Journal of NeuroVirology, 13: 195–202, 2007

ISSN: 1355-0284 print / 1538-2443 online
DOI: 10.1080/13550280701258407

informa
healthcare

Neuropsychological deficits in human immunodeficiency virus type 1 clade C–seropositive adults from South India

Jayashree Das Gupta,[1] P Satishchandra,[2] Kumarpillai Gopukumar,[1] Frances Wilkie,[3] Drenna Waldrop-Valverde,[3] Ronald Ellis,[4] Raymond Ownby,[3] D K Subbakrishna,[5] Anita Desai,[6] Anupa Kamat,[6] V Ravi,[6] B S Rao,[7] K S Satish,[7] and Mahendra Kumar[3]

[1]*Mental Health & Social Psychology,* [2]*Neurology,* [5]*Biostatistics and* [6]*Neurovirology, National Institute of Mental Health & Neuro Sciences, Bangalore, Karnataka, India;*
[3]*Department of Psychiatry and Behavioral Sciences, University of Miami School of Medicine, Florida, USA;*
[4]*HIV Neurobehavioral Research Center, University of California, San Diego, La Jolla, California, USA;*
[7]*Seva Free Clinic, Bangalore, Karnataka, India*

Most studies of cognitive functioning in human immunodeficiency virus type 1 (HIV-1)–seropositive (HIV-1+) subjects have been done in the United States and Europe, where clade B infections predominate. However, in other parts of the world such as South India, where clade C HIV is most common, the prevalence of HIV-1 is increasing. Standardized neuropsychological tests were used to assess cognitive functioning in a sample of 119 adults infected with clade C HIV-1 who were not on antiretroviral medications. The subjects did not have neurological or psychiatric illness and were functioning adequately. Neuropsychological test performance was compared with gender-, age-, and education-matched normative data derived from a sample of 540 healthy volunteers and a matched cohort of 126 healthy, HIV-1–seronegative individuals. Among the seropositive subjects, 60.5% had mild to moderate cognitive deficits characterized by deficits in the domains of fluency, working memory, and learning and memory. None of the subjects had severe cognitive deficits. The HIV-1+ sample was classified into groups according to the level of immune suppression as defined by CD4 count (<200, 201–499, and >500 cells/mm^3) and viral load (<5000, 5001–30,000, 30,001–99,999, 100,000–1,000,000, and >1,000,001 copies). Although the most immunosuppressed group (CD4 count <200 cells/mm^3 or viral load >1,000,001 copies) was small, their rate of impairment in visual working memory was greater when compared to groups with better immune functioning. Mild to moderate cognitive deficits can be identified on standardized neuropsychological tests in clade C–infected HIV-1+ adults who do not have any clinically identifiable functional impairment. The prevalence of cognitive deficits is similar to that reported in antiretroviral treatment–naïve individuals infected with clade B virus in the western world. *Journal of NeuroVirology* (2007) **13,** 195–202.

Keywords: clade C; cognition; HIV; neuropsychology; working memory

Introduction

The human immunodeficiency virus (HIV) enters the central nervous system (CNS) early in the course of infection (Scaravilli, 1993). Primary complications of HIV infection are those, which are directly related to the effect of HIV on the brain. These include neurocognitive complications as well as neurobiological

Address correspondence to Prof. Shobini Rao, Department of Mental Health & Social Psychology, National Institute of Mental Health & Neuro Sciences, Bangalore 560029, Karnataka, India. E-mail: shobini@nimhans.kar.nic.in

This study has been supported by grant R01 NS 41205 from NIH, USA, and approved by ICMR, New Delhi.

Received 27 October 2006; revised 18 December 2006; accepted 24 January 2007.

complications such as HIV meningitis and neuropathy (Grant *et al*, 1999). Most of the literature addressing cognition in HIV is from Europe and the United States where the predominant virus strain is clade B (Osmanov *et al*, 2002). In India the virus strain is predominantly clade C (Siddappa *et al*, 2004), although there are a few anecdotal reports of recombinant strains. Epidemiological trends show that HIV infection due to clade C is increasing in Asia and Africa.

Neuropsychological assessment is one of the important noninvasive techniques used to evaluate the impact of HIV infection on the CNS. Neurocognitive complications are common in HIV infection. Reports from the United States have identified impairments ranging from subsyndromal neuropsychological impairment to HIV-associated dementia (American Academy of Neurology AIDS Task Force, 1991). The early detection of neuropsychological impairment is important as subclinical changes have been associated with poor quality of life, work disability, and mortality (Osowiecki *et al*, 2000; Albert *et al*, 1995; Ellis *et al*, 1997; Heaton *et al*, 1994; Mayeux *et al*, 1993; Wilkie *et al*, 1998). In addition, the early detection of cognitive deficits in HIV-seropositive individuals could lead to better management. Some reports have suggested early treatment could reduce, minimize, or possibly reverse the cognitive deficits associated with HIV infection (Tartar *et al*, 2004).

