only 81 patients were eligible for the present study according to our radioclinical criteria. PML diagnosis was virologically confirmed in 59 patients. Neuropathological confirmation was obtained in 15 patients following brain biopsy (eight) or autopsy (seven), including ten with positive JCV detection by PCR. Therefore, a total of 64 patients were considered as having confirmed PML on virological or neuropathological grounds. Seventeen patients had a diagnosis of PML based only on radioclinical criteria. Most of these 81 patients were men (88%). The mean age was 36 years (range: 24–70 years). The mode of exposure for HIV infection was intravenous drug use (IDU) in 44 patients, homo or bisexual contact in 24 males, heterosexual contact in eight patients and blood transfusion in five patients. The median delay between clinical onset and PML diagnosis was 2 months. PML was the first AIDS-defining event in 47 patients (58%). Table 1 displays neurological features and topography of brain lesions on initial (magnetic resonance imaging) MRI (75 patients) or (computed tomographic scanning) CT scan (six patients). The most common initial signs and symptoms were weakness, speech disorders, incoordination and visual defects. The brain lesions on first MRI (or CT scan) were mainly located in the supratentorial area (73%) as against the infratentorial area (27%). Unilateral hemispheric lesions were discovered in 41 patients (51%), exclusive posterior fossa lesions in 14 patients (17%) and directly multifocal lesions in 26 patients (32%). A faint and irregular contrast enhancement was uncommonly observed in only five patients.

Table 1 Clinical and neuroimaging features of 81 AIDS patients with progressive multifocal leukoencephalopathy

	No	*%*
Neurological signs and symptoms at presentation		
Motor weakness	47	58.0
Speech disorders	29	35.8
Incoordination	25	30.9
Visual field deficits	17	21.0
Seizures	16	19.8
Cognitive disturbances	14	17.3
Sensory loss	13	16.0
Brainstem dysfunctions	9	11.1
Headache	9	11.1
Brain disturbance on first neuroimaging		
Sustentorial		
Frontal	52	64.2
Parieto-occipital	42	51.9
Centrum semiovale	40	49.4
Temporal	21	25.9
Internal capsule/Basal ganglia	9	11.1
Posterior fossa		
Cerebellum	23	28.4
Brainstem	22	27.2

Biological features are presented in Table 2. At the time of diagnosis, the median CD4 lymphocyte count was 35 cells/μL (mean: 67 cells/μL; range: 1–663). The CD4 cell count was higher than 100 cells/μL in 21 patients (26%) and exceeded 200 cells/μL in three patients (4%). The median plasma HIV load determined in 41 cases at PML diagnosis was 4.8 $\log_{10}$ copies/mL (range: 2.3–6.2). Cerebrospinal fluid (CSF) JC viral load was determined in 27 patients within 2 months following PML diagnosis and ranged from 3.2–7.0 $\log_{10}$ copies/mL with a median of 4.6 $\log_{10}$ copies/mL.

Course of ART following PML diagnosis

Following PML diagnosis, 22 patients remained either untreated (nine) or on the previous nucleoside reverse transcriptase inhibitor (NRTI) monotherapy (13). For 28 patients, NRTI therapy was

Table 2 Biological and neurological features at baseline of 81 AIDS patients with progressive multifocal leukoencephalopathy

	CD4 cell count (per μL)		*Plasma HIV load ($\log_{10}$ copies/mL)*		*CSF JCV load ($\log_{10}$ copies/mL)*		*EDSS*	
Population	*(No.)*	*Median (Quartiles)*	*(No.)*	*Median (Quartiles)*	*(No.)*	*Median (Quartiles)*	*(No.)*	*Median (Quartiles)*
Total	(81)	35 (10–101)	(41)	4.8 (4.3–5.4)	(27)	4.6 (3.4–5.4)	(81)	5.5 (4.5–6.5)
Confirmed PML	(64)	33 (11–99)	(34)	4.9 (4.0–5.4)	(27)	4.6 (3.4–5.4)	(64)	6.0 (4.75–6.75)
Radioclinical PML	(17)	35 (10–107)	(7)	4.6 (4.4–6.0)			(17)	5.0 (4.5–6.0)
P value†		0.43		0.47				0.12
Untreated or NRTI	(50)	23 (8–51)	(15)	4.7 (4.8–5.4)	(9)	5.0 (4.0–5.7)	(50)	6.0 (5.0–7.0)
Early CART	(23)	84 (24–134)	(22)	4.8 (4.0–5.5)	(14)	4.5 (3.9–5.2)	(23)	4.5 (4.0–6.0)
Delayed CART	(8)	62 (38–140)	(4)	5.0 (4.7–5.8)	(4)	3.5 (3.3–4.4)	(8)	5.25 (4.5–5.75)
Deceased	(63)	27 (8–71)	(28)	4.8 (4.3–5.5)	(16)	5.1 (4.4–5.7)	(63)	6.0 (5.0–7.0)
Alive*	(18)	91 (46–163)	(13)	4.8 (4.0–5.1)	(11)	3.9 (3.3–4.5)	(18)	4.5 (3.5–5.5)
P value†		$<10^{-3}$		0.33		$<10^{-2}$		$<10^{-3}$

