SHORT-TERM EFFECTIVENESS AND SAFETY OF HAART IN THE FORM OF A GENERIC FIXED-DOSE COMBINATION OF STAVUDINE, LAMIVUDINE AND NEVIRAPINE (TRIVIRO) IN HIV-1-INFECTED ADULTS IN ZIMBABWE

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Objectives. To assess the effectiveness and safety of a twice-daily regimen of a generic fixed-dose combination (FDC) of stavudine, lamivudine and nevirapine (Triviro) in a cohort of Zimbabwean HIV-1-positive adults.

Design. A prospective, open-label, one-arm study of antiretroviral-naïve adults with CD4 counts <200 cells/µl. Fifty-three intention-to-treat (ITT) patients were enrolled and monitored for 4 months.

Setting. Three primary health care facilities in Zimbabwe.

Outcome measures. Efficacy criteria included plasma HIV-1 RNA load, CD4 counts, patient weight and Karnofsky performance scores. Toxicity was assessed by clinical evaluation and laboratory tests.

Results. There was a significant 3.0 $\log_{10}$ decrease in viral load at weeks 8 and 16 for both groups. Viral loads ≤400 copies/ml were achieved in 96% of per protocol (PP) and 85% of ITT patients at 8 and 16 weeks. At 4 months 85% of the PP group and 76% of the ITT group achieved undetectable viral loads. There was a significant increase in median CD4 counts of 101 cells/µl for PP and 86 cells/µl for the ITT analysis. The number of PP patients with Karnofsky scores of 100 improved from 10 (21%) to 38 (81%) and BMI increased by an average of 1.15 kg/m^2. Of the 134 adverse events recorded, 4 (3%) were severe. Of 16 adverse drug reactions in 10 patients, 13 were ascribed to nevirapine. One adverse reaction resulted in withdrawal from the study.

Conclusion. The effectiveness and safety of Triviro was comparable to that seen with other formulations, and our results support the use of this FDC in Zimbabwe and elsewhere.

The success of HIV/AIDS treatment depends on the effective and appropriate use of antiretroviral agents (ARVs). Resource-limited countries bear the brunt of this public health crisis, and their responses necessarily differ from those of industrialised nations. Ideally, ARV administration and patient monitoring in public programmes should be standardised, with free, effective, tolerable drugs and simple regimens to promote patient compliance.

Most developing countries employ the World Health Organization (WHO) public health approach to antiretroviral treatment, which aims to maximise patient survival using highly active antiretroviral therapy (HAART) with a combination of three agents from different drug classes in order to enhance efficacy and minimise the development of resistance through the promotion of adherence.[1]

Adherence to treatment programmes is of particular importance to prevent treatment failure and development of drug resistance, since patient monitoring and availability of alternative drugs is often limited in resource-limited countries. Patient compliance in these countries has been found to be at least as good as that in developed regions,[2] but obstacles to long-term adherence include medication cost and affordability in the health care setting, access to treatment, side-effects and the complexities of treatment schedules.

Companies producing generic drugs were the first to market fixed-dose combination (FDC) ARV preparations, and these drugs have won broad acceptance in the developing world. A study by Chien in 2007 showed that the vast majority of first-line ARV drugs in sub-Saharan Africa are supplied by generic companies.[3] This is in large part due to the fact that the average price of generic drugs is usually a fraction of that of brand-name equivalents. FDCs comprise a third of the drugs purchased in the region, with the stavudine/lamivudine/nevirapine (d4T/3TC/NVP) combination constituting the largest proportion at 20%. FDCs form an important pillar in promoting adherence, since they greatly reduce pill burden, dosing frequency and prescription errors. As with brand-name drugs, generic formulations are subject to stringent quality control standards. However, in order to ensure patient safety and public support, it is essential to ensure that counterfeit generic or brand name drugs do not find their way onto the market.

The Triviro (d4T/3TC/NVP) (Ranbaxy Laboratories, Centurion, South Africa) combination HAART is similar in formulation to Triomune (Cipla, Mumbai, India) and GPO-VIR (Thai Government Pharmaceutical Organisation). It has been shown to be comparable both in terms of bioequivalence[4] and safety[5] to each of its components.

