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Toxoplasmic Encephalitis in an AIDS Cohort at Puerto Rico before and after Highly Active Antiretroviral Therapy (HAART)

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Abstract. Highly active antiretroviral therapy (HAART) significantly reduced the toxoplasmic encephalitis (TE) incidence in acquired immunodeficiency syndrome (AIDS) patients. The TE incidence and mortality were evaluated in an AIDS cohort followed in Puerto Rico before, during, and after HAART implementation in the Island. Of the 2,431 AIDS studied patients 10.9% had TE diagnosis, with an incidence density that decreased from 5.9/100 person-years to 1.1/100 person-years after HAART. Cox proportional hazard analysis showed substantial mortality reduction among TE cases who received HAART. No mortality reduction was seen in those cases who received TE prophylaxis. Although this study shows a TE incidence and mortality reduction in the AIDS cohort after HAART, the incidence was higher than those reported in the United States AIDS patients. Poor TE prophylaxis compliance might explain the lack of impact of this intervention. Strengthening the diagnostic and opportune TE diagnosis and prompt initiation of HAART in susceptible patients is important to control this opportunistic infection.

INTRODUCTION

Toxoplasmic encephalitis (TE) is one opportunistic infection of the central nervous system in human immunodeficiency virus (HIV)-infected patients. This acquired immunodeficiency syndrome (AIDS) defining condition is caused by an obligate intracellular protozoan parasite called *Toxoplasma gondii* (*T. gondii*) that has a worldwide distribution. The infection is usually transmitted by ingestion of undercooked pork or lamb meat, ingestion of contaminated vegetables, or direct contact with cat feces.[1–3] Prevalence of serologic evidence of *T. gondii* infection varies according to the geographic locale and population group at risk for the infection. Between 3% and 67% of adults in the United States (US) and up to 90% in Western Europe and tropical countries are positive for antibodies against *T. gondii.*[2] After the acute infection with *T. gondii* a cell-mediated immunity usually controls but does not eradicate the parasite that remains dormant in all organs, particularly in the central nervous system and in the myocardial, skeletal, and smooth muscles. This situation leads to the process of a chronic or latent infection. In patients with severe cell-mediated immune dysfunction, a reactivation of the infection produces the neurological manifestations of the infection.[1–3] Toxoplasmic encephalitis may produce a severe and often a life-threatening opportunistic infection that requires aggressive therapy and possibly lifelong prophylaxis for optimal outcome.

The risk of TE in HIV-infected persons decreased after the introduction of *T. gondii* prophylaxis and the use of effective antiretroviral therapy (ART).[4,5] Use of highly active antiretroviral therapy (HAART) leads to an improvement in the CD4 + T-cell count and a suppression of the HIV viral load. This improvement in the immune status of individuals reduces the reactivation of this infection.[4,5] The incidence of TE among US AIDS patients declined from 2.1/100 person years in 1992 to 0.7/100 person years in 1997 when HAART was implemented on a routine basis to most patients.[6]

Puerto Rico has a high prevalence of HIV infection and has the largest concentration of Hispanics within the United States and its territories.[7] Epidemiology of TE in this population has not been examined in more than a decade, since the introduction of HAART in the Island. A previous study reported a slight reduction of TE just after the implementation of HAART[8]; however, this study examines in more detail the trends in TE diagnosis and the variation in the morbidity and mortality of TE in a cohort of AIDS patients before and up to 10 years after the full implementation of HAART in the Island.

METHODS

Study population. This study determined the incidence of TE among AIDS persons followed by the Retrovirus Research Center (RRC) at Bayamon, Puerto Rico before, during, and after the full implementation of HAART. Demographic factors, clinical manifestations, laboratory findings, TE prophylaxis therapies, and ART prescriptions at the time of the TE diagnosis were tabulated and compared across three time periods. Period 1: 1992–1994, before HAART; Period 2: 1995–1997, introduction of managed HAART in Puerto Rico; and Period 3: 1998–2008, full implementation of HAART in the Island.

The study participants were followed at the Ramón Ruiz Arnau University Hospital or in the HIV ambulatory clinic of our institution until death or end of the study (September 2008). The study was conducted with the approval of the Institutional Review Board (IRB) of the Universidad Central del Caribe, at Bayamón, Puerto Rico.

