A randomized double-blind trial of the addition of lamivudine or matching placebo to current nucleoside analogue reverse transcriptase inhibitor therapy in HIV-infected children: the PENTA-4 trial

Paediatric European Network for Treatment of AIDS*

Objectives: To evaluate the toxicity, tolerability and effect on laboratory markers of adding lamivudine (3TC) to nucleoside analogue reverse transcriptase inhibitors (NRTI) in children with HIV-1 infection.

Design: Randomized double-blind trial.

Methods: HIV-1-infected children on stable NRTI therapy were randomized to receive 3TC syrup or tablets (4 mg/kg twice daily) or matching placebo in addition to existing therapy. Endpoints were serious adverse events, and changes in CD4 cell count and plasma HIV-1 RNA. Analyses were on an intention-to-treat basis.

Results: A total of 162 (81 on 3TC, 81 on placebo) children [median age, 6.5 years; interquartile range (IQR), 4.1–10.1 years] were included. At randomization, 52 were receiving zidovudine (ZDV), 39 didanosine (ddI), 54 ZDV–ddI and 17 ZDV–zalcitabine (ddC); 32 (20%) had AIDS; median CD4 cell count was 328×10^6/l (IQR, 127–696×10^6/l), and median HIV-1 RNA was 4.9 $\log_{10}$ copies/ml (IQR, 4.3–5.4 $\log_{10}$ copies/ml). Median follow-up was 40 weeks (IQR, 29–49 weeks) and 76% of follow-up was on blinded therapy for both 3TC and placebo groups. There were 11 serious adverse events in the blinded phase [two clinical (both placebo) and nine laboratory (five 3TC, four placebo)], five (two 3TC, three placebo) resulting in stopping trial drug. At 24 weeks, the CD4 cell count was greater in the 3TC group by a median of 47×10^6/l and HIV-1 RNA was lower by 0.30 $\log_{10}$ copies/ml ($P = 0.03$ and 0.002, respectively, versus the placebo group). The difference in reduction in HIV-1 RNA up to 24 weeks, as measured by area under the curve minus baseline, between 3TC and placebo groups was 0.38 $\log_{10}$ copies/ml (95% confidence interval, 0.12–0.65) greater in children taking ZDV-containing regimens at baseline, compared with those on ddI monotherapy ($P = 0.005$), after adjusting for other factors at baseline. Thirteen children developed new AIDS events (six on 3TC, four on placebo) of whom three died (all placebo).

Conclusions: The addition of 3TC to current NRTI therapy in children was safe and well-tolerated. There was evidence that treatment changes in HIV-1 RNA viral load were greater in children taking regimens that included ZDV.

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From the Paediatric European Network for Treatment of AIDS (PENTA). *See Appendix for committees and collaborators.

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Introduction

The use of combination antiretroviral therapy with two or more drugs in HIV-infected children increased following reports of improved efficacy over monotherapy [1,2]. However, licensing of drugs in children lags behind that in adults, often because there are insufficient pharmacokinetic, toxicity and tolerability data on paediatric formulations available from paediatric trials. Of the 10 antiretroviral therapies licensed for use in adults in 1998, only four have been licensed for children in most European countries.

At the time the Paediatric European Network for Treatment of AIDS (PENTA)-4 trial was set up, availability of formulations of antiretroviral drugs for HIV-infected children varied across Europe. Zidovudine (ZDV) was licensed in all countries and available as a syrup. Didanosine (ddI) was licensed in most countries but availability of the paediatric formulation varied. Zalcitabine (ddC) was available only in tablet form, although the syrup became available during the course of the trial through a named patient programme. No other drugs were available and thus many children were on ZDV monotherapy.