EDSS=expanded disability status scale; NRTI=nucleoside reverse transcriptase inhibitor; CART=combined antiretroviral therapy including protease inhibitor. *Alive on September 30, 1998; †Mann-Whitney test.

either modified (13) or started (15). All these 50 patients were similar as regards CD4 cell counts and were pooled for the survival analysis (group A). A total of 31 patients received CART including protease inhibitors (indinavir: 21, nelfinavir: seven, ritonavir - saquinavir: three). Twenty-three of them (group B) were started early on CART within 3 months after PML onset (median: 1 month). The remaining eight patients who were previously on NRTI bitherapy were started on CART late during the course of PML within a period of 8–37 months after PML onset (median: 14.5 months). These eight patients on delayed CART (group C) were still alive at the study endpoint and were considered separately for the survival analysis.

Prolonged survival in PML patients on CART

The median survival was 124 days (range: 15–1446 days) for the total study population. A significant difference (log-rank test, $P<10^{-4}$) in survival was found according to the ART regimens following PML (Figure 1). As shown in Table 3, all the patients from group A died with a median survival of 80 days. The 23 patients in group B had a median survival of 246 days; ten of them (43.5%) were still alive at the endpoint. Similarly, an age below 45 years, a CD4 cell count at baseline higher than 100 cells/μL or an (Expanded Disability Status Scale) EDSS score at baseline below 6.0 were significantly associated with survival prolongation and reduction of crude relative hazard (RH) of death. In contrast, no significant relationship was found between survival and sex, mode of HIV exposure, topography of brain lesions, AIDS status or ART regimen before PML onset. In the multivariate analysis using the Cox regression model (Table 3), the relationship between survival and age ($P<10^{-3}$), CD4 cell count at baseline (P=0.01), EDSS score at baseline (P=0.02) as well as CART ($P<10^{-4}$) was confirmed.

Neurological progression of PML patients on early CART

At the study endpoint, 13 of the 23 patients from group B had died. Their survival (median: 136 days; range: 43–654 days) was significantly shorter ($P<10^{-3}$) as compared to the ten patients who were still alive (median: 539 days; range: 187–734 days). The neurological progression of the ten survivors was variable. As compared to baseline, the neurological status at last point worsened in five patients, improved in four and remains stable in one. No significant change in EDSS score was observed between the last point and the baseline (median: +0.25; quartiles: −1.50 to +2.50) (Figure 2). In contrast, the median increment of CD4 lymphocytes from CART induction to the last point was 78 cells/μL in the survivors and 56 cells/μL in the deceased

Table 3 Survival after progressive multifocal leukoencephalopathy diagnosis: surrounding factors and Cox regression model

		Patients		Survival (days)*		Cox univariate model			Cox multivariate model		
Variables	*Categories*	*No.*	*(Alive)*	*Median*	*[Quartiles]*	*cRH*	*95% CI*	*Global* P *value*	*aRH*	*95% CI*	*Global* P *value*
Sex‡	Males	71	(15)	125	(70–293)	1.00		0.75			
	Females	10	(3)	101	(68–552)	0.88	0.40–1.94				
Age at baseline	⩾45 years	9	(1)	46	(34–111)	1.00			1.00		
	<45 years	72	(17)	132	(74–328)	0.32	0.15–0.68	$<10^{-2}$	0.22	0.09–0.51	$<10^{-3}$
Mode of exposure‡	Transfusion	5	(1)	80	(73–151)	1.00					
	Heterosexual	8	(1)	140	(104–416)	0.41	0.12–1.42				
	H/B sexual	24	(7)	122	(73–400)	0.52	0.17–1.55	0.56			
	IDU	44	(9)	122	(65–289)	0.56	0.20–1.59				
Brain localisation at baseline‡	Sustentorial	41	(7)	144	(73–243)	1.00					
	Posterior fossa	14	(3)	88	(38–320)	1.10	0.55–2.20	0.96			
	Multifocal	26	(8)	122	(75–464)	1.01	0.57–1.79				
EDSS score at baseline	⩾6.0	40	(4)	85	(57–160)	1.00			1.00		
	<6.0	41	(14)	187	(85–602)	0.54	0.33–0.90	0.02	0.52	0.31–0.89	0.02
CD4 cell count at baseline	⩾100	21	(8)	218	(90–602)	1.00			1.00		
	<100	60	(10)	119	(61–203)	1.90	1.03–3.52	0.04	2.31	1.19–4.50	0.01
PML as first AIDS-event‡	yes	47	(13)	122	(70–336)	1.00					
	no	34	(5)	128	(64–218)	0.92	0.56–1.51	0.74			
ART prior to PML onset‡	yes	39	(7)	136	(78–243)	1.00					
	no	42	(11)	123	(66–464)	1.27	0.77–2.09	0.35			
Course of ART after PML diagnosis	untreated or NRTI	50	(0)	80	(54–132)	1.00			1.00		
	early CART	23	(10)	246	(121–552)	0.24	0.13–0.45	$<10^{-4}$	0.28	0.14–0.53	$<10^{-4}$
	delayed CART†	*8*	(*8*)	*1137*	(*895–1286*)						