Once an informed consent was obtained, an enrollment questionnaire was completed and laboratory testing was performed including CD4 + T-lymphocyte count and HIV viral load. Enrollment included a retrospective abstraction of clinical data and laboratory findings from the last 12 months before study entry. The HIV risk behaviors and socio-demographic data were gathered during a personal face to face 30 minutes interview. The most frequent AIDS-defining illnesses found

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in the study group were tabulated and included *Pneumocystis jirovecii* pneumonia (PJP), TE, recurrent bacterial pneumonia (RP), pulmonary tuberculosis (PTb), and Kaposi's sarcoma (KS). The duration of HIV infection was estimated on the basis of the initial positive HIV test reported until death, last clinical encounter, or the predefined study end (September of 2008). The TE incidence rate was estimated by dividing the TE cases reported in each study period by the total of the HIV-infected persons with at least one AIDS-defining condition and or a CD4 + T-cell count < 200 cells/μL, followed in the same time period. The TE incidence density was obtained with the TE cases and the sum of years followed up since the diagnosis of AIDS through the TE diagnosis, death, or study end. The TE prophylaxis was defined as the prescription of trimethoprim-sulfamethoxazole and/or the combination of pyrimethamine and dapsone before the diagnosis of the TE. Prescriptions of ART, including nucleoside reverse transcriptase inhibitors, non-nucleoside reverse transcriptase inhibitors, and protease inhibitors were collected and tabulated. The use of HAART was defined as the use of three or more antiretroviral drugs of two or more different classes before the diagnosis of TE. Additional clinical, laboratory, and therapy data were collected every 6 months after enrollment from the participants' interviews and medical records.

To evaluate the TE-related mortality, the study analyzed the mortality trends within the first year after the date of the TE diagnosis. Mortality data were obtained from a review of the institutional medical records and the Puerto Rican AIDS surveillance system. Data from the Puerto Rico Demographic Mortality Registry were also reviewed to establish date and causes of death as reported by the death certificate. Death causes were tabulated and organized into systems or organ failure.

Statistical analysis. The Statistical Package of Social Sciences (SPSS Inc., Chicago, IL) program was used to perform univariate, bivariate, and proportional hazard analyses. Differences between patients grouped by years of TE diagnosis, including incidence density, clinical manifestations, laboratory findings, therapy used, prophylaxis, mortality rates, and causes of death, were analyzed with the χ^2 or Fisher exact test, and the Student's *t* test. In addition, Cox proportional hazard analysis with relevant covariates, including variables with *P* values ≤ 0.05 in the bivariate analyses, were use to evaluate 1-year mortality risk. Data were presented as percentage, median with interquartile rate, and mortality hazard ratios (HR) with their 95% confidence interval (CI). The *P* value used to determine statistical significance was < 0.05.

RESULTS

General findings. A total of 2,431 AIDS patients were enrolled and followed in the RRC from 1992 through 2008. We identified 266 (10.9%) patients with TE at some point in their follow-up and the overall incidence density was 4.3/100 person-years. Of these TE cases, 74.4% were male, 61.3% had the injecting drug use (IDU) behavior as a main HIV transmission risk factor, and their mean age was 37.8 ± 9.3 years. Approximately 13.5% had a history of or had concurrently PJP, 13.5% wasting syndrome (WS), 3.4% RP, 3.1% Herpes simplex, 2.6% PTb, and 2.3% KS at TE diagnosis. A CD4 + T-cell count < 200 cells/μL was observed in 89.3%, 57.1% had received any TE prophylaxis before the diagnosis, 25.2% had received any ART, and 11.3% had been prescribed HAART by the time of the TE diagnosis.

Characteristics of TE patients by study periods. The incidence and the incidence density of TE decreased significantly in the Island from 14.1% and 5.9/100 person-years before the introduction of HAART to 4.3% and 1.1/100 person-years after the full implementation of this therapy in the year 1998 (Table 1). Those TE cases diagnosed in the HAART era were more likely to be older, have more frequent RP, and were more likely to have received ART and TE prophylaxis (Table 1). Conversely, no significant variation was found in the CD4 + T-cell count median at TE diagnosis between the study

Table 1
Incidences, demographic, clinical manifestations, and outcome of TE by study group

	1992–1994 (*N* = 110)	1995–1997 (*N* = 95)	1998–2008 (*N* = 61)
TE incidence (%)	14.1	9.2	4.3*
TE incidence density	5.9	2.8	1.1*
Male (%)	75.5	74.7	72.1
Age at TE years M (IQR)	33.7 (29.8–40.5)	37.5 (32.4–41.5)	38.8 (34.3–45.6)*
IDU antecedent (%)	60.0	64.2	59.0
Years with HIV, M (IQR)	2.0 (1.0–4.3)	3.1 (1.5–6.3)	2.3 (0.6–7.3)**
Years HIV-TE, M (IQR)	1.1 (0.2–2.7)	2.1 (0.5–4.6)	0.6 (0.4–3.3)*
CD4 + T cell < 200/μL (%)	87.1	87.0	89.3
HIV load/mL M (IQR)	–	305,788	143,350
Clinical antecedents at TE diagnosis (%)			
PJP	13.6	17.9	6.6
Recurrent pneumonia	2.7	1.1	8.2*
Kaposi's sarcoma	1.8	3.2	1.6
Esophageal candidiasis	11.8	24.2	14.8
Wasting syndrome	9.1	16.8	16.4
ART previous year (%)	14.5	27.5	41.0**
HAART previous year (%)	0	8.4	34.4**
TE prophylaxis (%)	49.1	62.1	63.9
One year mortality (%)	79.1	70.5	57.4**