Because very little is known about the presence and/or pattern of cognitive deficits in clade C HIV infection, the present study examined cognitive functioning in a group of HIV-1–seropositive individuals from South India with clade C infection.

Results

The final sample studied consisted of 119 HIV-1–seropositive adults (men, $N=52$; women, $N=67$) ranging in age from 20 to 44 years (mean = 29.9, SD = 5.6). The participants had from 0 to 15 years of education (mean = 7.6, SD = 4.8). Of the sample, 22% had received no formal education. The predominant mode of transmission of HIV was through heterosexual contact. Mean CD4 count of the sample was 396.8 cells/mm^3 (SD = 212.5). The healthy, HIV-seronegative group consisted of 126 adults (64 men, 62 women) with mean age of 31.9 years (SD = 9.9) and mean number of years of education of 7.75 years (SD = 6.6). There was no difference in demographic characteristics of gender ($\chi^2=0.9$; $P=$ n.s. [nonsignificant]), age ($t=-1.9$; $P=$ n.s.), and education ($t=-1.9$; $P=$ n.s.) between the HIV-seropositive and healthy, HIV-seronegative group.

Prevalence and severity of cognitive deficits

A cognitive profile was obtained for each subject in the HIV-seropositive group by comparing test performance with the normative data. A deficit was considered to be present in a cognitive domain if performance was below the 15th percentile on even one test variable measuring performance in that particular domain, e.g., a deficit in the domain of motor speed was considered if performance was impaired only on the test variable Finger Tapping right hand. The number of cognitive deficits each subject had was calculated.

The severity of cognitive impairments was further classified based on the percentage of test variables falling below the 15th percentile. Subjects were considered to have mild impairment if they had deficits on more than one test variable but less than 25% of test variables, i.e., 2 to 3 of 12 test variables impaired. A deficit between 25% and 50% of test variables (i.e., 4 to 6 test variables impaired) was designated as mild to moderate impairment. Moderate to severe impairment was defined as deficits between 50% and 75% of test variables (i.e., 7 to 9 test variables impaired), and severe impairment was defined as deficits on greater than 75% of test variables, (i.e., >10 test variables impaired).

When compared with gender-, age-, and education-matched normative data, mild cognitive impairment was present in 35.3%, mild-moderate impairment was present in 21.9%, and moderate-severe impairment was present in 3.3% of the sample. None of the subjects had severe cognitive impairment. Combining the different categories of severity 60.5% of subjects had cognitive deficits on at least 2 variables.

Cognitive deficits and immune suppression

Cognitive deficits have been known to vary in different levels of immune suppression (Heaton *et al*, 1995). Data was analyzed to look at the difference in cognitive performance across groups differing on immune suppression as defined by CD4 count and plasma viral load. Differences in sociodemographic characteristics across the immune suppression groups in terms of age, gender, and years of education were analyzed using analysis of variance (ANOVA) or chi square. Analysis revealed significant age and gender differences across the groups based on CD4 count. Subjects in the <200 cells/mm^3 CD4 count group were significantly older (mean = 34.3 years, SD = 5.9). There were also a significantly greater number of males in this group ($\chi^2=8.19$, $df=2$, $P=.017$). Cognitive performance based on raw test scores was compared across the immune suppression groups using ANCOVA to control for the influence of sociodemographic covariates of age and gender. Groups defined by plasma viral load levels were comparable in terms of age ($F=0.78$, $df=4$, $P=$ n.s.), years of education ($F=0.53$, $df=4$, $P=$ n.s.), and gender ($\chi^2=0.79$, $df=4$, $P=$ n.s). Differences in cognitive performance across plasma viral load groups were analyzed using ANOVA.

Findings revealed test performance did not differ across levels of immune suppression for majority of the test variables (Table 1). However, visual working

Table 1 Comparison of deficits in HIV+ subjects with seronegative controls and difference in cognitive functioning across immune suppression groups

Test variable	X^1 (based on number of deficits in HIV+ and HIV-groups) df = 1	Level of significance	Differences across CD4 count groups ANCOVA (Based on raw test scores) F ratio (df = 2)	Level of significance	Difference across plasma viral load groups ANOVA (Based on raw test scores) F ratio (df = 4)	Level of significance
FT LH	2.10	0.15	0.08	0.92	0.38	0.83
FT RH	3.59	0.06	0.29	0.75	0.44	0.78
AF	0.69	0.40	0.86	0.43	0.17	0.95
PF	6.44	0.01	1.82	0.83	1.23	0.30
VM 1	0.14	0.71	0.67	0.52	1.22	0.31
VM 2	9.18	0.00	0.00	0.99	0.07	0.99
VIM 1	0.13	0.72	0.60	0.55	3.06	0.02
VIM 2	0.25	0.62	4.31	0.01	1.92	0.11
AVLTI-V Total	11.39	0.00	0.62	0.54	0.66	0.62
AVLT IR	7.33	0.00	1.06	0.35	0.55	0.70
AVLT DR	0.97	0.00	0.39	0.68	0.34	0.85
TNPMM	0.00	0.95	0.85	0.43	0.68	0.61

