Journal of NeuroVirology, 14: 89–101, 2008

ISSN: 1355-0284 print / 1538-2443 online
DOI: 10.1080/13550280701829793

informa
healthcare

Review

Second assessment of NeuroAIDS in Africa

Kevin Robertson,[1] Kathy Kopnisky,[2] James Hakim,[3] Concepta Merry,[4] Noeline Nakasujja,[5] Colin Hall,[1] Moussa Traore,[6] Ned Sacktor,[7] David Clifford,[8] Charles Newton,[9] Annelies Van Rie,[10] Penny Holding,[11] Janice Clements,[12] Christine Zink,[12] Jens Mielke,[13] Mina Hosseinipour,[14] Umesh Lalloo,[15] Farida Amod,[16] Christina Marra,[8] Scott Evans,[17] and Jeff Liner,[1] on behalf of the second Assessment of NeuroAIDS in Africa Conference Participants

[1]*Department of Neurology, University of North Carolina at Chapel Hill School of Medicine, Chapel Hill, North Carolina, USA*
[2]*Center for Mental Health Research on AIDS, National Institutes of Health/National Institute of Mental Health, Bethesda, Maryland, USA*
[3]*Department of Medicine, University of Zimbabwe Medical School, Avondale, Harare, Zimbabwe*
[4]*Pharmacology and Therapeutics, Trinity Centre, S J H, Trinity College Dublin, Dublin, Ireland*
[5]*Department of Psychiatry, Makerere University Medical School, Kampala, Uganda*
[6]*Department of Neurology, University of Bamako Medical School, Point G Hospital Bamako, Mali*
[7]*Department of Neurology, Johns Hopkins University School of Medicine, Baltimore, Maryland, USA*
[8]*Departments of Neurology and Medicine, Washington University in St. Louis St. Louis, Missouri, USA*
[9]*Neurosciences Department, Institute of Child Health, London, United Kingdom; and Kenya Medical Research Institute, Kilifi, Kenya*
[10]*Department of Epidemiology, University of North Carolina at Chapel Hill School of Medicine, Chapel Hill, North Carolina, USA*
[11]*Centre for Geographic Medicine Research, Coast, Kemri, Kenya; and University of Oxford, UK*
[12]*Department of Molecular and Comparative Pathobiology, Johns Hopkins University School of Medicine, Baltimore, Maryland, USA*
[13]*Department of Medicine, College of Health Sciences, University of Zimbabwe Harare, Zimbabwe*
[14]*Department of Medicine, Division of Infectious Diseases, University of North Carolina–Chapel Hill, University of North Carolina Project, Lilongwe, Malawi*
[15]*Enhancing Care Initiative KZN, University of KwaZulu-Natal, Durban, KwaZulu Natal, South Africa*
[16]*Nelson R. Mandela School of Medicine, University of KwaZulu-Natal, Durban ACTG-ICTU, Durban, KwaZulu-Natal, South Africa*
[17]*Department of Biostatistics, Harvard School of Public Health, Boston, Massachusetts, USA*

In July of 2006, the National Institute of Mental Health (NIMH) Center for Mental Health Research on AIDS (CMHRA) sponsored the second conference on the Assessment of NeuroAIDS in Africa, which was held in Arusha, Tanzania. The conference mission was to address the regional variations in epidemiology of HIV-related neurological disorders as well as the assessment and diagnosis of these disorders. Participants discussed and presented data regarding the relevance and translation of neuroAIDS assessment measures developed in resource intensive settings and the challenges of neuro-assessment in Africa, including the applicability of current tools, higher prevalence of confounding diseases, and the complexity of diverse cultural settings. The conference presentations summarized here highlight the need for further research on neuroAIDS in Africa and methods for assessing HIV-related neurological disorders. *Journal of NeuroVirology* (2008) **14,** 89–101.

Keywords: HIV; neurological disease; neuropsychological assessment; resource-limited settings

Address correspondence to Kevin R. Robertson, PhD, 3114 Bioinformatics Building, CB7025, Department of Neurology, University of North Carolina at Chapel Hill School of Medicine, Chapel Hill, NC 27599-7025, USA. E-mail: kevinr@neurology.unc.edu
This article is based on the Second Assessment of NeuroAIDS in Africa Conference, Arusha, Tanzania July 17–19, 2006.
Received 27 August 2007; revised 23 October 2007; accepted 26 November 2007.

Introduction

In July of 2006, the National Institute of Mental Health (NIMH) Center for Mental Health Research on AIDS (CMHRA) sponsored the second conference on the Assessment of NeuroAIDS in Africa, which was held in Arusha, Tanzania. The conference mission was to address the regional variations in epidemiology of human immunodeficiency virus (HIV)-related neurological disorders as well as the assessment and diagnosis of these disorders. Participants discussed and presented data regarding the relevance and translation of neuroAIDS (neuro–acquired immunodeficiency syndrome) assessment measures developed in resource-intensive settings and the challenges of neuroassessment in Africa, including the applicability of current tools, higher prevalence of confounding diseases, and the complexity of diverse cultural settings.

The overwhelming majority of HIV infections exist in sub-Saharan Africa (SSA), the countries below the southern border of the Sahara Desert, so progress in this region was of particular interest. The first conference on the Assessment of NeuroAIDS in Africa featured researchers from the African countries of Malawi, South Africa, Uganda, Zambia, and Zimbabwe. Some of the countries specifically discussed at the second conference included the Democratic Republic of Congo, Ethiopia, Kenya, Malawi, Mali, South Africa, Uganda, and Zimbabwe. Since the first conference in 2004, more progress has been made in developing culturally and contextually relevant neuropsychological tests and the use of screening instruments for more efficient allocation of neuropsychological resources has become more commonplace. In regards to HIV treatment issues, the second conference focused less on problems with adherence and hepatotoxicities and more on problems with access to therapy and the toxic neuropathies related to antiretroviral treatment. Concerning the status of NeuroAIDS in Africa, the first conference highlighted a study on the prevalence of HIV-associated neurological disease in Uganda as well as theoretical issues of culture and setting and their impact on self reports and neurological tests. The presentations at the second conference focused more on the specific neurological features presented at sites in three countries, Mali, Uganda, and Ethiopia and stressed that differences in HIV clades represented across regions as well as differences in study design and population studied can all affect the neurological outcomes reported. The conference presentations summarized here highlight the need for further research on neuroAIDS in Africa and methods for assessing HIV-related neurological disorders.

Epidemiology of HIV in Africa

Dr. Hakim from Harare, Zimbabwe, presented an overview of the epidemiology of HIV in Africa .(Hakim, 2006). The situation remains critical. Africa accounted for 2.7 million new HIV infections and 2 million deaths due to HIV in 2005; of the estimated 39 million people living with HIV in 2005, 25 million or 64% lived in SSA. Of those infected 13.2 million (53%) were women and 2 million (8%) were children, representing 75% of women and 90% of children living with HIV worldwide. By 2005 HIV prevalence in adults in some countries in SSA was as high as 34%. However, trends in HIV infections are peaking and showing decline in some areas. Uganda, Kenya, Zimbabwe, and urban Burkina Faso are showing declines possibly due to increased condom use, delay in sexual debut, and a decrease in the number of casual sexual partners. The leveling off of prevalence may also be a result of high mortality rates. Long-distance truck drivers, commercial sex workers, blood transfusion recipients, and special groups such as migrant workers, miners, and armed forces are still the main sources of transmission. The ramifications of HIV/AIDS include overwhelmed health services, decreased health providers due to "burn out," migration and death, struggling economies with increased poverty, and a decreased workforce in agriculture and industry. Education has suffered greatly as a result of school drop outs and increased death rate among teachers. A comprehensive program focused on prevention and treatment will continue to be necessary to reduce infection rates and death rates in SSA, where condom use remains uncommon and where fewer than 10% of infected people in the majority of countries are receiving antiretroviral therapy (ART).