* $P < 0.01$.
** $P < 0.05$. Incidence density × 100 person-years.
TE = toxoplasmic encephalitis; M (IQR) = median (interquartile rate); IDU = injecting drug use; HIV = human immunodeficiency virus; PJP = Pneumocystic jirovecii pneumonia; ART = antiretroviral therapy; HAART = highly active antiretroviral therapy.

TABLE 2
Demographic, HIV-related factors, and mortality causes of cases that died in the first year after TE diagnosis by study group

	1992–1994 N = 87	1995–1997 N = 67	1998–2008 N = 35
Male (%)	73.6	76.1	65.7
Age at death M (IQR)	34.4 (30.0–40.6)	38.3 (33.3–43.0)	39.4 (34.6–44.0)*
IDU antecedent (%)	56.3	64.2	60.0
Months AIDS-TE M (IQR)	0.0 (0.0–2.0)	0.0 (0.0–0.8)	0.2 (0.0–7.5)**
CD4 cell < 200/μL at TE (%)	90.5	90.0	90.9
TE prophylaxis (%)	46.0	65.7	68.5**
HAART (%)	0	4.5	28.6**
Death causes (%)			
Cardiovascular conditions	11.5	20.9	17.1
Pulmonary conditions	44.8	41.8	42.9
Gastrointestinal conditions	12.6	4.5	8.6
Metabolic conditions	6.9	10.4	8.6
Neurological conditions	17.2	14.9	11.4
Sepsis	6.9	16.4	25.7**
PJP	6.9	6.0	0
TE	57.5	53.7	48.6
KS	5.7	1.5	2.9

* $P < 0.01$ between years groups.
** $P < 0.05$ between years groups.
HIV = human immunodeficiency virus; TE = toxoplasmic encephalitis; M (IQR) = median interquartile rate; IDU = injecting drug use; AIDS = acquired immunodeficiency syndrome; HAART = highly active antiretroviral therapy; PJP = Pneumocystic jirovecii pneumonia; KS = Kaposi sarcoma.

periods. A high degree of immune dysfunction at the time of TE diagnosis was seen in all three study groups as expressed by the very low CD4 + T-cell count medians and the high percentages of cases with CD4 + T-cell count below 200.

Mortality findings. Among the 266 TE cases, 189 (71.0%) had died in the first year after the TE diagnosis, a significantly higher mortality was seen in patients diagnosed with TE before the implementation of HAART in the Island by the year 1998 (Table 1). In relation to the patients who died, those cases reported before 1998 were younger, had a shorter time between AIDS and TE, had less TE prophylaxis, and had less HAART at TE diagnosis time (Table 2). Death attributed to TE was more frequent in cases diagnosed before HAART than in those diagnosed afterward; post HAART death was most often attributed to sepsis (Table 2). Cox proportional hazard analysis confirmed that patients with a higher CD4 + T-cell count (HR = 0.62, 95% CI = 0.35–1.09), and those who used HAART (HR = 0.48, 95% CI = 0.17–0.88) at TE diagnosis had a lower mortality risk, after controlling for age, sex, estimated HIV disease duration, history of IDU, and TE prophylaxis (Table 3). Contrary, co-existence of other prevalent AIDS-related conditions at TE diagnosis were associated with a higher mortality, WS (HR = 2.17, 95% CI = 1.35–3.48), RP (HR = 2.53, 95% CI = 1.17–5.47), and KS (HR = 2.14, 95% CI = 0.90–5.08) after controlling by the same cofactor (Table 3).