IDU=intravenous drug user; PML=progressive multifocal leukoencephalopathy; EDSS=expanded disability status scale; NRTI=nucleoside reverse transriptase inhibitor; ART=antiretroviral therapy; CART=combined antiretroviral therapy including protease inhibitor; cRH=crude relative hazard of death; aRH=adusted relative hazard of death; CI=confidence interval. *End-date: September 30, 1998; †excluded for Cox model; ‡excluded for multivariate analysis.

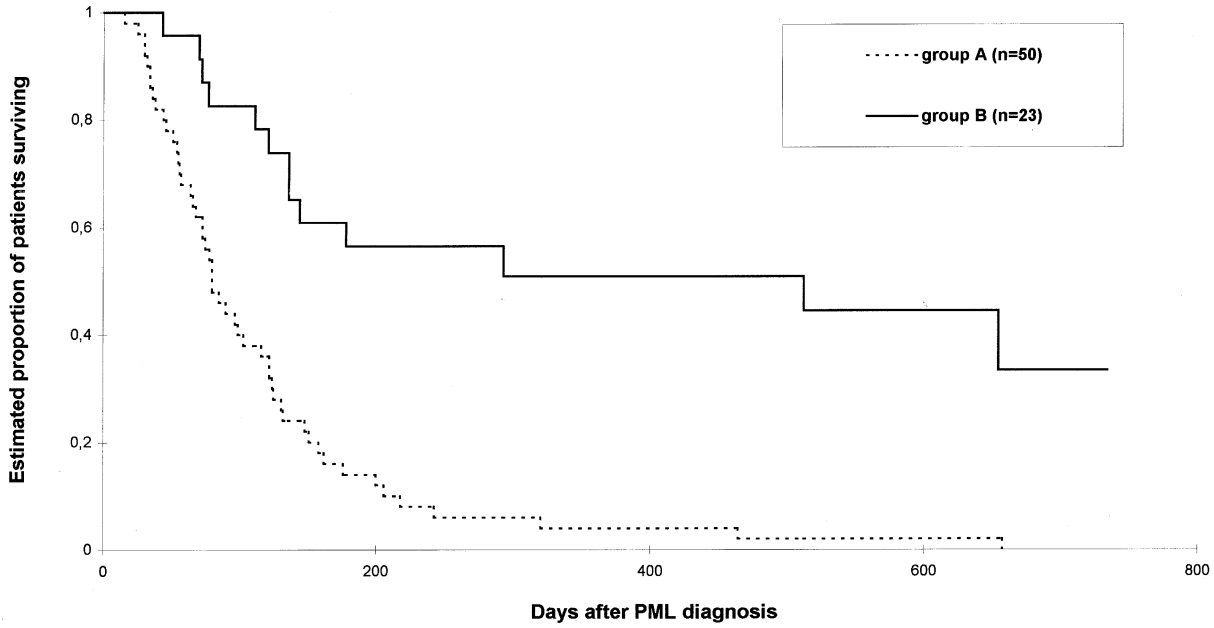

Figure 1 Kaplan Meier survival curve in AIDS-related progressive multifocal leukoencephalopathy. Longer survival time in 23 patients treated with potent combined antiretroviral therapy including protease inhibitors (group B, median survival: 246 days) *versus* 50 patients untreated or on previous nucleoside analogs (group A, median survival: 80 days). Difference is significant by logrank test ($P<10^{-4}$).