While several studies have been conducted in Central and West Africa using d4T/3TC/NVP,[6,7] few have reported the use of this FDC in the southern African region. Zimbabwe has one of the highest HIV prevalence rates in southern Africa, which in 2005 was estimated at 33% in adults aged 15 - 49.[8]

This study was designed to evaluate the short-term effectiveness and safety of Triviro in an eligible cohort of HIV-1-positive patients in Zimbabwe. The goals were to assess virological, immunological and clinical improvement, tolerability and safety profile in treatment-naïve participants.

METHODS

This prospective, open-label, one-arm observational study was conducted at three primary health care facilities in Zimbabwe (Zimbabwean Swiss AIDS Care Foundation, the Colin Saunders Hospital and the Well Women's Clinic). Enrolment took place from May 2005 until February 2006.

The enrolment criteria for patients who qualified were: >18 years of age, HIV-1 positive at WHO stage I, II or III with CD4 counts <200 cells/µl or advanced WHO stage III disease presenting with recurrent or persistent oral thrush and/or recurrent invasive bacterial infections irrespective of CD4 count. Exclusion criteria were inability to provide informed consent, a history of hypersensitivity to treatment components, alcohol or drug abuse, active pulmonary tuberculosis, acute/active opportunistic infections, a history of previous ARV therapy, pregnancy or breastfeeding, female patients not using contraception, evidence of peripheral neuropathy, haemoglobin concentration <9.0 g/dl, platelet count <75 000×10^9/l, aspartate aminotransferase (AST) or alanine aminotransferase (ALT) ≥3 times the upper limit of normal (ULN), serum creatinine ≥1.6 times the ULN, serum amylase ≥1.6 times the ULN, total bilirubin ≥30 µmol/l, and malabsorption or severe chronic persistent diarrhoea.

Informed consent was obtained from all participants. The Medicines Control Authority of Zimbabwe approved the protocol and provided a provisional licence for the use of Triviro before the study was conducted.

Two fixed-dose tablet formulations were considered for use as study medication: Triviro-30 (150 mg 3TC, 30 mg d4T and 200 mg NVP) for patients with a body weight <60 kg and Triviro-40 (150 mg 3TC, 40 mg d4T and 200 mg NVP) for patients with a body weight >60 kg. This study was performed before revision of the WHO guidelines on stavudine dosing, where 30 mg stavudine is the recommended dose regardless of body weight.[9]

A 2-week nevirapine lead-in dose was used to assess tolerability of the formulation, especially nevirapine, and to decrease the risk of adverse reactions. During this period patients were provided with a single tablet of Triviro-30 or Triviro-40 for administration in the morning and a single tablet of Coviro LS 30 or 40, a stavudine/lamivudine nucleoside combination, in the evening that corresponded with the dose of stavudine in the Triviro formulation. Patients who experienced no significant drug-related adverse events were advanced to therapy with Triviro-30 or Triviro-40 twice daily after the initial 2-week treatment period.

Patients were excluded from the study if they were lost to follow-up (failure to visit clinic within a week of scheduled follow-up visit), or experienced serious intolerable adverse events or clinical deterioration indicative of viral resistance. Treatment of other ailments was continued unless contraindicated for concomitant use in combination with the study medication.

Patient monitoring was performed within 2 weeks of treatment initiation to establish drug tolerability. Thereafter follow-up assessments at 4 weeks, 8 weeks and 4 months were performed. CD4 counts were measured at baseline and 4 months. HIV-1 RNA viral load testing was performed at baseline, 8 weeks and 4 months. Karnofsky scoring was assessed at baseline and 4 months. Potential toxicity of the study medication was assessed by clinical examination and monitoring of levels of haemoglobin, white blood cells and platelets assessed at the screening visit, week 8 and month 4. Serum transaminases were measured at the screening visit, 2, 4 and 8 weeks and 4 months. Serum creatinine and amylase were measured at screening, week 2 and month 4. The Aids Clinical Trials Group (ACTG) rating was used to assess toxicity.[10]

LABORATORY ASSAYS

All HIV viral load and CD4 measurements were performed by the laboratory of Vermaak and Partners, Pretoria, South Africa. The Nuclisens EasyQ HIV-1 assay (bioMerieux, Marcy l'Etiole, France) was used for viral load measurements.