Antiretroviral rollout in Africa

Dr. Merry discussed her experiences with CARE, the Cohort programme to evaluate Access to anti-Retroviral therapy and Education, with study sites in Dakar, Senegal, Abidjan, Cote d'Ivoire, Nairobi, Kenya, and Kampala, Uganda .(Merry, 2006). At these four sites, the percentage of patients with virological success as well as the mean CD4 count both rose significantly over time with increased access to ART. In spite of the overwhelming success of ART in improving systemic illness as seen at these sites, limited resources on the personal, national, and international levels hinder progress towards treating all of those with HIV in need of therapy. Compared to developed countries, SSA and other resource-poor areas have unmet basic needs and cultural issues that place seemingly insurmountable barriers to pervasive treatment and prevention efforts. People neglect personal healthcare when faced with the daily struggle for food, water, and shelter, while governments emphasize security and infrastructure development over healthcare services. Cultural issues include social stigma and a deeper concern with the present over the future. Her message was that, in addition to the obvious medical and humanitarian reasons,

failure to treat HIV has a profound and growing effect on economic and social stability, and on national security. Although inroads are being made on this crisis, much remains to be done to enlist the full support of resource-poor governments in this battle.

Dr. Nakasujja presented her experience with HIV/AIDS and the use of highly active antiretroviral therapy (HAART) in Uganda .(Nakasujja, 2006). Even though HIV prevalence has been stabilized in SSA, the actual number of people infected continues to grow due to population growth. The infection rate in Uganda in 2005 was 7%, compared to 18% in 1992. Uganda has a very high level of political commitment and awareness to HIV/AIDS treatment at all levels including a strong and comprehensive health sector that has become a model for many countries. The World Health Organization (WHO) "3 by 5" initiative to treat 3 million people living with HIV/AIDS by 2005 targeted 60,000 Ugandans to be on ART by the end of 2005. Uganda has committed to doubling the WHO target and aims to have 120,000 of the HIV-infected population on ART by the end of 2007. Donor supported programs began in 2003, and thus far have treated approximately 57% of the target (around 69,000 people) at 185 accredited sites. Both generic and brand name antiretroviral drugs are offered at these sites but with a preference for the cheaper generics. Despite initial concerns that HIV treatment could divert both resources and attention away from prevention, treatment scale-up in Uganda has actually increased opportunities to undertake effective prevention. Increased awareness of HIV status and better access to treatment and care have created an upsurge in demand for HIV counseling and testing services. New approaches to testing and counseling include home-visit testing, family, couples' and discordant couples' therapy. Uganda has the theoretical capacity to rapidly scale up antiretroviral therapy if adequate financial resources are made available. However, significant problems remain. The costs of drugs and laboratory services remain high. A shortage of human resources, compounded by low salaries, lack of incentives, and the hold on hiring in the public sector, creates obstacles. The capacity for scaling up is weak at the district and subdistrict levels, and the referral systems are inadequate. Facilities for data management are inadequate and there is a lack of coordination between the multiple HIV care providers and stakeholders. Infrastructural problems such as inadequate roads and incorrect addresses, false beliefs, and quack healthcare givers all complicate care giving. Although less marked than in the past, the stigma associated with the diagnosis remains a problem for many Ugandans.

HIV-associated dementia in the HAART era

Dr. Hall reviewed the clinical features and trends in HIV-associated dementia (HAD) during the HAART era (Hall, 2006). HAART has completely changed the clinical picture of HAD. Although there has not yet been a reduction in the prevalence of HAD or its less severe phenotype minor cognitive motor disorder (MCMD), multiple studies show improvement, sometimes dramatic, in neuropsychological function post-HAART in adults and children (Cysique *et al*, 2004; Ferrando *et al*, 2003; Robertson *et al*, 2004; Saavedra-Lozano *et al*, 2006; Sacktor *et al*, 2000, 2006; Shanbhag *et al*, 2005; Tozzi *et al*, 1999, 2001). Improvement usually continues for at least 3 months, but some studies show improvement maintained for up to 2 years after treatment begins when treatment is maintained throughout the 2 years and the patient is adherent. However, there are still significant deficits in treated populations, probably a result of pretreatment neurological damage. Progressive deficits have been reported in some treated subjects, but the relevance of this is not yet clear as serial testing has demonstrated changes in both positive and negative directions.

Most patients on HAART have dramatic improvements in systemic disease markers like CD4 count and plasma viral load, but the picture is less clear for the central nervous system (CNS). Most studies prior to HAART showed some correlation between dementia and CD4 level, plasma viral load, or cerebrospinal fluid (CSF) viral load (Childs *et al*, 1999; Ellis *et al*, 1997; McArthur *et al*, 1997; Robertson *et al*, 1998). However, in the HAART era, CSF virological failures are still fairly common and plasma viral load, CSF viral load, and CSF immune markers are not correlated with or predictive of dementia (Cysique *et al*, 2006; Sevigny *et al*, 2004; Stankoff *et al*, 1999; Tozzi *et al*, 2007).

The poor penetration of antiretroviral drugs (ARVs) across the blood-brain barrier allows the potential for viral sequestration in the brain and could potentially cause continuing neurological decline. These viral reservoirs may also increase the potential for drug resistance and reseed the systemic compartment(Antinori *et al*, 2003; Boffito *et al*, 2006; Cashion *et al*, 1999; Clements *et al*, 2005; Enting *et al*, 1998; Kramer-Hammerle *et al*, 2005; Langford *et al*, 2003; Wynn *et al*, 2002). Some studies have reported a correlation between CSF drug penetrance and neuropsychological test scores but others have not (Eggers *et al*, 2003; Robertson *et al*, 2004). Although these studies show mixed results, it is probably reasonable to add ARVs with higher CSF penetrance into treatment regimens of neurologically impaired patients.

The optimal time to start HAART has not been established, but several lines of evidence suggest that, as it relates to the nervous system, the earlier the better. In both developed and developing nations, early treatment must be weighed against cost, treatment side effects and the likelihood of resistance.

NeuroAIDS in Africa

Most of the research on the neurological outcomes of HAART has been limited to studies carried out

in developing countries with adequate access to primary HIV treatment. In SSA, an area where people are of a much lower socioeconomic status, antiretroviral drugs are expensive and hard to come by, and cultures are vastly different from the United States and Western Europe, the effects of HAART on HIV-associated neurological disease are still being elucidated. Experiences from three countries in SSA, Mali, Uganda, and Ethiopia are described below.

The overall prevalence of HIV infection in Mali is 1.8% (Traore, 2006). ARVs have been available in three clinics in the capital Bamako since 2001, and five further centers have recently been established. Mali faces the same logistical and financial problems as other resource-poor countries. Computed tomographic (CT) scanning is available, but magnetic resonance imaging (MRI) is not, CD4 evaluation is possible but not universal, and there is limited ability for further diagnostic testing. Dr. Traore discussed the spectrum of neurological diseases associated with HIV infection at a clinic in Mali. From January 1991 to December 2002, 5128 patients were admitted to the clinical neurology department and 239 (4.66%) were infected with HIV. Of the 239 patients with HIV, 110 had focal or diffuse brain disorder, 55 had cranial lesions, 41 had spinal disorders, and 33 had peripheral nerve disorders. The outcome of this study noted the frequency and diversity of neurological disorders associated with HIV infection. In this Mali clinic, where HIV subtypes G and CRF06_CPX are common, CNS complications were more frequent than peripheral nervous system complications.