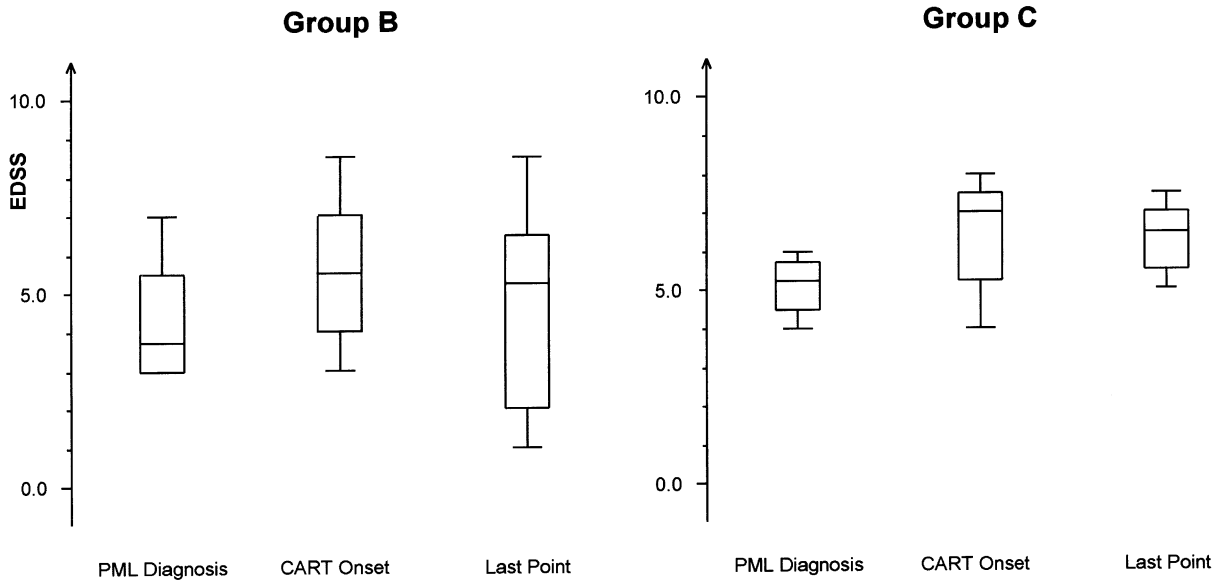

Figure 2 EDSS assessment: Evolution on combined antiretroviral therapy (CART) in 18 living AIDS patients with progressive multifocal leukoencephalopathy (PML). Group B: ten patients started early on CART. No significant change (Wilcoxon signed rank test) in EDSS score from baseline (median: 3.75) to CART onset (5.50) and from CART onset to last point (5.25). Group C: eight patients started later on CART. Significant impairment (P=0.01) in EDSS score between baseline (median 5.25) and CART onset (7.0). No significant change in EDSS score from CART onset to last point (6.5). In both groups B and C, no significant change in EDSS score from baseline to last point.

patients (P=0.13). Plasma HIV load was undetectable (limit of detection: 200 copies/mL) at the last point in eight of the ten survivors and in seven of the 13 deceased.

Long-lived PML patients on delayed CART
The eight patients in group C had a survival exceeding 2.5 years after PML clinical onset (median: 44 months; range: 31–63 months). All were still alive at the study endpoint. All these patients received CART late in the course of PML (median: 14.5 months). At CART onset, they were similar to group B patients in terms of CD4 cell count (median: 56 cells/μL; range: 13–352) and HIV viral load (median: 4.9 $\log_{10}$ copies/mL; range: 2.7–5.0). Plasma HIV load was undetectable at the last point in four of them. As compared to the evaluation at baseline, the neurological status at last point worsened in one patient, improved in four and remains stable in three. No significant change in EDSS score was observed between the last point and the baseline (median: +1.0; quartiles: 0.0 to +2.0).

Specific anti-JC virus treatments
Twenty patients underwent specific anti-JCV therapies. Eleven patients from group A were treated with different regimens of alpha-interferon (five) or cytarabine (six) with no statistically significant improvement in survival. One patient from group B received topotecan infusions (0.3 mg/m^2 for 21 consecutive days per month) and died 178 days after PML diagnosis. Eight patients from group B received cidofovir infusions (5 mg/kg bimonthly); at the study endpoint, no statistically significant difference in survival (P=0.12) was found as compared to the 14 patients on early CART alone.

Discussion

Our study shows a significant benefit of CART on survival, however without clear neurological improvement in a series of 81 patients with AIDS-related PML. Fifty-one of these consecutive cases were observed in the last 4 years. PML was the first AIDS-defining event in 58% of them, in agreement with other series (Gillepsie *et al*, 1991; Fong *et al*, 1995a; Albrecht *et al*, 1998). In this study, diagnosis of PML was based only on radioclinical criteria in 17 patients, including 13 from group A, three from group B and one from group C. At baseline, these patients had similar neurological and biological features as compared to the 64 patients with virological and neuropathological confirmation of PML (Table 2). Exclusion of these 17 patients has not modified study results as regards survival analysis, multivariate analysis of factors affecting RH of death and neurological progression.