STATISTICS

Viral loads and CD4 cell counts were summarised using descriptive statistics. Changes in the medians of viral loads (using $\log_{10}$ transformation) and CD4 cell counts relative to baseline were tested for significance using the Wilcoxon signed rank test. Proportions of subjects with plasma HIV-1 RNA levels ≤25, <50 and <400 copies/ml were assessed using the binomial (exact) 95% confidence interval. Changes in blood pressure, body mass index (BMI), pulse rate, respiratory rate, temperature and haemoglobin were analysed using Student's paired *t*-test. Changes in median serum aspartate transaminase, alanine transaminase, bilirubin, serum creatinine, serum amylase, white cell counts, platelet counts and Karnofsy score were tested for significance using the Wilcoxon signed rank test. The proportion of patients who had a Karnofsky score of 100 at baseline and at month 4 was also summarised. Missing data were replaced with the last available observation for CD4 cell count and viral load data. Statistical analysis was performed using PC SAS 8.2 (SAS Institute Inc., Cary, NC) at 5% two-sided alpha levels.

RESULTS

Out of 55 patients screened, 53 were enrolled at three participating primary health care clinics in Zimbabwe. Of this intention-to-treat (ITT) group (i.e. patients who received at least 1 dose of drug), 6 patients withdrew or were excluded from the study due to: adverse events (2), non-compliance (2), loss to follow up (1) and failure to attend the clinic (1). One of the 2 patients who withdrew as a consequence of reported adverse events presented with immune reconstitution syndrome (tuberculosis) and died 2 weeks after withdrawal from the study, and the other presented with a severe rash thought to be caused by nevirapine. The demographics of the ITT group are shown in Table I.

TABLE I. DEMOGRAPHIC CHARACTERISTICS OF THE ITT GROUP AT BASELINE

Variable	Statistics
Age (yrs)	
N	53
Mean±SD	41.6±7.63
Min, max	28.6, 56.4
Sex (*N* (%))	
Female	27 (50.9%)
Male	26 (49.1%)
Race (*N* (%))	
Black	48 (90.6%)
Caucasian	2 (3.8%)
Mixed race	3 (5.7%)
Viral load	2 700
CD4	117

VIRAL AND IMMUNOLOGICAL RESPONSE

There was a significant decrease in HIV-1 RNA levels from the median (interquartile range (IQR)) at baseline of 4.4 $\log_{10}$ copies/ml to 1.4 $\log_{10}$ copies/ml (3.0 $\log_{10}$ decrease) at week 8, which was maintained at month 4, for both ITT and PP (i.e. patients who followed the study protocol strictly) groups (p<0.0001) (Table II). The proportion of patients with viral loads of ≤400 copies/ml was 96% in the PP group and 85% in the ITT group at both 8 and 16 weeks of treatment. These results are presented in Table III and Fig. 1. At 4 months, 87.2% of the PP group had viral RNA levels ≤50 copies/ml, with 85.1% achieving undetectable levels (≤25 copies/ml). Of the ITT group, 77.4% had viral loads ≤50 copies/ml and 75.5% ≤25 copies/ml (Table III).

TABLE II. VIRAL LOAD, $\log_{10}$ (HIV RNA COPIES/ml) FOR ITT AND PP POPULATIONS AT BASELINE, WEEK 8 AND MONTH 4

	Baseline		Week 8		Month 4	
Statistic	ITT	PP	ITT	PP	ITT	PP
N	53	47	53	46*	53	47
Median	4.431	4.3979	1.398	1.3979	1.398	1.3979
IQR (Q1 - Q3)	4.2041 - 4.9085	4.1139 - 4.8195	1.3979 - 1.9638	1.3979 - 1.6435	1.3979 - 1.3979	1.3979 - 1.3979

*Data for 1 subject not available.