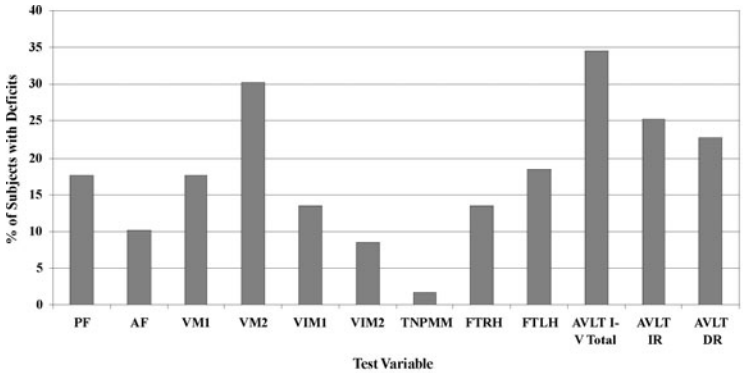
Note: PF = Phonemic Fluency, AF = Animal Fluency, FT LH = Finger Tapping left Hand, FT RH = Finger Tapping right Hand, VM1 = Verbal Working Memory 1-back hits, VM2 = Verbal Working Memory 2-back hits, VIM1 = Visual Working Memory 1-back hits, VIM2 = Visual Working Memory 2-back hits, TNPMM = Total number problems solved with minimum moves on Tower of London Test, AVLT = Auditory Verbal Learing Test, IR = Immediate Recall, DR = Delayed Recall.

memory was significantly poorer in groups with advanced immune suppression, i.e., when there were low CD4 counts or high plasma viral loads. This suggests greater deficits are seen in working memory when CD4 counts fall below 200 cells/mm^3 or plasma viral load is > 1,000,001 copies.

Profile of cognitive deficits in HIV-1–seropositive adults

The percentage of patients with deficits on each test variable in the HIV-seropositive group was analyzed to identify any emergent pattern of cognitive deficits. Results show the sample of HIV-1 seropositive adults have cognitive deficits occurring across various domains. As depicted in Figure 1, the percentage of subjects with deficits on each test variable varied from 2.5% (total number problems solved with minimum moves on TOL) to 33.6% (AVLT Trials I–V Total). The largest proportion of subjects with deficits was found in verbal learning (33.6% sample with deficits) and manipulation of verbal information in working memory (30% sample with deficits).

A comparison of deficits between the HIV-seropositive group and the matched cohort of healthy seronegative individuals who had also undergone complete neuropsychological assessment was done using chi-square analysis. A significantly greater number of subjects in the HIV seropositive group had deficits in fluency (PF $\chi^2 = 6.4$, $df = 1$, $P = .01$), learning and memory (AVLT Trials I–V Total $\chi^2 = 11.4$, $df = 1$, $P = .00$; AVLT IR $\chi^2 = 7.3$, $df = 1$, $P = 0.00$; AVLT DR $\chi^2 = 6.9$, $df = 1$, $P = .00$), and verbal



Note: PF= Phonemic Fluency, AF=Animal Fluency, FT LH=Finger Tapping Left Hand, FT RH= Finger Tapping Right Hand, VM1=Verbal Working Memory 1-back hits, VM2= Verbal Working Memory 2-back hits, VIM1= Visual Working Memory 1-back hits, VIM2= Visual Working Memory 2-back hits, TNPMM= Total number problems solved with minimum moves on Tower of London Test, AVLT= Auditory Verbal Learning Test, IR=Immediate Recall, DR=Delayed Recall.

Figure 1 Percentage of Subjects with Deficits on Test Variables.

working memory (VM2 $\chi^2 = 9.1$, $P = .00$). These findings suggest significant differences in cognitive profiles between HIV-seropositive adults and healthy seronegative individuals.

Discussion

The present study examined cognitive deficits in a sample of HIV-1–seropositive adults from South India, where the infection was due to clade C virus. At the time of entry to the study, clinically, none of the seropositive subjects had either active systemic illness, or symptoms associated with nervous system involvement, including neurocognitive complaints or any recurrent opportunistic infections. Subjects were excluded from the study if they had a previous history suggestive of neurological or psychiatric illness, head injury, substance dependence, hypertension, or diabetes. Subjects on any CNS-active medication or already receiving antiretroviral therapy were also excluded. Therefore adequate precautions had been taken to exclude factors other than the HIV-1 infection from influencing their cognitive performance. However, on standardized neuropsychological tests, cognitive deficits were identified in the sample. There were mild and mild to moderate cognitive deficits in 57.2% of subjects. Moderate to severe deficits were present in a very small number, i.e., 3.3%, and none had severe deficits. The total prevalence of cognitive deficits being 60.5%. However, none of the subjects included in the present cohort had a cognitive impairment amounting to either mild motor/cognitive impairment or HIV-associated dementia. Subjects did not report any significant change in their ability to carry out routine daily activities. In addition the subjects were satisfied with their quality of life (Chandra *et al*, 2006).