Dr. Sacktor presented a study of HIV-associated cognitive impairment at an infectious disease clinic in Kampala, Uganda where clades A and D are the predominant subtypes (Sacktor, 2006). Of 78 ambulatory HIV+ patients studied, 31% met criteria for HIV dementia, 47% for mild cognitive impairment, and 22% of the patients had no impairment on neuropsychological testing. These figures are similar to the pattern found in patients with advanced immunosuppression in the pre-HAART Dana cohort study in the United States. Neuropsychological tests of verbal memory (WHO-UCLA Verbal Learning Test trial 5 and delayed recall) and executive function (Color Trails Test parts 1 and 2) were most likely to demonstrate impairment. In a pilot study, 23 HIV+ individuals had detailed neuropsychological testing assessments at baseline and after 6 months of HAART therapy. At baseline, 61% of the HIV+ individuals were diagnosed with HIV dementia. At the 6-month follow-up, the mean CD4 count improved from 71 cells/μl at baseline to 222 cells/μl ($P < .001$) and only 4% were diagnosed with HIV dementia. There was also improvement in functional performance as measured by the Karnofsky scale. These results suggest that HAART should be provided for HIV+ patients in SSA with neurocognitive impairment (regardless of CD4 status) if confounding factors such as CNS opportunistic infections, delirium from an acute systemic process, or preexisting conditions have been ruled out as a cause for cognitive impairment.

Dr. Clifford discussed results of the Ethiopian Netherlands AIDS Research Project (ENARP), a longitudinal natural history study of HIV in two communities in rural Ethiopia where general healthcare is available (Clifford, 2006). HIV assessments were performed at each of the communities in this cohort as a part of the Ethiopian Neurologic AIDS Research Consortium. (ENARC) (Clifford *et al*, 2007). Both HIV positives and controls came from the same community, and all HIV positives available were studied. The investigators also evaluated a new rapid screening measure for dementia called the International HIV Dementia Scale (IHDS). Patients were given four tasks: naming four objects, fingertapping, the "Luria" psychomotor learning task, and a delayed recall of the four objects previously named. The IHDS revealed no significant differences between the control population and the HIV-affected population. However, the traditional testing, largely using motor speeded tasks, did reveal significant slowing of finger tapping in the HIV-affected group, whereas all other tests showed no difference. The investigators detected less HIV-associated disability than was anticipated for this study. Performance of both controls and HIV positives were impaired by Western standards, but did not significantly differ from each other. The IHDS did not demonstrate differences between HIV positives and negatives consistent with clinical impression. Neuropathy was found in approximately 15% of both positive and negative populations.

Compared to the Ethiopian project, the untreated population from the Ugandan clinic showed more cognitive impairment, as well as more advanced CD4/Karnofsky status. However, population differences as well as study design could explain disparities in the results. Researchers emphasized that norms for tests must be developed locally and require an appropriate normal population. Examination conditions and examiners should be the same for HIV+ and control populations. Demographic influences may be meaningful and unrecognized; thus, populations should be well matched. It is also possible that the genetic diversity of different viral subtypes could play a part in the disparity seen in different populations (Liner *et al*, 2007; Sacktor *et al*, 2007). Although this has been inadequately studied, there have been several recent reports that HIV subtype D is associated with faster disease progression, specifically over subtype A (Kaleebu *et al*, 2002, 2007; Kanki *et al*, 1999; Vasan *et al*, 2006). With subtype C predominant in Ethiopia and subtype D predominant in Uganda, regional differences in disease progression could be a result of the genetic diversity of different viral subtypes. Unfortunately, the relative neurovirulence of different subtypes is not well known.

Pediatric neuroAIDS in Africa

HIV-related pediatric CNS disorders usually exhibit broad variability in severity and timing (Epstein *et al*, 1986; Lobato *et al*, 1995). CNS involvement is the first AIDS-defining symptom in as many as 18% of pediatric patients (Gabuzda and Hirsch, 1987). It can occur before significant immunosuppression and generally presents as HIV encephalopathy with developmental delay or loss of developmental milestones, microcephaly, and pyramidal tract signs (Belman *et al*, 1988). Risk factors for pediatric neuroAIDS include the timing of infection, advanced maternal disease at delivery, rapid progression with early advanced immune suppression, high plasma viral load in infancy, and factors such as home environment and socioeconomic status.

Over 95% of children in SSA acquire their infections from their mothers. The cumulative rates of HIV transmission from the mother are 25% to 40%. Under 10% are infected during pregnancy, 10% to 20% at birth, and 10% to 20% during breast feeding (Dabis and Ekpini, 2002). This results in more than 2.2 million children infected with HIV in SSA, of whom approximately 35% die by 1 year and 52% by 2 years of age.

Dr. Newton and colleagues are studying pediatric HIV encephalopathy in Kilifi, the second poorest area in Kenya, where as many as 20% of children are underweight with stunted growth, and 10% have microcephaly .(Newton, 2006). There was a high prevalence of neurodevelopmental impairment in the community. However, because over half of children infected with HIV die before their second birthday, its effect on neurological involvement is difficult to detect. In this age group, motor function was the easiest to assess. Lack of culturally appropriate cognitive and language tests make information on these domains difficult to interpret. They have developed a simplified definition of HIV encephalopathy for children in SSA, which consists of (1) any child who is HIV positive; (2) lack of growth in head circumference assessed by serial measurements at least 3 months apart; (3) neurodevelopmental milestones such as loss of skills (particularly motor) and lack of acquisition of skills; (4) diffuse symmetrical hyperreflexia; and (5) lumbar puncture to exclude CNS infections.

Dr. Newton discussed the need for a study of HIV-positive children in SSA with "encephalopathy" to identify CNS infections, obtain neuroimaging, and investigate possible metabolic causes that may influence neurodevelopment of undernourished children in poorer communities. Further research is also needed to identify neurodevelopmental skills that can be assessed in children across SSA in the age range of 6 to 48 months. Culturally and language appropriate assessments of neurodevelopment also need to be developed and standardized across large populations.

Dr. Van Rie also stressed the lack of neurodevelopmental assessment tools evaluated and validated outside of the Unites States and Europe ..(Van Rie, 2006). In addition, disentangling the direct effect of HIV on the CNS from the environmental and social influences of HIV on the neurodevelopment of HIV-infected children in the sub-Saharan African context poses significant problems to assessment. In a pilot study in Kinshasa, Democratic Republic of Congo (DRC), three groups of children age 18 to 71 months were evaluated, including 35 HIV-infected children initiating HAART, 35 HIV-affected but uninfected children (AIDS orphans and children of parents with symptomatic AIDS), and 90 control (HIV-unexposed healthy) children living with healthy parents from Kinshasa. Neurodevelopmental assessment, maternal quality of life, demographic parameters, family structure, and clinical and immunological parameters were collected at baseline, 6 and 12 months. Neurodevelopmental assessment tools included the Bayley motor scale, Bayley mental scale, Peabody motor scale, and Snijders-Oomen Nonverbal Intelligence Test (SON $2\frac{1}{2}$–7). Although the mean and standard deviation (SD) of the motor scores for the control group was not significantly different from the mean and SD of the normative populations (determined in settings of Western culture such as the United States and Europe), lower mean mental scores were observed. This may indicate the need to adapt some of the items to the sub-Saharan African context.

Compared to the normative group, motor, mental, and language development were delayed in an overwhelming majority of HIV-infected children. HIV-affected children had significant but less marked delays in their motor development and tended to have slower mental development suggesting both a direct biological (HIV) and an environmental component. Behavioral problems were identified in both HIV-infected and -affected children. Language expression was more delayed than comprehension in both the HIV-infected and -affected groups. The impact of CNS involvement appeared to be more pronounced in the younger age group of HIV-infected children. However, this could be due to the use of different tools in different age groups, or the effect of a "survival cohort." Confounders also included malnutrition and socioeconomic status. The follow-up assessment indicated that both mental development and motor development improved in a greater proportion of HIV-infected children compared to control children, possibly due to the effect of ART. Over time, the control group showed greater improvement of mental development compared to motor development scores, revealing a possible learning effect.