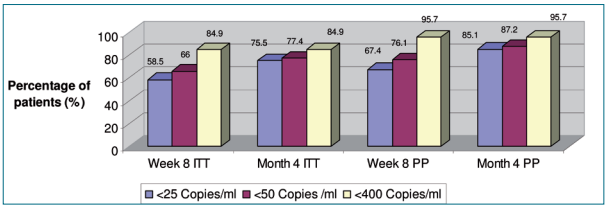

Fig. 1. Viral loads in the ITT and PP groups at week 8 and month 4.

A significant improvement in the CD4 cell counts from baseline to month 4 was observed, with an increase in the median count of 86 cells/µl for the ITT group and 101 cells/µl for the PP group (Fig. 2).

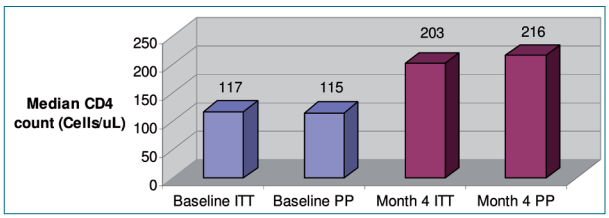

Fig. 2. CD4 counts for ITT and PP populations at baseline and month 4.

PHYSIOLOGICAL PARAMETERS

Participant body weight increased significantly after 4 months of treatment in the PP group, with a mean increase in weight of 3.22 kg, corresponding to a BMI increase of 1.15 kg/m^2. Respiratory rate decreased significantly from baseline (an average decrease of 1.3 breaths/min), while blood pressure, pulse rate and temperature remained essentially unchanged. The Karnofsky performance score at 100 was found to markedly improve in the PP group from 10 patients (21.3%) at baseline to 38 (80.9%) at the conclusion of the study.

TOLERABILITY

Laboratory analyses of markers to potentially indicate toxicity was performed. AST levels were shown to decrease 2 weeks after initiation of treatment, but approached baseline levels at later visits. ALT levels were elevated at month 4, and serum creatinine was increased at week 2. Amylase was shown to decrease at month 4, while significant declines in bilirubin were seen at all time points. While the median and IQR values for all the enzymatic parameters did not exceed grade 1 according to the ACTG guidelines,[10] there were instances where the maximum values were indicative of grade 2 or 3. A single patient had AST and ALT levels in the grade 2 (2.6 - 5.0 times ULN) range at screening, which decreased to below threshold during treatment. Another patient showed elevated levels of AST and ALT (grade 3; 5.1 - 10 times ULN) at week 8, which fell to normal at week 16. Serum creatinine levels were within the grade 1 (1.1 - 1.3 times ULN) range, except for 1 patient at grade 2 (1.4 - 1.8 times ULN) at 2 weeks, which normalised by 16 weeks. Analysis of amylase levels showed 4 patients at grade 2 (1.1 - 1.5 times ULN) and grade 3 (1.6 - 2.0 times ULN), respectively. This changed to 5 patients at grade 2 and 1 at grade 3 at 16 weeks. One patient had a grade 3 (2.6 - 5.0 times ULN) bilirubin level at week 4, which normalised by week 16. Although slightly elevated levels of haemoglobin and platelets were noted at 4 months, no changes were seen in the lymphocyte counts.

ADVERSE EVENTS AND DRUG REACTIONS

One hundred and thirty-four adverse events were recorded, the commonest being headache in 6 patients (11.3%), diarrhoea in 5 (9.4%), cough in 4 (7.5%), and loss of appetite and vomiting in 3 (5.7%). The majority (74.6%) of these events were judged to be mild, with 22.4% moderate and 3% severe. Eighty-one per cent of these events were considered to be unrelated to the study medication, 3.7% to have a remote possibility of causal relatedness, 11.9% a possible connection and 3% a probable connection. More than 1 adverse event was noted in 40 patients (76%). Sixteen adverse drug reactions were noted in 10 patients, of which 13 were ascribed to nevirapine (81.3%) and 3 (18.8%) to stavudine. The majority (56.3%) of these reactions were mild, 37.5% were moderate, and there was one instance (6.3%) of a severe rash that resulted in withdrawal from the study.