A preliminary report from South India on 30 seropositive individuals with advanced HIV-1 clade C virus infection reported that 56% of patients had cognitive deficits in two cognitive domains (Yepthomi *et al*, 2006). The performance of seropositive individuals was considered to be deficient if the score was 1.5 SD below the mean performance of a comparison group of 30 healthy seronegative controls. The sample was restricted to individuals with severe levels of immune suppression, characterized by median CD4 cell count of 97. In contrast, our sample is large consisting of 119 HIV-1 clade C–seropositive adults with immune suppression levels ranging from mild to severe. The tests used in the present study have been standardized and validated in the Indian population (Rao *et al*, 2004). The definition of a cognitive deficit is based on a large normative database of 540 normal individuals. In addition, the tests have been found suitable for even subjects with no formal education. Validation of neuropsychological tests is of particular importance in developing countries considering cultural variations and specific challenges such as suitability of tests for individuals with lower levels of education. Cognitive impairment has also been graded into different categories. It is remarkable that the two studies on HIV-1 infection with clade C virus have found similar prevalence rates of cognitive dysfunction, i.e., 60.5% in our study and 56% in the study of Yepthomi *et al* (2006). The presence of mild to moderate deficits is consistent with findings from prior studies, which have documented deficits in clade B virus (Heaton *et al*, 1995; Reger *et al*, 2002). These reports have noted the prevalence rates of cognitive deficits to vary between 30% and 66% (White *et al*, 1995).

In the present study, cognitive performance on the neuropsychological test battery did not differ according to immune suppression levels, except for visual working memory. Previous HIV research from the United States, where clade B viral strain predominates, have found that in individuals with advanced immune suppression, the severity of cognitive deficits is associated with lower CD4 counts (Ellis *et al*, 1997). However, when the CD4 counts are high, indicating better immune functioning, cognitive impairment is only weakly associated with immune suppression (Grant *et al*, 1999). The similarity of cognitive profiles across different levels of immune suppression in the present study could be due to the fact that in clade C virus, the dicysteine motif of the Tat protein is conserved and this mutation may be helping to conserve cognition. The low, i.e., less than 2% to 4%, prevalence of HIV-associated dementia in India (Satishchandra *et al*, 2000; Wadia *et al*, 2001) has been hypothesized to be related to the above mutation of Tat protein (Ranga *et al*, 2004). Compared with Tat protein in clade B, the Tat protein in clade C virus is associated with greater replication due to nuclear factor kappa B (NFκB) transcription factor (Kurosu *et al*, 2002). Despite this greater replication, damage to the neuronal cells in the brain is less in clade C virus as compared with clade B virus (Seth, 2006). Further, the inflammatory response to the clade C virus is less compared with clade B virus (Siddappa *et al*, 2006). The decreased damage to the brain neuronal cells and the decreased inflammatory response in clade C virus could be associated with lower levels of cognitive impairment even when the disease has progressed, as indicated by severe levels of immune suppression.

The cognitive profile in the seropositive individuals of the present study is suggestive of frontostriatal pathology. The cognitive profile in the seropositive sample differed from that of the healthy seronegative controls in the domains of phonemic fluency, verbal working memory, and verbal learning and memory. Thus cognitive deficits in these domains are associated with HIV-1 infection. Previous studies have also reported similar deficits that support the involvement of subcortical and frontostriatal brain processes in clade B infection (Aylward *et al*, 1993; Kieburtz *et al*, 1996; Stern *et al*, 1992; York *et al*, 2001). Again,

working memory is compromised in severe immune suppression, i.e., CD4 below 200 cells/mm^3 and plasma viral loads higher than >1,000,001 copies. Deficits in working memory are suggestive of prefrontal involvement (Baddeley, 1992). Earlier studies have shown evidence of working memory deficits in HIV-infected subjects supporting frontal lobe pathology in HIV infection (Everall *et al*, 1991; Hinkin *et al*, 2002; Stout *et al*, 1995).

Previous studies have reported a lower prevalence of HIV dementia in clade C infection. However, the absence of severe cognitive deficits in the present sample may be due to the stringent exclusion criteria resulting in a sampling bias. Individuals with any symptoms associated with nervous system involvement, including neurocognitive complaints and history of neurological or psychiatric illness, were excluded from the study. Therefore, by definition, none of the subjects could be diagnosed with HIV-associated dementia. The sampling criteria may have also excluded individuals with advanced HIV who may have severe cognitive impairment related to advanced HIV disease. The cohort in the present study is being followed up as part of an ongoing longitudinal study. Further research is required to address questions regarding the progression of cognitive deficits in clade C infection.