Dr. Holding presented on pediatric neuroassessment in Kenya, emphasizing the need for the development of tools appropriate to the context, to ensure that they are sensitive to potential subtle effects of disease (Holding, 2006). She described the

development of just such a battery of measures for infants and toddlers that has identified poorer development in psychomotor and language skills in children growing up in East Africa exposed to HIV infection in utero. Differences from unexposed children were found to increase with age, and to extend to emotional regulation. Dr. Holding also emphasized the need to research the neurodevelopment of children with maternal HIV in the context of coinfections such as malaria. Maternal malaria had a higher prevalence in HIV+ mothers (41%) compared to HIV− mothers (21%). Adverse birth outcomes are more common in the presence of both HIV and malaria. The developmental outcome in children born to HIV and malaria co-infected women may therefore be mediated both directly by maternal HIV transmission and indirectly by mechanisms involving complications of coinfection.

Peripheral nerve disease in HIV-infected subjects

Although there are no recent epidemiological data from Africa, Dr. Mielke from Harare, Zimbabwe, discussed peripheral nerve diseases in HIV-infected subjects, with a focus on their classification into several major clinical neuropathies based on when they occur with respect to CD4 count or disease stage, as well as clinical course and major symptoms (Mielke, 2006). Several recent studies have found neuropathy in over 60% of subjects (Moore *et al*, 2000; Morgello *et al*, 2004; Schifitto *et al*, 2002; Simpson *et al*, 2006).

Distal sensory polyneuropathy (DSP), a "dying back" axonal neuropathy progressing distally from the toes and featuring distal pain, paresthesia, and numbness, is the most common HIV-associated neuropathy. DSP usually occurs during advanced HIV infection with a subacute and chronic course. Toxic neuropathies from the "d-drugs" zalcitabine (ddC), stavudine (d4T), and didanosine (ddI) share clinical features with DSP, making them hard to differentiate, but typically have a more acute onset, 1 week to 6 months, and more prominent pain than DSP. Symptomatic improvement can usually be seen over weeks to months after discontinuation of treatment, but is often preceded by an initial period of worsening symptoms. However, because DSP and toxic neuropathies are often indistinguishable and overlapping, about one third of patients fail to improve after treatment is stopped. Elevated serum lactate levels and mitochondrial DNA have been used to help distinguish "d-drug"neuropathy. A small study has demonstrated that use of l-acetyl carnitine improved toxic neuropathy symptoms, supporting mitochondrial toxicity as the mechanism for nucleoside reverse transcriptase inhibitor (NRTI)-induced neuropathy (Herzmann *et al*, 2005; Osio *et al*, 2006).

Dr. Mielke's discussion also included a review of the symptoms, time course, and diagnosis of several other HIV-associated neuropathies, including acute inflammatory demyelinating polyneuropathy (AIDP), chronic inflammatory demyelinating polyneuropathy (CIDP), mononeuropathy multiplex and progressive polyradiculopathy.

Lactic acidosis and ascending neuromuscular syndrome

Dr. Hosseinipour presented initial data on ascending neuromuscular syndrome in Lilongwe, Malawi (Hosseinipour, 2006). As ART is rapidly scaling up in resource-poor countries, the primary combination used in many countries is stavudine/lamivudine (d4T/3TC) and nevirapine (NVP) since it is inexpensive and available in fixed dose combinations. A potential toxicity related to long-term use of this combination is lactic acidosis (LA), a potentially fatal condition that requires cessation of ART. LA is commonly associated with an ascending neuromuscular weakness and rapid-onset neuropathy with/without muscle weakness may indicate lactic acidosis. Risk factors for LA include female sex, duration of therapy, pregnancy, preexisting liver disease, and obesity (Bonnet *et al*, 2003; Calza *et al*, 2005; Coghlan *et al*, 2001; Imhof *et al*, 2005; Moyle *et al*, 2002). Drug associated risks for LA are d4T/ddI > d4T > ddI = zidovudine (ZDV) > 3TC = abacavir (ABC). LA is not yet seen in Tenofovir (TDF) treatment. In 2002, the FDA reported 25 cases of LA with neuromuscular toxicity .(FDA, 2002). Twenty-two of the cases were on stavudine containing regimens, 8 were pregnant women, and there were 7 deaths. The FDA case definition included neuromuscular weakness, lactic acidosis or symptomatic hyperlactatemia, and events occurring within 4 to 5 weeks of each other. Clinical symptoms of several known LA cases in Lilongwe included drastic weight loss, nausea, abdominal distension, and neuropathic pain followed by progressive muscle weakness. Higher lactate levels corresponded with higher degrees of pain and weakness and prognosis of death. Of the 20 known cases of LA identified in Lilongwe, 100% were on stavudine.

This ascending neuromuscular syndrome may be due to mitochondrial toxicity, and progressive neurological dysfunction can persist and worsen after cessation of ART when associated with lactic acidosis. Better characterization of the rate of this syndrome is required as the population at risk is great and increasing. Standardized, clinical assessments, and evaluation among prospectively identified cases is required.

Recent developments in the SIV macaque model of HIV CNS disease

Dr. Clements described results of research from the Retrovirus Lab at Johns Hopkins on an accelerated, consistent simian immunodeficiency virus

(SIV)/macaque model where the asymptomatic phase was virtually eliminated and the macaques progressed to the terminal stage within 84 days (Clements, 2006). The SIV model examined virus genotypes throughout infection and found specific genotypes in the brain during the acute and asymptomatic stages of infection. There was evidence in the SIV model for reseeding of the blood with various viral genotypes from the brain. Correlates of neurological disease included viral entry into the CNS during acute infection, infection and activation of macrophages and astrocytes, viral RNA in the brain and viral load in the CSF, up-regulation of CCL2 in the CSF, and finally CNS inflammation with immunosuppression. All contribute to neuronal loss and degeneration in the CNS and peripheral nervous system (PNS). Dr. Clements also discussed the therapeutic effects of interferon-β (IFNβ) on HIV infection. During the innate immune response in the CNS, rapid production of IFNβ induces the production of anti-inflammatory cytokines. This slows virus spread and suppresses replication at the transcriptional level before the induction of virus specific immune responses. The research indicates that IFNβ inhibits SIV. Based on the predictable SIV model, the studies suggest that IFNβ or a therapeutic that would prolong the innate immune control might maintain innate immune responses and lengthen the asymptomatic phases in the CNS.

Dr. Zink reported on the potential neuroprotective benefits of minocycline in conjunction with HIV treatment (Zink, 2006). Minocyline has been shown to be neuroprotective in animal models of multiple sclerosis, stroke, brain trauma, Parkinson's disease, amyotrophic lateral sclerosis (ALS), and Huntington's disease. It has been on the market for 30 years, so it is off patent, has proven safe when used daily for years, and has good CNS penetrance. In this SIV study, minocycline reduced macrophage activation measured by increased expression of CD68 and major histocompatibility complex (MHC) class II and in turn reduced the severity of SIV encephalitis. Minocycline appears to possess both anti-inflammatory actions by drastically reducing CCL2 (monocyte chemoattractant protein [MCP]-1) expression, and antiviral effects as evidenced by suppressing SIV/HIV viral replication in lymphocytes and macrophages. Interestingly, virus cultured in the presence of minocycline did not develop resistance and plasma virus from SIV-infected, minocycline macaques was not resistant to later suppression by minocycline. Minocycline also presents therapeutic PNS effects, especially for those suffering from toxic neuropathies. It was shown to prevent dorsal root ganglion loss in terms of neuronal area fraction to almost to the same level as controls, and minocycline combined with tenofovir was shown to prevent epidermal nerve fiber loss to a level intermediate between SIV uninfected and infected.

These recent findings based on the SIV model may provide important alternate and adjunctive therapies for HIV-associated neurological problems. In SSA, where access to primary treatment is still limited and many of the more toxic drugs are still relied on, alternate therapies such as minocycline and IFNβ may be vital to treat the inflammatory aspects of HAD and toxic neuropathies and to strengthen innate immune responses and lengthen the asymptomatic phases in the CNS.