DISCUSSION

In this observational study of Triviro usage in Zimbabwe, we found effective viral suppression and immunological recovery at month 4 after initiation of generic FDC HAART in the majority of the participants. A significant virological improvement after 4 months was noted, with a 3.0 $\log_{10}$ decline in the median HIV-1 RNA levels from

TABLE III. NUMBER (%) OF ITT AND PP PATIENTS WITH VIRAL LOADS ≤25, ≤50 AND ≤400 COPIES/ml AT WEEK 8 AND MONTH 4

	Week 8		Month 4	
	ITT	PP	ITT	PP
≤25 copies/ml 95% CI	31 (58.5%) 44.1 - 71.9%	31 (67.4%) 52.0 - 80.5%	40 (75.5%) 61.7 - 6.2	40 (85.1%) 71.7 - 93.8%
p-value†	0.2164	0.0183*	0.0002**	<0.0001**
≤50 copies/ml 95% CI	35 (66.0%) 51.7 - 78.5%	35 (76.1%) 61.2 - 7.4%	41 (77.4%) 63.8 - 87.7%	41 (87.2%) 74.3 - 95.2%
p-value†	0.0195*	0.0004**	<0.0001**	<0.0001**
≤400 copies/ml 95% CI	45 (84.9%) 72.4 - 93.3%	44 (95.7%) 85.2 - 99.5%	45 (84.9%) 72.4 - 93.3%	45 (95.7%) 85.5 - 99.5%
p-value†	<0.0001**	<0.0001**	<0.0001**	<0.0001**

*Significant at 5% level, **Significant at 1% level.
†Two-sided binomial test (H_0: proportion = 0.5).

baseline. Similarly, a significant immunological improvement was seen, with an average increase of 101 CD4 cells/µl for the PP-treated group. These results compare favourably with, and in some cases exceed, the endpoints of similar studies.[6,7] Patient well-being was also found to be enhanced, with a marked increase in BMI and in the number of patients with a Karnofsky score of 100 by the end of 4 months. The majority of adverse events were not judged to be severe, although there was one withdrawal due to debilitating rash. The adverse drug reactions observed were also typical for the drugs used, and are mentioned in the product information.

The d4T/3TC/NVP FDC used in this study has previously been used as HAART therapy for advanced AIDS care[11] and is increasingly being used for paediatric treatment, with oral formulations facilitating easier dosage and increased compliance.[12] Although adult trials have been performed using d4T/3TC/NVP formulations in other regions, few studies have been performed in southern Africa. The reports to date are on the use of the combination for the treatment of AIDS-associated Kaposi's sarcoma in KwaZulu-Natal, South Africa[13] and for prevention of vertical transmission of HIV-1 in Mozambique,[14] and an initial report of its use in adults in South Africa.[15]

The present study has some limitations. A longer-term study is needed to establish toxicity in patients using what could be lifelong treatment, especially given reports from Thailand of metabolic complications such as dyslipidaemia.[16] Testing for resistance was not performed in our study, but will become critical in light of cross-resistance that has been described in second-line treatment-naïve patients failing the d4T/3TC/NVP treatment.[17]

The positive longer-term experiences of other sites with Triomune[18,19] and Triviro[20] suggest that use of these generic HAART FDCs in southern Africa could be considered as first-line therapy.

In conclusion, our short-term results support the use of Triviro, and d4T/3TC/NVP FDCs in general, to treat HIV-1 positive adults.