DISCUSSION

The data show that the incidence of TE before 1998 in this Puerto Rican AIDS cohort was higher than one reported by previous studies in the United States for the same time period.[6] For example, Jones and others in their study of 12,982 AIDS persons in eight US states and Puerto Rico, reported a TE incidence three times lower than the one found in our AIDS group. After the implementation of HAART in the Island the incidence of TE decreased significantly; nevertheless, it remains higher than the one reported in the continental United States after the introduction of HAART. Unfortunately, no data has been reported in the US AIDS population on the TE incidence after 1997 so that a direct comparison is not possible. The study also indicates several changes in the clinical and socio/demographic characteristics of HIV-infected persons with TE, since the introduction of HAART, that is consistent with previously published studies.[1,5,9,10] In Puerto Rico the full implementation of HAART was present in 1998, and our study detected a significant decline of TE incidence and mortality afterward. Furthermore, we showed a substantial reduction in the 1-year mortality among those TE patients who received HAART (HR 0.48) and a beneficial trend in those with a higher CD4 + T-cell count (HR = 0.62) at the time of TE diagnosis. It is well accepted that HIV infection leads to a severe CD4 + T-cell count depletion with severe immunological damage that promotes the reactivation of *T. gondii*, which generates the target organ damage.[11] Therefore, the more competent the T-cells mediated cellular immunity, the better the immunological host capacity to maintain quiescence of the *T. gondii* chronic infection.[12] The use of ART and in particular HAART produce an effective HIV viral load reduction, with

TABLE 3
One year mortality risk after TE diagnosis by Cox proportional hazard*

	Mortality Hazard ratio	95% CI	*P*
Female	1.00	0.68–1.51	0.964
Age at study entry	1.01	0.99–1.03	0.279
IDU antecedent	1.09	0.75–1.56	0.654
Time from AIDS to TE	1.30	1.03–1.64	0.028
TE prophylaxis	0.91	0.94–1.30	0.613
PJP	1.21	0.74–2.09	0.417
Recurrent pneumonia	2.53	1.17–5.47	0.018
Wasting syndrome	2.17	1.35–3.48	< 0.01
Kaposi sarcoma	2.14	0.90–5.08	0.084
CD4 + T cell ≥ 200/μL	0.62	0.35–1.09	0.101
HAART	0.48	0.17–0.88	0.017

* TE = toxoplasmic encephalitis; CI = confidence interval; IDU = injecting drug use; AIDS = acquired immunodeficiency syndrome; PJP = *Pneumocystic jirovecii* pneumonia; HAART = highly active antiretroviral therapy.

a resultant improvement in the quantity and quality of the CD4 + T-cells, leading to an improved immune coordination and synergy that should decrease the magnitude and extent of target organ damage caused by *T. gondii*.

This study found a significant increment in the TE prophylaxis beginning in 1995 that concord with the time when this recommendation was implemented. However, this prophylaxis did not produce a significant protective effect in the mortality risk of the studied TE cases. Inadequate compliance with the TE prophylaxis therapies might explain this finding. Unfortunately, this dataset does not include information on this prophylaxis adherence.

Despite the important reduction in the incidence and improvement in the prognostic outlook of TE in these Puerto Rican AIDS persons with the use of ART and HAART, an important number of patients continue to suffer from this devastating infection. Strategies for the prevention of TE, prompt recognition, adequate prophylaxis and management of the infection, and the institution of adequate and prompt ART, continue to be of vitally important for controlling this infection. Consequently, primary prevention measures that minimize the risk for acquiring *T. gondii* should be advised and reinforced in Hispanic HIV-infected persons, such as this group; not eating raw or undercooked meat, including pork or venison, and washing hands after contact with raw meat, after gardening, or after having contact with soil need to be emphasized. In addition, fruits and vegetables should be washed before eating them raw. If patients own cats, their litter box should be changed daily preferable by other persons. These animals should not be given raw or undercooked foods. Furthermore, HIV-infected patients should be assessed for *T. gondii* antibodies at the time of HIV diagnosis and high risk populations for developing TE, including *T. gondii* seropositive persons with CD4 + T-cell count < 100 cell/μL should receive TE prophylaxis promptly and adequately.[13]

Limitations of our study include that the AIDS persons were selected from a passive surveillance cohort, in which patients had come to the hospital or outpatient facilities to have the data collected, otherwise the data would be lost. Thus, increasing the probability of lost to follow/up can lead to missing data that could affect the study findings. The study is not a population-based survey that could be generalized to all the AIDS patients on the Island.

In conclusion, the study showed a marked reduction of the TE incidence and mortality in this cohort of AIDS persons after the implementation of HAART in the Island. However, it continued to be higher than the one reported in the AIDS mainland population before HAART. An early diagnosis and referral for HIV treatment is highly recommended for this high-risk population. The immunological improvement produced by these opportune and adequate HIV therapies will reduce significantly the *T. gondii* reactivation. In addition, TE preventive and prophylaxis efforts directed to reduce the *T. gondii* infection and reactivation need to be emphasized and evaluated in further studies.

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