In Phase II trials in adults with CD4 cell counts over 100×10^6/l, combination therapy with lamivudine (3TC) and ZDV was reported to result in increases in the CD4 cell count and decreases in HIV-1 RNA viral load similar to other nucleoside combinations and sustained for up to 48 weeks [3–6]. The most common adverse events attributed to 3TC were neutropenia, gastrointestinal disturbances and headaches. Phase III trials in adults were ongoing at the time this trial commenced. Preliminary data from an open Phase I/II study of 3TC syrup in children showed evidence of antiretroviral activity and good tolerability [7]. However, seven out of 89 children in follow-up after this study and a further five out of 13 who received 3TC through a compassionate release programme developed pancreatitis (total 11.8%). Because all children had advanced disease and were also receiving other concurrent antiretroviral therapy, it was unclear whether this was 3TC-related. In previously untreated symptomatic children, a Phase III trial (AIDS Clinical Trials Group 300) designed to compare the efficacy and toxicity of ZDV–3TC with ddI–ZDV and ddI monotherapy had just commenced in the United States. The PENTA-4 trial was designed to assess the safety, tolerability and effect on immunological and virological parameters of adding 3TC in children who had received prior antiretroviral therapy with nucleoside analogue reverse transcriptase inhibitors (NRTI).

Participants and methods

Participants

Children were eligible if they were aged between 3 months and 16 years, and were HIV-infected through mother-to-child transmission or via infected blood or blood products. Children at any disease stage (Centers for Disease Control and Prevention classification N, A, B or C [8]) and with any duration of previous NRTI (but not protease inhibitor or non-NRTI) therapy could be enrolled provided they had been on their current antiretroviral therapy for at least 3 months. Criteria for exclusion included concurrent cytotoxic therapy for a malignancy, acute therapy for an opportunistic infection (enrolment could take place after acute treatment), history of pancreatitis, hepatitis B infection with decompensated liver disease, haemoglobin less than 9 g/dl, neutrophil count less than 1×10^9/l, bilirubin greater than 3 times the upper limit of normal (ULN), aspartate or alanine transaminases greater than 5× ULN, creatinine greater than 135 μmol/l, and amylase or lipase greater than 2× ULN (unless pancreatic amylase was less than 2× ULN). The protocol was approved by the relevant ethics committee for each participating centre. All parents or primary caregivers, and children where appropriate, gave written consent to participate.

Trial design and treatment

PENTA-4 was a randomized, double-blind trial. The planned intake was 150 children to be enrolled over 12 months and followed for a minimum of 24 weeks. Children were randomly allocated to receive 3TC 4 mg/kg twice daily as a syrup (10 mg/ml) or tablets (150 mg), or matching placebo in a 1 : 1 ratio. Randomization was stratified by country and a minimization algorithm was employed to achieve a balance amongst the groups for current antiretroviral therapy (ZDV, ddI, ZDV–ddI, or ZDV–ddC). Randomization was undertaken centrally by the National Trials Centres in London (for United Kingdom, Germany, Ireland, Italy, Brazil) and Paris (for France, Belgium, Portugal, Spain, Switzerland) by telephone or fax. Because there were a large number of centres, each with small numbers of children, trial drugs were distributed individually for each child to be started 2 weeks after randomization. The median time between randomization and trial drug prescription was 15 days [interquartile range (IQR), 14–21 days].

The protocol allowed children to start open-label 3TC during the trial if there was progression of HIV disease, if considered appropriate by paediatricians and caregivers, who remained blind to the original randomization. An amendment in April 1996 allowed children entering the trial on ZDV monotherapy to add or switch to ddI during the trial while remaining on blinded trial therapy. A further amendment in September 1996 allowed children to switch to open-label 3TC after 24 weeks even if there was no evidence of disease progression, after preliminary results from the CAESAR trial of 3TC in adults became available

(unpublished report from Glaxo-Wellcome, Greenford, Middlesex, UK). Concomitant medication was allowed, including primary prophylaxis for *Pneumocystis carinii* pneumonia (PCP) [9] or bacterial infections.