The spectrum of initial symptoms and signs observed in our series was similar to that reported in other series of either AIDS- (Gillepsie *et al*, 1991; Von Einsiedel *et al*, 1993; Fong *et al*, 1995a; Albrecht *et al*, 1998; Berger *et al*, 1998a) or non-AIDS-related PML (Brooks and Walker, 1984). MRI contrast enhancement was observed in only 6% of our patients in contrast to the 15% reported in another series (Berger *et al*, 1998a).

At PML diagnosis, plasma HIV loads obtained in 41 patients were high and CD4 cell counts were low (see Table 2), suggesting that PML occurs mainly in the setting of severe immunodeficiency consecutive to ART absence or failure.

Several studies had previously demonstrated the high sensitivity and specificity of JCV DNA detection by PCR in CSF (Moret *et al*, 1993; Gibson *et al*, 1993; Weber *et al*, 1994; Fong *et al*, 1995b; McGuire *et al*, 1995). This method is currently supplanting brain biopsy because it is a less invasive procedure (Weber *et al*, 1996; Cinque *et al*, 1997; Antinori *et al*, 1997). JCV DNA detection by PCR in CSF was a reliable marker for PML diagnosis in our patients.

The natural course of PML is usually rapidly fatal with a median of 4 months from diagnosis to death in our series as in other HIV series (Berger and Concha, 1995) or in other immunosuppressive diseases (Brooks and Walker, 1984). In this series, prolonged survival was correlated with a CD4 cell count at PML diagnosis higher than 100 cells/μL, as reported in other studies (Fong *et al*, 1995a; Von Einsiedel *et al*, 1993; Berger *et al*, 1998b). We had previously shown the prognostic value of CSF JC viral load in PML patients (Taoufik *et al*, 1998b). In the present study, JC viral load, determined in CSF of 27 patients within 2 months following PML diagnosis, was significantly higher in the 16 deceased patients than in the 11 who were alive at the study endpoint (see Table 2).

Prolonged survival, exceeding 2.5 years, was observed in eight patients (group C). These long-lived PML patients had a low CD4 cell count (median: 62 cells/μL; range: 26–266) and a high level of plasma HIV load (median: 5.0 $\log_{10}$ copies/mL; range: 4.6–6.0) at baseline. Interestingly, CSF JC viral load, determined at baseline in four of these patients, was moderate (3.2, 3.3, 3.7 and 4.7 $\log_{10}$ copies/mL). This might explain the unusual PML course, which seemed unrelated to CART, as the latter was initiated with a median delay of 14.5 months following PML onset.

Prolonged survival in PML patients treated with CART has been suggested in anecdotal case reports (Power *et al*, 1997; Elliot *et al*, 1997; Baqi *et al*, 1997) and in series of five patients (Albrecht *et al*, 1998) and ten patients (Cinque *et al*, 1998). Our data confirm the significant improvement of survival time in a large series of 31 patients.

In our series, the 18 survivors from both groups B (early CART) and C (delayed CART) had a median survival of 724 days with an undetectable plasma HIV load in 67% of them and a median increase in CD4 counts of 112 cells/μL following initiation of potent CART. In agreement with a preliminary multicentre study (Clifford *et al*, 1998; Hall *et al*, 1998a), our results show that prolongation of survival is associated with a sustained virological and immunological response to CART. Moreover, CSF JC viral load became undetectable in nine of the 12 tested survivors. Our data support the hypothesis of restoration of an anti-JCV immune response (Taoufik *et al*, 1998b). On the other hand, the median time to death was 3 months in the 63 deceased patients including the 13 on early CART. These results also suggest an initial critical period in the course of AIDS-related PML with a dual potentiality of prognosis. The severe form of PML may be related to more virulent JCV strains that rapidly induce a lethal destruction of cerebral white matter. The absence of significant neurological improvement as we observed in the 18 survivors suggests that CART, as the only PML treatment is not sufficient. The rapid, and often irreversible, deterioration of neurological condition is not prevented by CART. One possible explanation is that the CART-induced immune reconstitution (Autran *et al*, 1997) might require a given latency to be effective. Therefore, an early specific anti-JCV therapy would be of interest to limit extensive destruction of cerebral white matter.