Findings of the present study show the presence of mild to moderate cognitive deficits on standardized neuropsychological tests in HIV-1 clade C–infected adults from South India. In addition, patients did not have any clinically identifiable functional impairment. They were maintaining well and able to carry out routine daily activities. The prevalence of cognitive deficits of 60.5% is similar to that seen in clade B infection. Nature of the deficits is suggestive of frontostriatal involvement.

Methods

Participants

The present study is part of an ongoing NIH funded research project studying neurological progression in HIV infection. Participants were recruited between October 2003 and December 2004 after an initial screening from a peripheral outpatient clinic for HIV-seropositive people, and a HIV outpatient weekly clinic at the National Institute of Mental Health and Neuro Sciences (NIMHANS), Bangalore. Subjects were excluded from the study if they had a previous history suggestive of neurological or psychiatric illness, head injury, substance dependence, hypertension, or diabetes. Subjects on any CNS-active medication or already receiving antiretroviral therapy were not included in the study. Subjects with any symptoms associated with nervous system involvement or any recurrent opportunistic infections were excluded. Subjects with neurocognitive complaints were also excluded.

The purpose of the study was explained to persons meeting these criteria and written informed consent was obtained for their participation. The ethical board of NIMHANS, Bangalore, and Institutional Review Board (IRB), University of Miami, USA, approved the study.

Participants underwent a detailed clinical examination. At the time of screening, 18 participants were excluded in view of neurological deficits, which were mainly peripheral neuropathy, mononeuritis multiplex, and hemiplegia. Only 2 of the 18 participants excluded had evidence suggestive of HIV encephalopathy and complained of cognitive deficits. None of the participants included in the present study complained of marked deficits in everyday functioning due to cognitive impairment. The significant other accompanying the participants also corroborated no marked change in the participant's every day functioning. Clinically there were no symptoms of dementia in any of the participants. Serological examination was conducted according to National AIDS Controls Organization (NACO) (Government of India) guidelines (NACO, 1999). HIV serostatus was confirmed using the NACO (Government of India) guidelines and strategy. Serum samples were tested using a single enzyme-linked immunosorbent assay (ELISA)/rapid immunoassay. Reactive samples were reconfirmed using two more ELISA-based assays as well as Western blot. CD4 counts were obtained for each subject using a flow cytometric analysis of peripheral venous blood (FACS Count, Becton Dickenson, USA). Viral loads (real time polymerase chain reaction [PCR]) were also assessed using standard laboratory procedures. All patients were subtyped and were confirmed as cases of clade C infection using the procedure for specific subtyping described by Siddappa *et al* (2004).

A review of patient chart details revealed the following number of patients in the sample had previous history of systemic illness for which they had been treated: 16 had pulmonary tuberculosis, 6 had extra pulmonary tuberculosis mainly in the form of tuberculous lymphadenitis, 26 had herpes zoster, 2 had herpes labialis, 26 had herpes genitalis, and 2 had oral candidiasis. Of the sample 14% had history of more than one systemic illness. At study entry, all subjects had completely recovered from systemic illness and had no active symptoms. None of the subjects had significant weight loss, persistent fever, or diarrhea associated with acquired immunodeficiency syndrome (AIDS). In addition, patients did not have any clinically identifiable functional impairment and were otherwise maintaining well and able to carry out routine daily activities. Hence according to NACO guidelines, none of the patients met criteria for AIDS (NACO, 1999).

In India, CD4 count is not determined routinely as a part of HIV testing in most centers because of the cost involved. For this reason, the NACO

guidelines do not classify patients according to CD4 count. In the present study, in order to examine the relationship between immune suppression and cognitive functioning, participants were categorized into three groups according to their CD4 cell counts (Centers for Disease Control and Prevention [CDC], 1992), i.e., <200 (16.4%), 201–499 (55.2%), and >500 (28.4%) cells/mm^3. Plasma viral loads were available for 114 subjects. Subjects were classified into the following categories according to viral load <5000 (11.4%), 5001–30,000 (21.9%), 30,001–99,999 (18.4%), 100,000–1,000,000 (36.0%), and >1,000,001 (12.3%) copies.

Neuropsychological evaluation

A comprehensive neuropsychological assessment was carried out by trained neuropsychologists to assess cognitive functioning. The following cognitive domains were assessed: motor speed, fluency (category and phonemic fluency), working memory (verbal and visual), planning, verbal learning, and verbal memory. None of these were paper-pencil tests, or tests that required reading, making the battery suitable even for individuals with no formal schooling. The tests used for the present study have been standardized for use in the Indian population (Rao *et al*, 2004).

Motor speed: To assess the domain of motor speed the *Finger Tapping Test* (Halstead, 1947) was administered. The test consists of a tapping key mounted on a box with an attached electronic counter. The subject was required to tap the key as many times as possible within a 10-s trial. Five 10-s trials were administered for both the right and left hand with a rest pause of 30 s after the third trial. Number of taps with the forefinger of each hand was recorded. The average number of taps across trials for the right hand and left hand was used for analyses.