HIV-related neurological disease in Africa—discussions of CNS opportunistic infections

Central nervous system tuberculosis

Dr. Lalloo, from Durban, South Africa, reviewed central nervous system tuberculosis (CNS TB) in HIV-infected patients (Lalloo, 2006). TB is the second most common cause of death from a single infectious agent and the most common opportunistic infection associated with HIV. Approximately one third of the world's population is infected with TB, with about 8 million new cases and 3 to 4 million deaths occurring each year. The risk of TB infection increases after HIV seroconversion; in SSA where nearly two thirds of the world's HIV-infected individuals reside, the two problems are inextricably connected. SSA accounts for about one third of new TB cases each year and both the incidence and prevalence continue to rise. Around 10 million people are coinfected with HIV and TB, and some researchers estimate that TB kills 1 out of every 3 AIDS patients. TB is one of the factors that result in neurologic signs and symptoms in 70% to 90% of HIV patients during the course of the disease. Clinical syndromes associated with neuro-TB include meningitis, tuberculoma, brain abscess, encephalitis, myeloradiculopathy, spinal cord abscess, and vertebral TB. CNS TB is more common in select populations such as intravenous drug users, the poor and marginalized, and persons with advanced HIV disease (CD4 <200). However, there is no evidence yet that the neurological manifestations of TB are different in HIV+ versus HIV− patients.

Multidrug-resistant TB has a mortality rate of $>60\%$ and is an increasing problem as more and more people have a poor response to second line treatment. It must be expected in any patient with prior exposure to TB drugs and meningitis. There is currently no data to support any standard strategy combining HAART and TB treatment. Treatment must be individualized to each patient, but experience with pulmonary TB (PTB) suggests that concomitant treatment is beneficial. Some studies have suggested a decrease in the manifestations of neuro-TB in patients on HAART (Bottieau *et al*, 2003; Neuenburg *et al*, 2002); however, as a likely manifestation of immune reconstruction immunodeficiency syndrome (IRIS), some HAART-treated patients suffer a paradoxical

worsening of TB while on appropriate TB treatment (Aaron *et al*, 2004; Breen *et al*, 2006; Dean *et al*, 2002). This paradoxical worsening is thought to represent the host's reconstituted immune response to the TB antigen, but treatment failure, poor adherence, malabsorption, adverse drug reactions, lymphoma, and drug resistance must be excluded. Under these conditions, it has been suggested that HAART should be delayed until TB is under control, but more studies are needed to determine optimal treatment.

AIDS-associated toxoplasmosis

Dr. Amod from Durban, South Africa, discussed toxoplasmosis, the commonest opportunistic infection (OI) causing focal brain lesions in AIDS patients (Amod, 2006). Worldwide seropositive prevalence rates vary geographically from 20% to 75% and are notably higher in Europe than in the United States. The estimated toxoplasmosis seroprevalence in South Africa is around 34%. Many African countries hit hard by HIV/AIDS also have a high seroprevalence of toxoplasmosis. Untreated toxoplasmosis infections can result in debilitating neurological disease and are often fatal in AIDS patients. Incidence of toxoplasmosis encephalitis (TE) correlates with the prevalence of immunoglobulin G (IgG) antibodies and 95% of cases are due to a reactivation of latent disease during advanced immunosuppression. CT or MRI usually shows multiple focal brain lesions. If lesions are detected, and the patient is positive for *Toxoplasma* IgG antibodies, then antitoxoplasma therapy is started. Toxoplasmosis usually targets the CNS in 80% of cases and the retina in 5% to 10% of cases. Pneumonitis, myocarditis, and disseminated multiorgan involvement are far less common. TE has a subacute onset, with neurological and constitutional symptoms progressing over days to weeks. Clinical features include fever and headache in 40-% to 70% of cases, focal neurological signs (hemiparesis and cranial nerve palsies) in 50% to 60%, seizures in 30% to 40%, and diffuse neurological dysfunction such as confusion and lethargy in 40%. Primary prophylaxis has been shown to prevent the reactivation of disease and should be instituted in resource-limited settings once CD4 is $<200/\mu$l. In resource-limited settings, the recommended treatment is cotrimoxazole because it is inexpensive, readily available, and effective. After treatment, neurological response occurs by day 3 in 50% and by day 14 in 90% of patients. Radiological improvement usually appears by the third week of treatment. In resource-limited settings where pharmaceuticals and testing supplies must be used efficiently, healthcare providers have limited access to powerful diagnostic instruments such as CT and MRI, and many patients are diagnosed with HIV only after developing OI such as toxoplasmosis, the subacute nature of TE poses a significant problem to successful and timely diagnosis and treatment.

HAART has been associated with a decline in the incidence of Ois, including toxoplasmosis. Observational and randomized studies show that for primary prophylaxis (no previous episode of toxoplasmosis), therapy can be discontinued when CD4 >200 for ≥ 3 months. More limited data is available regarding stopping secondary prophylaxis, but it should be considered when CD4 count >200 for >6 months and patient is asymptomatic.

Neurosyphilis

Dr Marra discussed neurosyphilis (Marra, 2006). The WHO estimates that, worldwide, 12 million people acquire syphilis each year. Most cases occur in parts of the world where HIV is highly prevalent, and 10% to 50% of patients with syphilis also have HIV. This translates to 180,000 to 1.2 million cases of symptomatic neurosyphilis every year. Neurosyphilis can occur at any time after infection, even in patients who have primary syphilis. The gold standard test to establish a diagnosis of neurosyphilis is a reactive cerebrospinal fluid (CSF)–Venereal Disease Research Test (VDRL). Unfortunately, this test is nonreactive in 30% to 70% of cases of neurosyphilis. Also, it is not available in many areas of the world where syphilis and thus neurosyphilis are most prevalent. Dr. Marra and colleagues have examined the use of immunochromatographic tests (ICTs) for neurosyphilis diagnosis. These commercial tests are rapid and easy to perform and have high sensitivity and specificity for syphilis diagnosis when used on whole blood or serum. In preliminary studies, they optimized three ICTs for diagnosis of neurosyphilis and showed that, using an undiluted CSF sample, it may be possible to use these tests to exclude the diagnosis of neurosyphilis. Conversely, using a dilution of the sample may enable the tests to be used to establish the diagnosis of neurosyphilis with a high degree of certainty. Larger studies are needed to confirm these preliminary findings.

After years of control, syphilis is resurging in the developed world and many cases are coinfected with HIV. In SSA and other developing regions of the world where the risk for HIV infection is high, there is also a high risk of syphilis and other sexually transmitted diseases. In the case of concurrent syphilis and HIV infection, the two diseases can synergize in their efforts to survive as well as in attacking the nervous system. Syphilis increases the risk of mother to child transmission of HIV and HIV-induced immunity impairs the clearance of *Treponema pallidum*, the spirochete bacteria that cause syphilis (and neurosyphilis) from the CNS. Neurorelapse, or the development of neurosyphilis after treatment for early syphilis, may be more common in HIV-infected individuals. Current WHO guidelines suggest prescribing ART in Africa and resource-limited settings when the peripheral blood CD4 T-cell count drops below 200 cells/μl (or when symptoms of OI manifest). However, HIV-infected persons with syphilis are at

increased risk of developing neurosyphilis if their CD4 T-cell counts are less than or equal to 350 cells/μl. Syphilis is a treatable, even curable, disease; however, diagnosis remains a problem. In these settings where resources are scarce and primary HIV treatment usually does not begin until a later stage of immunodeficiency, effectively diagnosing and treating opportunistic infections is very important. Dr. Marra and colleagues are making progress in this area by adapting immunochromatographic tests for cheaper, easier, and more effective diagnosis of neurosyphilis.