Despite the recent *in vitro* confirmation of cytarabine effectiveness against JCV (Hou and Major, 1998), AIDS Clinical Trials Group 243, a three-armed controlled trial with cytarabine, conducted before the era of CART, failed to demonstrate any benefit of cytarabine in survival of PML patients (Hall *et al*, 1998b). This result may reflect the weak bioavailability of cytarabine in cerebral white matter. In contrast, a clinical trial of alpha-interferon in association with CART might be of interest in light of the survival prolongation of PML observed in a recent open label study (Huang *et al*, 1998). Cidofovir exhibits *in vitro* activity against the polyomavirus SV40 (Andrei *et al*, 1997). Preliminary reports in AIDS-related PML patients treated with CART have suggested a benefit of cidofovir alone (Brosgard *et al*, 1997; Matheron *et al*, 1998) or in association with cytarabine (Sadler *et al*, 1998). At the endpoint of this study, no statistically significant benefit in survival was observed in PML patients started early on CART in association with cidofovir. However, extended follow-up is necessary to draw conclusions on effect of cidofovir in PML progression.

Taken together, these results show a significant benefit of combined antiretroviral therapy including protease inhibitors on survival of AIDS-related PML patients, although without neurological improvement. In this regard, the early association of CART and an efficient specific anti-JCV therapy early after PML onset should be considered in future trials.

Materials and methods

Study design

This study was a chart review collected retrospectively between January 1990 and December 1994 and prospectively from January 1995 through June 1998. The sources of the records were the Department of Internal Medicine and Clinical Immunology (Antoine Béclère Hospital, F-Clamart, France) and the Unit of NeuroAIDS Rehabilitation Care (Department of Internal Medicine and Infectious Diseases, Bicêtre Hospital, F-Le Kremlin-Bicêtre, France).

All subjects were in-patients documented for HIV infection and with clinical AIDS diagnosis according to the CDC 1993-revised definition. PML diagnosis was established by a neurologist on radioclinical grounds according to the following criteria: (1) presence of a focal cerebral disease with a subacute rate of progression; (2) correlated with focal white matter lesions on brain MRI or CT scan consistent with PML (without mass effect or nodular contrast enhancement); (3) prior antitoxoplasmic prophylaxis or failure of acute antitoxoplasmic treatment and (4) no evidence for another etiology during follow-up. Patients with positive JCV DNA PCR in CSF were considered as virologically confirmed cases. Moreover, 15 patients were considered as pathologically diagnosed PML cases on the basis of the histopathological triad: (1) demyelination sometimes associated with necrosis; (2) enlarged bizarre reactive astrocytes and (3) dense amphophilic nuclear inclusions in oligodendrocytes. Papovaviruses were detected in oligodendroglial nuclear inclusions by electronic microscopy (n=6) or by using immunocytochemical staining (n=9) with a rabbit polyclonal SV40 antibody (Oncogene Science Inc, Mineola, NY, USA).

Analysis parameters

As the time of PML clinical onset was not precisely known for all patients, the date of PML diagnosis was used as the baseline for the survival analysis. In this study, the date of the initial brain MRI or CT scan was considered as the date of PML diagnosis. Living patients were right-censored to the study endpoint on September 30, 1998. For patients who died before this date, survival time was defined as the period from baseline to death. The following variables were considered for their relationship to survival: sex, age, mode of HIV exposure, initial distribution of brain lesions, CD4 cell count and plasma HIV viral load at baseline, AIDS status before PML onset, ART regimens before and after PML onset and specific anti-JCV medications.

No standardized neuroimaging protocol was performed in this study. The distribution of brain lesions at PML diagnosis was assigned by correlation of clinical status and neuroimaging findings as supratentorial, infratentorial or directly multifocal location. The neurological disability was measured at baseline by the same neurologist (J Gasnault) with the EDSS (Kurtzke, 1983), a standardized neurological score graded from 0 (normal) to 10 (death). For most patients followed since 1995, the EDSS score was determined every 2 months until the study endpoint. The change in EDSS score was calculated by subtraction of the baseline value from the final score.

Virological methods
Detection of JCV in CSF by PCR assay was performed in 49 patients at the Unit of Virology (Department of Microbiology, Paul Brousse Hospital, F-Villejuif, France). JCV quantitation by PCR in CSF was performed in the same laboratory since 1996, as previously described (Taoufik *et al*, 1998a, b). The threshold of JCV quantitation was 3.2 $\log_{10}$ copies/mL. Twenty-three CSF samples were investigated for JCV detection in other laboratories of virology (Assistance Publique - Hôpitaux de Paris, France). Plasma HIV load was determined by reverse transcriptase PCR assay (Amplicor HIV Monitor kit, Roche Diagnostic Systems, F-Neuilly, France). The limit of HIV quantitation was 200 copies/mL.