Verbal fluency: To assess phonemic fluency, the *Phonemic Fluency Test* (Rao *et al*, 2004) was administered. This test is analogous to the Controlled Oral Word Association Test (Benton and Hamsher, 1989). To make the test suitable for use in Indian languages, the phonemes "Ka," "Pa," and "Ma" were used. The subject was required to generate words beginning with each of the phonemes. One-minute time was given for each phoneme. The score used was the average number of words generated. The *Animal Names Test* (Lezak, 1995) was administered to assess category fluency. The subject was asked to generate animal names, excluding fish, birds, and snakes, for 1 min. The score used was the total number of animal names generated.

Working memory: To assess the domain of verbal working memory, *Verbal N-Back Task* (Rao *et al*, 2004) was administered. The test is a modification of the task developed by Smith and Jonides (1999). Test stimuli comprise two lists of 30 randomly arranged phonemes, which are read aloud to the subject. In the 1-back task, the subject is instructed to say, "yes" whenever the phoneme is repeated. In the 2-back task, the subject is instructed to respond whenever a phoneme is repeated after an unrelated phoneme. The score is the number of correct responses in the 1-back and 2-back tasks. The *Visual N-Back Task* (Rao et al, 2004) was administered to assess visual working memory. The test is modified from the task developed by Smith and Jonides (1999). The 1-back and 2-back procedures are similar to that of the Verbal N-back Task. The test stimuli are 36 cards containing dots printed randomly in positions around an imaginary circle. In the 1-back task, the subject is instructed to respond whenever the dot appears in the same position on two consecutive cards. In the 2-back task, the subject is required to respond whenever the dot appears in the same position on alternate cards. The score used for analyses is the number of correct responses on the 1-and 2-back tasks.

Planning: To assess the domain of planning, the *Tower of London Test* (TOL) was administered (Shallice, 1982). The test stimuli consist of two boards with three pegs of different sizes and three different colored balls (red, blue, and green). The subject is required to match the arrangement on the examiner's board using the minimum number of moves. A total of 14 problems of increasing complexity are given. The total number of problems solved with minimum moves (TNPMM) was used for analyses in the present study.

Verbal learning and memory: The Auditory Verbal Learning Test (AVLT) was administered, which assesses immediate memory span, new learning, susceptibility for interference, and recognition memory (Schmidt, 1996). Subjects were required to recall as many words as possible from a 15-word list that was read aloud to them. Five such learning trials were administered. Following this an interference trial was administered in a similar manner using a different word list. An immediate recall, a 20-min delayed recall without prior warning, and a recognition trial of the original list was also administered. The present study used the World Health Organization (WHO) word lists (Maj *et al*, 1993), which were translated into different Indian languages. The test was administered and scored according to the procedure given by Schmidt (1996). For analyses, the scores Trial I-V Total, Immediate Recall, and Delayed Recall of the word list were considered.

Data analysis

Cognitive functioning in the HIV-seropositive sample was compared with normative data derived from a group of 540 normal healthy volunteers (Rao *et al*, 2004). Healthy volunteers were recruited from

relatives of patients admitted at the hospital, students, and from the community at large. Healthy volunteers who obtained a score of >2 on the General Health Questionnaire (GHQ-12) developed by Goldberg and Williams (1988) were excluded, as these persons met criteria for psychological distress, that itself could influence cognitive functioning. Healthy volunteers were also excluded if they had a previous history of neurological, neurosurgical, or psychiatric illness or substance dependence and family history of alcohol dependence, schizophrenia or bipolar disorder. The sample was divided into three age ranges (16–30, 31–50, and 51–65 years) and three education groups (no formal schooling, 1 to 10 years, and greater than 10 years of formal education) separately for males and females. For each test variable, percentile scores were calculated. The 15th percentile score (1 SD below the mean) was taken as the cut off score (Heaton *et al*, 1995). Cutoff scores were calculated for each group based on age, education, and gender. A deficit was defined as a test score falling below the 15th percentile (Heaton *et al*, 1995). Validation for each of the tests has been carried out on patient groups with focal lesions, refractory epilepsy, head injury, and Parkinson disease (Rao *et al*, 2004).

In order to examine the prevalence of cognitive deficits, a cognitive profile was obtained for each patient in the HIV-seropositive group by comparing test performance with the normative data described above. Based on the number of test variables falling below the 15th percentile, the severity of cognitive impairment was further classified. In addition to comparison with normative data, the cognitive profile of the HIV-seropositive group was compared with that of healthy HIV-seronegative individuals matched for age, education, and gender with the HIV-seropositive group to confirm that the cognitive profile is unique to the HIV-seropositive sample.