Neurological studies in resource-limited settings: lessons from ACTG5199

Based on experiences with ACTG5199: The International Neurological Study, Dr. Evans addressed several design and statistical issues that arise when planning and conducting neuro-AIDS clinical trials in Africa (Evans, 2006). First, evidence presented at the meeting based on data from Uganda, Ethiopia, Malawi, Zimbabwe, and South Africa suggests that there is considerable site diversity of neurological responses. For example, Clifford reports that the International HIV Dementia Scale, a brief screening tool, did not detect differences in cognitive status between HIV-positive and -negative subjects in Ethiopia consistent with clinical impression; contrastingly, Sacktor reports that in Uganda 31% of patients met criteria for HIV dementia and 47% of patients met criteria for mild cognitive impairment after extensive neuropsychological testing. Baseline results of neuropsychological tests from ACTG 5199 also suggest considerable site variation in neuropsychological function. Possible reasons for these differences include site differences in viral genetics and associated neurotoxicity, the distribution of HIV-associated complications and opportunistic infections, cultural or socioeconomic factors, or a variation in test administration (lack of standardization and training). Generally, increased variation results in a loss of precision and power, and decreases the ability to detect treatment effects. As a result, stratified or subgroup analyses may be required when conducting such multinational studies. Researchers should try to minimize variation by using standardized methods and definitions, objective and simple end points, central scoring and reading of data whenever possible, and design options such as matching or cross-over studies. HIV clinical trials in Africa tend to recruit subjects quickly. However, appropriate infrastructure is important to retain patients and ensure protocol compliance. Study drop-out and noncompliance can threaten the integrity of study results.

Neuroassessment issues

Because assessment of HIV neurology is less well studied in Africa than the United States, assumptions made in the design phases of studies are questionable when using data from US trials to design African trials. Studies may require interim assessment of these assumptions for validity. Considering the number of biological, demographic, and socioeconomic variables, adaptive designs such as "internal pilot studies" are necessary to keep studies on target and relevant to the context. For example, sample size calculations may be conducted utilizing estimates of variability or response rates obtained from studies conducted in the United States. These assumptions may not be valid in international settings, and thus sample-size reestimation at an interim time point may be prudent.

Another consideration is that clinical trial subjects may not be representative of the general population. Whereas sites in the United States have difficulty identifying eligible HIV patients due in large part to a low HIV prevalence (less than 3% of global HIV infections), African sites have relatively large pools of potentially eligible subjects to consider against inclusion/exclusion criteria. Site investigators in Africa may select patients for enrollment into longer duration trials who have more favorable clinical prognoses. This can result in a selection bias for a healthier patient group that can affect the ability to generalize the trial results. For these reasons, clinical trials may not be the most appropriate mechanism for estimating prevalence and incidence of neurological disease in these settings.

A particular challenge exists for studying neuropsychological function. It is critical to standardize the application of neurological and neuropsychological examinations and to have excellent translation of tests and associated instructions into local languages and dialects. The complexity of translation into various languages may have an impact on standardization. Questions that are of comparable difficulty in one language/culture may not have comparable difficulty in another, and may affect the interpretation of changes over time. Appropriate standardization can be accomplished through appropriate quality assurance and intersite and intrasite training. Appropriate normative data to improve interpretation is wanting in international settings. Without such normative data, there are difficulties in assessing the clinical relevance of changes and identifying individual patient impairment. Statistical significance of changes can still be obtained and valid treatment comparisons can still be made in randomized studies. Confounding can be controlled using statistical methods, but there will be difficulty interpreting the clinical relevance of any identified changes or treatment differences.

Summary and future research

The central and peripheral nervous system complications of HIV disease, the mechanisms of neuropathogenesis, and treatment-related effects have been well

characterized in the context of developed countries. Ongoing international treatment and research efforts are increasingly providing the infrastructure and access to HIV+ patient populations in developing countries such that the medical community has begun making inroads into studying specialized scientific areas, such as the nervous system, and learning about and treating the complications of HIV that are unique in those settings. The increased access to HIV physicians, researchers, caregivers and patients in Africa has helped to define and tailor a more specific neuroAIDS research agenda.

Although some common challenges and barriers exist, it is becoming clearer that each region and country in Africa must be considered independently in the fight against HIV/AIDS. For instance, the reported regional differences in neurocognitive outcome measures in response to ARV treatment highlight several important areas for more intensive research, including the need to further develop and improve the sensitivity and specificity of existing neuroassessment measures, to develop tests that can be appropriately compared across sites, and to improve methods for translating the tests from one language and context to another. Additional recognized research needs include normative data collection and general epidemiology research related to HIV neurological complications.

Beyond neuroassessment tools, preliminary research projects in Africa and other developing regions of the world suggest that different viral clades may have different pathogenicities and could therefore account for the regional differences in the clinical assessment of cognition and neuropathogenesis, thus indicating that viral clade analysis is an important area for continued and future research. In the United States, studies are addressing whether neuroprotective adjunctive agents or treatment strategies involving the use of ARV drugs with improved CNS penetration will improve the CNS/PNS complications of HIV for those individuals who are vulnerable to nervous system impairment; we anticipate that such studies will be extended in and important to developing countries as drug access is improved. Finally, coinfections and opportunistic infections are typically a larger problem in Africa than in the developed world. Studying HIV-related brain disease in the context of coinfections and treatments for conditions such as cryptococcal meningitis, tuberculosis, toxoplasma encephalitis, and syphilis are also important areas of investigation for learning about neuropathogenesis and clinical management of such patient populations.

Action items

1. More research is required to further examine the possible effects of population, demographic, environmental and cultural influences as well as the impact of viral genetic variation (HIV clades) on neurological disease progression.
2. Development of culturally specific and regionally sensitive assessment tools will be vital to the accurate comparison of neurological and neuropsychological data across sites.
3. Standardization of assessments and accumulation of appropriate normative data in international settings is required.
4. Further studies are needed to identify CNS infections, obtain neuroimaging, and investigate possible metabolic causes that may influence neurodevelopment of undernourished children in poorer communities.
5. More research is also needed to identify neurodevelopmental skills that can be assessed in children across SSA and to tailor culturally and language appropriate assessments across populations.
6. There is a need to research the neurodevelopment of children with maternal HIV in the context of coinfections such as malaria.
7. Because the more toxic ARVs are less expensive and more available in resource-poor settings, the populations at risk for toxic neuropathies and lactic acidosis are great and increasing; better characterization of the rates of these syndromes are required and the potential of adjunctive therapies such as minocycline for these symptoms warrants further investigation.
8. The major hurdles with diagnosis and treatment of OIs are primarily resource based. More research into cheaper and more effective diagnostic tools for diseases like neurosyphilis may lead to decreased morbidity and mortality in HIV-infected persons in regions at high risk of these diseases.

References

Aaron L, Saadoun D, Calatroni I, Launay O, Memain N, Vincent V, Marchal G, Dupont B, Bouchaud O, Valeyre D, Lortholary O (2004). Tuberculosis in HIV-infected patients: a comprehensive review. *Clin Microbiol Infect* **10:** 388–398.

Amod F (2006). AIDS associated toxoplasmosis. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Antinori A, Cingolani A, Giancola ML, Forbici F, De Luca A, Perno CF (2003). Clinical implications of HIV-1 drug resistance in the neurological compartment. *Scand J Infect Dis Suppl* **35(Suppl 106):** 41–44.

Belman AL, Diamond G, Dickson D, Horoupian D, Llena J, Lantos G, Rubinstein A (1988). Pediatric acquired immunodeficiency syndrome. Neurologic syndromes. *Am J Dis Child* **142:** 29–35.

Boffito M, Pillay D, Wilkins E (2006). Management of advanced HIV disease: resistance, antiretroviral brain penetration and malignancies. *Int J Clin Pract* **60:** 1098–1106.