Statistical analysis
All statistical analyses were carried out with Statview 4.5 software (Abacus Concepts). Analysis of survival time according to the ART regimen was done using the Kaplan Meier product limit method. Survival curves were compared using log-rank tests. Non-parametric tests (Mann-Whitney test, Kruskall-Wallis test) were used to compare continuous and discrete variables. Crude relative hazards (RH) of death were computed for all of the following variables (sex, age, mode of HIV exposure, initial distribution of brain lesions, CD4 cell count and EDSS score at baseline, prior AIDS diagnosis, ART regimen before and after PML diagnosis) using the Cox proportional hazards model. Variables with crude RH of statistical significance less than 0.2 were included in a multivariate model. The Wilcoxon signed rank test was used to evaluate the change of EDSS score in survivors treated with CART.

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References

Albrecht H, Hoffmann C, Degen O, *et al* (1998). Highly active antiretroviral therapy significantly improves the prognosis of patients with HIV-associated progressive multifocal leukoencephalopathy. *AIDS* **12:** 1149–1154.

Andrei G, Snoeck R, Vandeputte M, de Clercq E (1997). Activities of various compounds against murine and primate polyomaviruses. *Antimicrob Agents Chemoth* **41:** 587–593.

Antinori A, Ammassari A, De Luca A, *et al* (1997). Diagnosis of AIDS-related focal brain lesions: a decision-making analysis based on clinical and neuroradiologic characteristics combination with polymerase chain reaction assays in CSF. *Neurology* **48:** 687–694.

Autran B, Garcelain G, Li TS, *et al* (1997). Positive effects of combined antiretroviral therapy on $CD4^+$ T cell homeostasis and function in advanced HIV disease. *Science* **277:** 112–116.

Baqi M, Kucharczyk W, Walmsley SI (1997). Regression of progressive multifocal leukoencephalopathy with highly active antiretroviral therapy. *AIDS* **11:** 1526–1527.

Berger JR, Kaszovitz B, Post JD, Dickinson G (1987). Progressive multifocal leukoencephalopathy associated with human immunodeficiency virus infection. A review of the literaure with a report of sixteen cases. *Ann Int Med* **107,** 78–87.

Berger JR, Concha M (1995). Progressive multifocal leukoencephalopathy: the evolution of a disease once considered rare. *J Neurovirol* **1:** 5–18.

Berger JR, Pall L, Lanska D, Whiteman M (1998a). Progressive multifocal leukoencephalopathy in patients with HIV infection. *J Neurovirol* **4:** 59–68.

Berger JR, Levy RM, Flomenhoft D, Dobbs M (1998b). Predictive factors for prolonged survival in acquired immunodeficiency syndrome-associated progressive multifocal leukoencephalopathy. *Ann Neurol* **44:** 341–349.

Brooks BR, Walker DL (1984). Progressive multifocal leukoencephalopathy. *Neurol Clinics* **2:** 299–313.

Brosgard C, Roberts T, Safrin S, Marra CM, Colson N, Jaffre HS (1997). Cidofovir therapy for progressive multifocal leukoencephalopathy in two AIDS patients (abstract). 37th Interscience Conference on Antimicrobial Agents and Chemotherapy, Toronto, Canada.

Cinque P, Scarpellini P, Vago L, Linde A, Lazzarin A (1997). Diagnosis of central nervous system complications in HIV-infected patients: cerebrospinal fluid analysis by polymerase chain reaction. *AIDS* **11:** 1–17.

Cinque P, Casari S, Bertelli D (1998). Progressive multifocal leukoencephalopathy, HIV and highly active antiretroviral therapy. *New Engl J Med* **339:** 838–849.

Clifford D and Neurologic AIDS Research Consortium (1998). Natural history of progressive multifocal leukoencephalopathy (PML) in AIDS modified by antiretroviral therapy. *J Neurovirol* **4:** 346.

Domingo P, Guardiola JM, Iranzo A, Margall N (1997). Remission of progressive multifocal leukoencephalopathy after antiretroviral therapy. *Lancet* **349:** 1554–1555.

Elliott B, Aromin I, Gold R, Flanigan T, Mileno M (1997). 2.5 year remission of AIDS-associated progressive multifocal leukoencephalopathy with combined antiretroviral therapy. *Lancet* **349:** 850.

Fong IW, Toma E and the Canadian PML study group (1995a). The natural history of progressive multifocal leukoencephalopathy. *Clin Infect Dis* **20:** 1305–1310.

Fong IW, Britton CV, Luinstra KK, Toma E, Mahony JB (1995b). Diagnostic value of detecting virus DNA in cerebrospinal fluid of patients with progressive multifocal leukoencephalopathy. *J Clin Microbiol* **33:** 484–486.