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REFERENCES

1. Gilks CF, Crowley S, Ekpini R, *et al.* WHO public-health approach to antiretroviral treatment against HIV in resource-limited settings. *Lancet* 2006; 368: 505-510.
2. Mills EJ, Nachega JB, Buchan I, *et al.* Adherence to antiretroviral therapy in sub-Saharan Africa and North America. *JAMA* 2006 296: 679-690.
3. Chien CV. HIV/AIDS drugs for sub-Saharan Africa: how do brand and generic supply compare? *PLoS ONE* 2007; 2: e278.
4. Monif T, Tippabhotla SK, Garg M, Singla AK, Vijan T. Nevirapine/lamivudine/stavudine as a combined-formulation tablet: bioequivalence study compared with each component administered concurrently under fasting condition. *Int J Clin Pharmacol Ther* 2006; 44: 276-283.
5. Marier JF, Dimarco M, Guilbaud R,*et al.* Pharmacokinetics of lamivudine, zidovudine, and nevirapine administered as a fixed-dose combination formulation versus coadministration of the individual products. *J Clin Pharmacol* 2007; 47: 1381-1389.
6. Laurent C, Kouanfack C, Koulla-Shiro S, *et al.*Effectiveness and safety of a generic fixed-dose combination of nevirapine, stavudine, and lamivudine in HIV-1-infected adults in Cameroon: open-label multicentre trial. *Lancet* 2004; 364: 29-34.
7. Jarrousse B, Ba M, Dakouo ML, *et al.* Fixed-dose combination of stavudine 30 mg/lamivudine/nevirapine as standard first-line regimen in a sub-Saharan cohort of HIV-1 infected adult patients. XVI International AIDS Conference, Toronto, Canada, 13 - 18 August 2006, abstract CDB0497.
8. Zimbabwe Human Development Report 2003, Poverty Reduction Forum, Institute of Development Studies, University of Zimbabwe. http://hdr.undp.org/en/reports/nationalreports/africa/zimbabwe/zimbabwe_2003_en.pdf (accessed 25 November 2007).
9. Addendum to 2006 Who Guidelines On Antiretroviral Therapy for HIV Infection in Adults and Adolescents: New Dosage Recommendations for Stavudine (D4T). http://www.who.int/hiv/art/ARTadultsaddendum.pdf (accessed 20 March 2008).
10. Division of Aids Table for Grading the Severity of Adult and Pediatric Adverse Events. Published December 2004. www.aactg.org/textmenu//science_sciguidelines.asp
11. Manosuthi W, Chimsuntorn S, Likanonsakul S, Sungkanuparph S. Safety and efficacy of a generic fixed-dose combination of stavudine, lamivudine and nevirapine antiretroviral therapy between HIV-infected patients with baseline CD4 <50 versus CD4 > or = 50 cells/mm3. *AIDS Res Ther* 2007; 4: 6.
12. Pensi T. Fixed dose combination of lamivudine, stavudine and nevirapine in the treatment of pediatric HIV infection: a preliminary report. *Indian Pediatr* 2007; 44: 519-521.
13. Mosam A, Cassol E, Page T, *et al.* Generic antiretroviral efficacy in AIDS-associated Kaposi's sarcoma in sub-Saharan Africa. *AIDS* 2005; 19: 441-443.
14. Marazzi MC, Germano P, Liotta G, *et al.* Safety of nevirapine-containing antiretroviral triple therapy regimens to prevent vertical transmission in an African cohort of HIV-1-infected pregnant women. *HIV Med* 2006; 7: 338-344.
15. Mosam A, Coovadia H, Sharon C, *et al.* Generic HAART in South Africa. Efficacy of fixed dose nevirapine, stavudine and lamivudine. XVI International AIDS Conference, Toronto, Canada, 13 - 18 August 2006, abstract CDB0598.
16. Manosuthi W, Chaovavanich A, Prasithsirikul W, *et al.* Efficacy and metabolic complications after 96 weeks of a generic fixed-dose combination of stavudine, lamivudine and nevirapine among advanced HIV-infected patients. IAS Conference, 22-25 July, Sydney, Australia, abstract WEPEB057.
17. Sungkanuparph S, Manosuthi W, Kiertiburanakul S, Chantratitra W. Tenofovir resistance among HIV-infected patients failing a fixed-dose combination of stavudine, lamivudine, and nevirapine in a resource-limited setting. *AIDS Patient Care STDS* 2007; 21: 711-714.
18. Laurent C, Kouanfack C, Koulla-Shiro S, *et al.* Long-term safety, effectiveness and quality of a generic fixed-dose combination of nevirapine, stavudine and lamivudine. *AIDS* 2007; 21: 768-771.
19. Calmy A, Pinoges L, Szumilin E, *et al.* Generic fixed-dose combination antiretroviral treatment in resource-poor settings: multicentric observational cohort. *AIDS* 2006; 20: 1163-1169.
20. Pujari SN, Patel AK, Naik E, *et al.* Effectiveness of generic fixed-dose combinations of highly active antiretroviral therapy for treatment of HIV infection in India. *J Acquir Immune Defic Syndr* 2004; 15: 1566-1569.