References

Albert SM, Marder KM, Dooneief G, Bell K, Sano M, Todak G, Stern Y (1995). Neuropsychologic impairment in early HIV infection: a risk factor for work disability. *Arch Neurol* **52:** 525–530.

Aylward EH, Henderer JD, McArthur JC, Bretschneider PD, Harris GD, Barta PE, Pearlson GD (1993). Reduced basal ganglia volume in HIV-1 associated dementia: results from quantitative neuroimaging. *Neurology* **10:** 2099–2104.

American Academy of Neurology AIDS Task Force. (1991). Nomenclature and research case definitions for neurologic manifestations of human immunodeficiency virus-type 1 (HIV-1) infection. *Neurology* **41:** 778–785.

Baddeley A (1992). Working memory. *Science* **255:** 556–559.

Benton AL, Hamsher KS (1989). *Multilingual Aphasia Examination.* New York: Oxford University Press.

Centers for Disease Control and Prevention (CDC). (1992). 1993 Revised classification system for HIV infection and expanded surveillance case definition for AIDS among adolescents and adults. *MMWR* **41(RR-17):** 1–19.

Chandra PS, Gandhi C, Satishchandra P, Kamat A, Desai A, Ravi V, Ownby RL, Subbakrishna DK, Kumar M. (2006). Quality of life in HIV subtype C infection among asymptomatic subjects and its association with CD4 counts and viral loads—a study from South India. Qual Life Res Oct 11, **15:** 1597–1605.

Ellis RJ, Deutsch R, Heaton RK, Marcotte TD, McCutchan JA, Nelson JA, Abrahamson I, Thal L, Atkinson JH, Walace MR, Grant I, the HNRC Group (1997). Neurocognitive impairment is an independent risk factor for death in HIV infection. *Arch Neurol* **54:** 416–424.

Everall IP, Luthert PJ, Lantos PL (1991). Neuronal loss in the frontal cortex in HIV infection. *Lancet* **337:** 1119–1121.

Grant I, Marcotte TD, Heaton RK, the HNRC Group (1999). Neurocognitive complications of HIV disease. *Psychol Sci* **10:** 191–195.

Halstead WC (1947). *Brain and intelligence.* Chicago: University of Chicago Press.

Heaton RK, Grant I, Nelson B, White DA, Kirson D, Atkinson HJ (1995). The HNRC500-neuropsychology of HIV infection at different disease stages. *J Int Neuropsychol Soc* **1:** 231–251.

Heaton RK, Velin RA, McCutchan JA, Gulevich SJ, Atkinson JH, Wallace MR, Godrey HPD, Kirson A, Grant I, the HNRC Group (1994). Neuropsychological impairment in human immunodeficiency virus-infection: implications for employment. *Psychosomat Med* **56:** 8–17.

Hinkin CH, David JH, Karen IM, Steven AC, Mona NL, Marta S, Bryan S (2002). Verbal and spatial working memory performance among HIV-infected adults. *J Int Neuropsychol Soc* **8:** 532–538.

Kieburtz K, Ketonen L, Cox C, Gossman H, Holloway R, Booth H, Hickey C, Feigin A, Caine ED (1996). Cognitive performance and regional brain volume in human immunodeficiency virus type 1 infection. *Arch Neurol* **53:** 155–158.

Kurosu T, Mukai T, Komoto S, Ibrahim MS, Li Y-G, Kobayasi T, Tsuji S, Ikuta K (2002). Human immunodeficiency virus type 1 subtype C exhibits higher transactivation activity of Tat than subtypes B and E. *Microbiol Immunol* **46:** 787–799.

Lezak MD (1995). *Neuropsychological assessment,* 3rd ed. New York: Oxford University Press.

Maj M, Satz P, Janssen R, Zudig M, Starace F, D'Elia LF, Sugondhbirom B, Mussa M, Naber D, Ndetei D, Schulte G, Sartorius N (1994). WHO neuropsychiatric AIDS study, cross sectional phase II: neuropsychological and neurological findings. *Arch Gen Psychiatry* **51:** 51–61.

Mayeux R, Stern Y, Tang M-X, Todak G, Sano J, Richards M, Stein Z, Erhardt A, Gorman J (1993). Mortality risks in gay men with human immunodeficiency virus infection and cognitive impairment. *Neurology* **43:** 176–182.

McArthur JC, Hoover DR, Becellar H, Millier EN, Cohen BA, Becker JT, Graham NMH, McArthur JH, Selnes OA,

Jacobson LP, Visscher BR, Concha M, Saah A (1993). Dementia in AIDS patients: incidence and risk factors. *Neurology* **42:** 1707–1712.

National AIDS Control Organization (1999). http://www.nacoonline.org/guidelines/guideline_8.pdf

Osmanov S, Pattou C, Walker N, Schwardlander B, Esparza J, WHO-UNAIDS Network for HIV Isolation and Characterization (2002). Estimated global distribution and regional spread of HIV-1 genetic subtypes in the year 2000. *J Acquir Immune Defic Syndr* **29:** 184–190.