Gibson PE, Knowles WA, Hand JF, Brown DWG (1993). Detection of JC virus DNA in the cerebrospinal fluid of patients with progressive multifocal leukoencephalopathy. *J Med Virol* **39:** 278–281.

Gillepsie SM, Chang Y, Lemp G, *et al* (1991). Progressive multifocal leukoencephalopathy in persons infected with human immunodeficiency virus, San Francisco 1981–1989. *Ann Neurol* **30:** 597–604.

Hall CD, Dafni U, Simpson D and the AIDS Clinical Trial Group 343 Team (1998a). Failure of cytarabine in progressive multifocal leukoencephalopathy associated with human immunodeficiency virus infection. *N Eng J Med* **338:** 1345–1351.

Hall CD, Yiannoutsos C, Clifford DB (1998b). Progressive multifocal leukoencephalopathy, HIV and highly active antiretroviral therapy. Reply. *New Engl J Med* **339:** 848–849.

Holman RC, Janseen RS, Buehler JW, Zelasky MT, Hooper WC (1991). Epidemiology of progressive multifocal leukoencephalopathy in the United States: analysis of national mortality and AIDS surveillance. *Neurology* **41:** 1733–1736.

Hou J, Major EO (1998). The efficacy of nucleoside analogs against JC virus multiplication in a persistently infected human fetal brain cell line. *J Neurovirol* **4:** 451–456.

Huang SS, Skolasky GJ, Dal Pan, Royal W, McArthur JC (1998). Survival prolongation in HIV-associated progressive multifocal leukoencephalopathy treated with alpha-Interferon: an observational study. *J Neurovirol* **4:** 324–332.

Krupp LB, Lipton RB, Swerdlow ML, Leeds NE, Llena J (1985). Progressive multifocal leukoencephalopathy: clinical and radiographic features. *Ann Neurol* **17:** 344–349.

Kurtzke JF (1983). Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS). *Neurology* **33:** 1444–1452.

Major EO, Ault GS (1995). Progressive multifocal leukoencephalopathy: clinical and laboratory observations on a viral induced demyelinating disease in the immunodeficiency patient. *Curr Opin Neurol* **8:** 184–190.

Matheron S, Girard PM, Duval X, Petitdidier C, Bortolotti V, Gasnault J (1998). Evaluation of cidofovir for progressive multifocal leukoencephalopathy (abstract). 5th Conference on Retroviruses and Opportunistic Infection. Chicago, USA.

McGuire D, Barhite S, Hollander H, Miles M (1995). JC virus DNA in cerebrospinal fluid of human immunodeficiency virus-infected patients: predictive value for progressive multifocal leukoencephalopathy. *Ann Neurol* **37:** 395–399.

Miller J, Barrett R, Britton C (1982). Progressive multifocal leukoencephalopathy in a male homosexual with T-cell immune deficiency. *New Engl J Med* **307:** 1436–1438.

Moret H, Guichard M, Matheron S, *et al* (1993). Virological diagnosis of progressive multifocal leukoencephalopathy: detection of JC virus DNA in cerebrospinal fluid and brain tissue of AIDS patients. *J Clin Microbiol* **31:** 3310–3313.

Power C, Nath A, Aoki FY, Del Bigio M (1997). Remission of progressive multifocal leukoencephalopathy following splenectomy and antiretroviral therapy in a patient with HIV infection. *N Engl J Med* **336:** 661–662.

Sadler M, Chinn R, Healy J, Fisher M, Nelson MR, Gazzard BG (1998). New treatments for progressive multifocal leukoencephalopathy in HIV-1 infected patients. *AIDS* **12:** 533–535.

Taoufik Y, Froger D, Benoliel S, *et al* (1998a). Quantitative Elisa-polymerase chain reaction at saturation using homologous internal DNA standards and chemiluminescence revelation. *Eur Cytokine Network* **9:** 197–204.

Taoufik Y, Gasnault J, Karaterki A, *et al* (1998b). Prognostic value of JC Virus load in cerebrospinal fluid of patients with progressive multifocal leukoencephalopathy. *J Infect Dis* **178:** 1816–1820.

Von Einsiedel RW, Fife TD, Aksamit AJ, *et al* (1993). Progressive multifocal leukoencephalopathy in AIDS: a clinicopathological study and review of the literature. *J Neurol* **240:** 391–406.

Weber T, Turner RW, Frye S, *et al* (1994). Specific diagnosis of progressive multifocal leukoencephalopathy by polymerase chain reaction. *J Infect Dis* **169:** 1138–1141.

Weber T, Frye S, Bodemer M, Otto M, Lüke W (1996). Clinical implications of nucleic acid amplification methods for the diagnosis of viral infections of the nervous system. *J Neurovirol* **2:** 175–190.