Bonnet F, Bonarek M, Morlat P, Mercie P, Dupon M, Gemain MC, Malvy D, Bernard N, Pellegrin JL, Beylot J (2003). Risk factors for lactic acidosis in HIV-infected

patients treated with nucleoside reverse-transcriptase inhibitors: a case-control study. *Clin Infect Dis* **36:** 1324–1328.

Bottieau E, Noe A, Florence E, Colebunders R (2003). Multiple tuberculous brain abscesses in an HIV-infected patient successfully treated with HAART and antituberculous treatment. *Infection* **31:** 118–120.

Breen RA, Swaden L, Ballinger J, Lipman MC (2006). Tuberculosis and HIV co-infection: a practical therapeutic approach. *Drugs* **66:** 2299–2308.

Calza L, Manfredi R, Chiodo F (2005). Hyperlactataemia and lactic acidosis in HIV-infected patients receiving antiretroviral therapy. *Clin Nutr* **24:** 5–15.

Cashion MF, Banks WA, Bost KL, Kastin AJ (1999). Transmission routes of HIV-1 gp120 from brain to lymphoid tissues. *Brain Res* **822:** 26–33.

Childs EA, Lyles RH, Selnes OA, Chen B, Miller EN, Cohen BA, Becker JT, Mellors J, McArthur JC (1999). Plasma viral load and CD4 lymphocytes predict HIV-associated dementia and sensory neuropathy. *Neurology* **52:** 607–613.

Clements JE (2006). The SIV macaque model of HIV CNS disease. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Clements JE, Li M, Gama L, Bullock B, Carruth LM, Mankowski JL, Zink MC (2005). The central nervous system is a viral reservoir in simian immunodeficiency virus–infected macaques on combined antiretroviral therapy: a model for human immunodeficiency virus patients on highly active antiretroviral therapy. *J NeuroVirol* **11:** 180–189.

Clifford DB (2006). Ethiopian NeuroAIDS assessment. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Clifford DB, Mitike MT, Mekonnen Y, Zhang J, Zenebe G, Melaku Z, Zewde A, Gessesse N, Wolday D, Messele T, Teshome M, Evans S (2007). Neurological evaluation of untreated human immunodeficiency virus infected adults in Ethiopia. *J NeuroVirol* **13:** 67–72.

Coghlan ME, Sommadossi JP, Jhala NC, Many WJ, Saag MS, Johnson VA (2001). Symptomatic lactic acidosis in hospitalized antiretroviral-treated patients with human immunodeficiency virus infection: a report of 12 cases. *Clin Infect Dis* **33:** 1914–1921.

Cysique LA, Maruff P, Brew BJ (2004). Prevalence and pattern of neuropsychological impairment in human immunodeficiency virus-infected/acquired immunodeficiency syndrome (HIV/AIDS) patients across pre- and post-highly active antiretroviral therapy eras: a combined study of two cohorts. *J NeuroVirol* **10:** 350–357.

Cysique LA, Maruff P, Brew BJ (2006). Variable benefit in neuropsychological function in HIV-infected HAART-treated patients. *Neurology* **66:** 1447–1450.

Dabis F, Ekpini ER (2002). HIV-1/AIDS and maternal and child health in Africa. *Lancet* **359:** 2097–2104.

Dean GL, Edwards SG, Ives NJ, Matthews G, Fox EF, Navaratne L, Fisher M, Taylor GP, Miller R, Taylor CB, de Ruiter A, Pozniak AL (2002). Treatment of tuberculosis in HIV-infected persons in the era of highly active antiretroviral therapy. *AIDS* **16:** 75–83.

Eggers C, Hertogs K, Sturenburg HJ, van Lunzen J, Stellbrink HJ (2003). Delayed central nervous system virus suppression during highly active antiretroviral therapy is associated with HIV encephalopathy, but not with viral drug resistance or poor central nervous system drug penetration. *AIDS* **17:** 1897–1906.

Ellis RJ, Hsia K, Spector SA, Nelson JA, Heaton RK, Wallace MR, Abramson I, Atkinson JH, Grant I, McCutchan JA (1997). Cerebrospinal fluid human immunodeficiency virus type 1 RNA levels are elevated in neurocognitively impaired individuals with acquired immunodeficiency syndrome. HIV Neurobehavioral Research Center Group. *Ann Neurol* **42:** 679–688.

Enting RH, Hoetelmans RM, Lange JM, Burger DM, Beijnen JH, Portegies P (1998). Antiretroviral drugs and the central nervous system. *AIDS* **12:** 1941–1955.

Epstein LG, Sharer LR, Oleske JM, Connor EM, Goudsmit J, Bagdon L, Robert-Guroff M, Koenigsberger MR (1986). Neurologic manifestations of human immunodeficiency virus infection in children. *Pediatrics* **78:** 678–687.

Evans S (2006). Assessment of NeuroAIDS in Africa: statistical issues and design. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

FDA (2002). d4T and lactic acidosis with neuromuscular weakness: FDA warning. *AIDS Treat News* **378:** 5–6.

Ferrando SJ, Rabkin JG, van Gorp W, Lin SH, McElhiney M (2003). Longitudinal improvement in psychomotor processing speed is associated with potent combination antiretroviral therapy in HIV-1 infection. *J Neuropsychiatry Clin Neurosci* **15:** 208–214.

Gabuzda DH, Hirsch MS (1987). Neurologic manifestations of infection with human immunodeficiency virus. Clinical features and pathogenesis. *Ann Intern Med* **107:** 383–391.

Hakim J (2006). Epidemiology of HIV in Africa. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Hall C (2006). HIV associated dementia in the HAART era. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Herzmann C, Johnson MA, Youle M (2005). Long-term effect of acetyl-l-carnitine for antiretroviral toxic neuropathy. *HIV Clin Trials* **6:** 344–50.

Holding P (2006). Pediatric neuroassessment in Kenya: children exposed to multiple risks. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Hosseinipour M (2006). Lactic acidosis and ascending neuromuscular syndrome. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Imhof A, Ledergerber B, Gunthard HF, Haupts S, Weber R (2005). Risk factors for and outcome of hyperlactatemia in HIV-infected persons: is there a need for routine lactate monitoring? *Clin Infect Dis* **41:** 721–728.

Kaleebu P, French N, Mahe C, Yirrell D, Watera C, Lyagoba F, Nakiyingi J, Rutebemberwa A, Morgan D, Weber J, Gilks C, Whitworth J (2002). Effect of human immunodeficiency virus (HIV) type 1 envelope subtypes A and D on disease progression in a large cohort of HIV-1-positive persons in Uganda. *J Infect Dis* **185:** 1244–1250.

Kaleebu P, Nankya IL, Yirrell DL, Shafer LA, Kyosiimire-Lugemwa J, Lule DB, Morgan D, Beddows S, Weber J, Whitworth JA (2007). Relation between chemokine receptor use, disease stage, and HIV-1 subtypes A and D: results from a rural Ugandan cohort. *J Acquir Immune Defic Syndr* **45:** 28–33.

Kanki PJ, Hamel DJ, Sankale JL, Hsieh C, Thior I, Barin F, Woodcock SA, Gueye-Ndiaye A, Zhang E, Montano M, Siby T, Marlink R, I ND, Essex ME, S MB (1999). Human immunodeficiency virus type 1 subtypes differ in disease progression. *J Infect Dis* **179:** 68–73.

Kramer-Hammerle S, Rothenaigner I, Wolff H, Bell JE, Brack-Werner R (2005). Cells of the central nervous

system as targets and reservoirs of the human immunodeficiency virus. *Virus Res* **111:** 194–213.

Lalloo U (2006). CNS TB. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Langford TD, Letendre SL, Larrea GJ, Masliah E (2003). Changing patterns in the neuropathogenesis of HIV during the HAART era. *Brain Pathol* **13:** 195–210.