Osowiecki DM, Cohen RA, Morrow KM, Paul RH, Carpenter CC, Flanigan T, Boland RJ (2000). Neurocognitive and psychological contributions to quality of life in HIV-1-infected women. *AIDS* **14:** 1327–1332.

Pereda M, Ayuso-Mateos JL, Barrio AGD, Echevarria S, Farinas MC, Palomo DG, Macias JG, Vazquez-Barquero JL (2000). Factors associated with neuropsychological performance in HIV-seropositive subjects without AIDS. *Psychol Med* **30:** 205–217.

Rao SL, Subbakrishna DK, Gopukumar K (2004). *NIMHANS Neuropsychology Battery—2004 manual.* India: NIMHANS Publication.

Reger M, Welsh R, Razani J, Martin DJ, Boone KB (2002). A meta-analysis of the neuropsychological sequelae of HIV infection. *J Int Neuropsychol Soc* **8**: 410–424.

Satishchandra P, Nalini A, Gourie-Devi M, Khanna N, Santosh V, Ravi V, Desai A, Chandramuki A, Jayakumar PN, Shankar SK. (2000). Profile of neurologic disorders associated with HIV/AIDS from Bangalore, South India (1989–96). *Ind J Med Res* **11:** 14–23.

Scaravilli F (1993). *Neuropathology of HIV infection.* London: Springer-Verlag London.

Schmidt M (1996). *Rey Auditory Verbal Learning Test: A handbook.* Los Angeles: Western Psychological Services.

Seth P (2006). Neuronal apoptosis and chemokine production by HIV-1 Tat protein derived from clade B and C in Human Fetal Neural Precursor cells. Abstract accepted for presentation at the 24th Annual Conference of the Indian Academy of Neurosciences, Lucknow, India, December, 2006.

Shallice T (1982). Specific impairments of planning. *Philos Trans R Soc Lond* **13:** 199–209.

Siddappa NB, Dash PK, Mahadevan A, Jayasuryan N, Hu F, Dice B, Keefe R, Satish KS, Satish B, Sreekanthan K, Chatterjee R, Venu K, Satishchandra P, Ravi V, Shankar SK, Shankarappa R, Ranga U (2004). Identification of subtype C human immunodeficiency virus type 1 by subtype specific PCR and its use in the characterization of viruses circulating in southern parts of India. *J Clin Microbiol* **42:** 2742–2751.

Siddappa NB, Venkataramanan M, Venkatesh P, Janaki MV, Jayasuryan N, Desai A, Ravi V, Ranga U (2006). Transactivation and signaling functions of Tat are not correlated: biological and immunological characterization of HIV-1 subtype-C Tat protein. *Retrovirology* **3:** 53.

Smith EE, Jonides J (1999). Storage and executive processes in the frontal lobes. *Science* **283:** 1657–1661.

Stern RA, Arruda J, Somerville JA, Cohen RA, Boland RJ, MD, Martin EM (1992). Neurobehavioral functioning in a nonconfounded group of asymptomatic HIV-seropositive homosexual men. *Am J Psychiatry* **149:** 1099–1102.

Stout JC, Salmon DP, Butters N, Taylor M, Peavy G, Heundel WC, Delis DC, Ryan L, Atkinson JH, Chandlier JL, Grant I, the HNRC group (1995). Decline in working memory associated with HIV infection. *Psychol Med* **25:** 1221–1232.

Tartar JL, Sheehan CM, Nash AJ, Starratt C, Puga A, Widmayer S (2004). ERPs differ from neurometric tests in assessing HIV-associated cognitive deficit. *Neuroreport* **15:** 1675–1678.

Wadia RS, Pujari SN, Kothari S, Udhar M, Kulkarni S, Bhagat S, Nanivadekar A (2001). Neurological manifestations of HIV disease. *J Assoc Physicians India* **49:** 343–348.

White DA, Heaton RK, Monsch AU, the HNRC Group (1995). Neuropsychological studies of asymptomatic human immunodeficiency virus-type 1 infected individuals. *J Int Neuropsychol Soc* **1:** 304–315.

Wilkie FL, Goodkin K, Eisdorfer C, Feaster D, Morgan R, Fletcher MA, Blaney N, Baum M, Szapocznik J (1998). Mild cognitive impairment and risk of mortality in HIV-1 infection. *J Neuropsychiatry Clin Neurosci* **10:** 125–132.

Yepthomi T, Paul R, Vallabhaneni S, Kumarasamy N, Tate DF, Solomon S, Flanigan T. (2006). Neurocognitive consequences of HIV in southern India: a preliminary study of clade C virus. *J Int Neuropsychol Soc* **12:** 424–430.

York MK, Franks JJ, Henry RR, Hamilton WJ (2001). Verbal working memory storage and processing deficits in HIV-1 asymptomatic and symptomatic individuals. *Psychol Med* **31:** 1279–1291.