Liner KJ, 2nd, Hall CD, Robertson KR (2007). Impact of human immunodeficiency virus (HIV) subtypes on HIV-associated neurological disease. *J NeuroVirol* **13:** 291–304.

Lobato MN, Caldwell MB, Ng P, Oxtoby MJ (1995). Encephalopathy in children with perinatally acquired human immunodeficiency virus infection. Pediatric Spectrum of Disease Clinical Consortium. *J Pediatr* **126:** 710–715.

Marra CM (2006). Neurosyphilis. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

McArthur JC, McClernon DR, Cronin MF, Nance-Sproson TE, Saah AJ, St Clair M, Lanier ER (1997). Relationship between human immunodeficiency virus-associated dementia and viral load in cerebrospinal fluid and brain. *Ann Neurol* **42:** 689–698.

Merry C (2006). Antiretroviral rollout in Africa. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Mielke J (2006). Peripheral nerve disease in HIV-infected subjects. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Moore RD, Wong WM, Keruly JC, McArthur JC (2000). Incidence of neuropathy in HIV-infected patients on monotherapy versus those on combination therapy with didanosine, stavudine and hydroxyurea. *AIDS* **14:** 273–278.

Morgello S, Estanislao L, Simpson D, Geraci A, DiRocco A, Gerits P, Ryan E, Yakoushina T, Khan S, Mahboob R, Naseer M, Dorfman D, Sharp V (2004). HIV-associated distal sensory polyneuropathy in the era of highly active antiretroviral therapy: the Manhattan HIV Brain Bank. *Arch Neurol* **61:** 546–551.

Moyle GJ, Datta D, Mandalia S, Morlese J, Asboe D, Gazzard BG (2002). Hyperlactataemia and lactic acidosis during antiretroviral therapy: relevance, reproducibility and possible risk factors. *AIDS* **16:** 1341–1349.

Nakasujja N (2006). HIV/AIDS and the use of HAART in Uganda. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Neuenburg JK, Brodt HR, Herndier BG, Bickel M, Bacchetti P, Price RW, Grant RM, Schlote W (2002). HIV-related neuropathology, 1985 to 1999: rising prevalence of HIV encephalopathy in the era of highly active antiretroviral therapy. *J Acquir Immune Defic Syndr* **31:** 171–177.

Newton C (2006). The definition of HIV encephalopathy in children living in Africa. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Osio M, Muscia F, Zampini L, Nascimbene C, Mailland E, Cargnel A, Mariani C (2006). Acetyl-l-carnitine in the treatment of painful antiretroviral toxic neuropathy in human immunodeficiency virus patients: an open label study. *J Peripher Nerv Syst* **11:** 72–76.

Robertson K, Fiscus S, Kapoor C, Robertson W, Schneider G, Shepard R, Howe L, Silva S, Hall C (1998). CSF, plasma viral load and HIV associated dementia. *J NeuroVirol* **4:** 90–94.

Robertson KR, Robertson WT, Ford S, Watson D, Fiscus S, Harp AG, Hall CD (2004). Highly active antiretroviral therapy improves neurocognitive functioning. *J Acquir Immune Defic Syndr* **36:** 562–566.

Saavedra-Lozano J, Ramos JT, Sanz F, Navarro ML, de Jose MI, Martin-Fontelos P, Mellado MJ, Leal JA, Rodriguez C, Luque I, Madison SJ, Irlbeck D, Lanier ER, Ramilo O (2006). Salvage therapy with abacavir and other reverse transcriptase inhibitors for human immunodeficiency-associated encephalopathy. *Pediatr Infect Dis J* **25:** 1142–1152.

Sacktor N (2006). Assessment of HIV dementia in Uganda: results from the Academic Alliance Cohort. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Sacktor N, Nakasujja N, Robertson K, Clifford DB (2007). HIV-associated cognitive impairment in sub-Saharan Africa—the potential effect of clade diversity. *Nat Clin Pract Neurol* **3:** 436–443.

Sacktor N, Nakasujja N, Skolasky R, Robertson K, Wong M, Musisi S, Ronald A, Katabira E (2006). Antiretroviral therapy improves cognitive impairment in HIV+ individuals in sub-Saharan Africa. *Neurology* **67:** 311–314.

Sacktor NC, Skolasky RL, Lyles RH, Esposito D, Selnes OA, McArthur JC (2000). Improvement in HIV-associated motor slowing after antiretroviral therapy including protease inhibitors. *J NeuroVirol* **6:** 84–88.

Schifitto G, McDermott MP, McArthur JC, Marder K, Sacktor N, Epstein L, Kieburtz K (2002). Incidence of and risk factors for HIV-associated distal sensory polyneuropathy. *Neurology* **58:** 1764–1768.

Sevigny JJ, Albert SM, McDermott MP, McArthur JC, Sacktor N, Conant K, Schifitto G, Selnes OA, Stern Y, McClernon DR, Palumbo D, Kieburtz K, Riggs G, Cohen B, Epstein LG, Marder K (2004). Evaluation of HIV RNA and markers of immune activation as predictors of HIV-associated dementia. *Neurology* **63:** 2084–2090.

Shanbhag MC, Rutstein RM, Zaoutis T, Zhao H, Chao D, Radcliffe J (2005). Neurocognitive functioning in pediatric human immunodeficiency virus infection: effects of combined therapy. *Arch Pediatr Adolesc Med* **159:** 651–656.

Simpson DM, Kitch D, Evans SR, McArthur JC, Asmuth DM, Cohen B, Goodkin K, Gerschenson M, So Y, Marra CM, Diaz-Arrastia R, Shriver S, Millar L, Clifford DB (2006). HIV neuropathy natural history cohort study: assessment measures and risk factors. *Neurology* **66:** 1679–1687.

Stankoff B, Calvez V, Suarez S, Bossi P, Rosenblum O, Conquy L, Turell E, Dubard T, Coutellier A, Baril L, Bricaire F, Lacomblez L, Lubetzki C (1999). Plasma and cerebrospinal fluid human immunodeficiency virus type-1 (HIV-1) RNA levels in HIV-related cognitive impairment. *Eur J Neurol* **6:** 669–675.

Tozzi V, Balestra P, Bellagamba R, Corpolongo A, Salvatori MF, Visco-Comandini U, Vlassi C, Giulianelli M, Galgani S, Antinori A, Narciso P (2007). Persistence of neuropsychologic deficits despite long-term highly active antiretroviral therapy in patients with HIV-related neurocognitive impairment: prevalence and risk factors. *J Acquir Immune Defic Syndr* **45:** 174–182.

Tozzi V, Balestra P, Galgani S, Narciso P, Ferri F, Sebastiani G, D'Amato C, Affricano C, Pigorini F, Pau FM, De Felici A, Benedetto A (1999). Positive and sustained effects of highly active antiretroviral therapy on HIV-1-associated neurocognitive impairment. *AIDS* **13:** 1889–1897.

Tozzi V, Balestra P, Galgani S, Narciso P, Sampaolesi A, Antinori A, Giulianelli M, Serraino D, Ippolito G (2001). Changes in neurocognitive performance in a cohort of patients treated with HAART for 3 years. *J Acquir Immune Defic Syndr* **28:** 19–27.

Traore M (2006). Mali and NeuroAIDS. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Van Rie A (2006). Kinshasa, Congo and children with HIV. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.

Vasan A, Renjifo B, Hertzmark E, Chaplin B, Msamanga G, Essex M, Fawzi W, Hunter D (2006). Different rates of disease progression of HIV type 1 infection in Tanzania based on infecting subtype. *Clin Infect Dis* **42:** 843–852.

Wynn HE, Brundage RC, Fletcher CV (2002). Clinical implications of CNS penetration of antiretroviral drugs. *CNS Drugs* **16:** 595–609.

Zink MC (2006). Impact of minocycline on SIV CNS and PNS disease. In: *Second Assessment of NeuroAIDS in Africa*: Arusha, Tanzania.