Management and follow-up

Children were assessed at randomization (week −2), at the date of start of trial drug (week 0), then at 2 and 4 weeks, then every 4 weeks to week 24, and every 8 weeks thereafter (to a maximum of 72 weeks). At each visit there was a clinical assessment including measurement of height, weight and, for children under 2 years, head circumference. Full blood count, creatinine (or urea), electrolytes, alanine or aspartate aminotransferase, alkaline phosphatase, amylase, and CD4 cell count were measured locally in the clinical centres. If the amylase was elevated, it was fractionated to determine whether it was of salivary or pancreatic origin. Blood samples for plasma HIV-1 RNA determination were taken at each visit into an EDTA tube, separated within 6 h, and the plasma stored at −70°C. Parents were asked at each visit whether any doses of trial drug or other antiretroviral therapy had been missed. Serious adverse event reports were faxed to the trials centres within 24 h. The Data and Safety Monitoring Committee reviewed the unblinded data in May 1996 and March 1997. All eligibility and baseline data, serious adverse events, clinical events (AIDS or death) and 10% of clinical and laboratory data collected were validated against the original clinic records. Clinical status at entry, serious adverse events and AIDS events were reviewed by the trial coordinators based at the trials centres, who were blind to the allocated treatment.

Endpoints

The primary endpoint of the trial was the development of serious clinical or laboratory events considered to be probably or possibly attributable to trial medication. Serious adverse events were defined as either fatal, life-threatening, causing or prolonging hospital admission, permanent disability, cancer, clinical pancreatitis, raised pancreatic amylase (grade 2) or other clinical or laboratory (grade 3 or 4) events, categorized according to a paediatric modification of the National Cancer Common Toxicity Criteria. All adverse events were classified according to whether they resulted in permanent discontinuation, interruption or other modification of trial drug, and whether they were probably, possibly or unlikely to be related to trial drug. Secondary endpoints were changes in HIV-1 RNA viral load, CD4 cell counts, and age-adjusted height and weight. Progression to AIDS or death was recorded.

Laboratory measurements

CD4 cell percentages and absolute cell counts were measured using flow cytometry in the clinical centres. HIV-1 RNA was measured in 200 μl plasma using the Roche Amplicor Monitor assay with add-in primers (version 1.5; Roche Diagnostic Systems, Branchburg, New Jersey, USA) in a central laboratory (Covance, Harrogate, Yorkshire, UK), using a single batch of kits. This laboratory was accredited and run according to good laboratory practice. The lower limit of detection was 400 copies/ml (2.6 $\log_{10}$ copies/ml).

Statistical analysis

The frequency of adverse events was compared between the two randomized groups using χ^2 tests. The analyses were undertaken for events occurring on blinded trial therapy or within 30 days after stopping blinded therapy, and for all events, by treatment group. Time to first serious event was compared using Kaplan–Meier plots and log-rank tests. CD4 cell counts were expressed as absolute counts and percentages. Counts were also adjusted for age by calculating CD4 cell z-scores, based on data from uninfected children born to HIV-infected mothers in the European Collaborative Study [10]. HIV-1 RNA levels were expressed as $\log_{10}$ copies/ml.

Laboratory and clinical responses were analysed on an intention-to-treat basis, including all children who were prescribed trial therapy at week 0, irrespective of whether they started or subsequently discontinued therapy. The median changes from baseline of CD4 cell counts, CD4 cell z-scores, HIV-1 RNA viral load, height, weight, height and weight adjusted for age [11] and head circumference in children younger than 2 years, were compared between the 3TC and placebo groups using Wilcoxon rank-sum tests at fixed assessment times and for the mean change as measured by the area under the curve minus baseline (AUCMB) up to week 24 and to week 48. Two-sided tests of significance were used.

Results

Between 6 December 1995 and 28 November 1996, 172 children were enrolled from 34 paediatric centres in Belgium (n = 6), France (n = 10), Germany (n = 1), Ireland (n = 6), Italy (n = 68), Portugal (n = 5), Spain (n = 42), Switzerland (n = 14), United Kingdom (n = 10) and Brazil (n = 10) participating in PENTA (Fig. 1). Ten children were not prescribed trial drug and were excluded from all analyses. Of these, four had been randomized although full consent had not been given, one withdrew consent, one did not return to clinic after randomization, one developed AIDS between weeks −2 and 0, and three had laboratory abnormalities at week −2 and the paediatrician decided not to prescribe trial drug. Twenty-one children had minor violations of eligibility criteria for entry in the trial, including grade 2 haematological abnormalities (n = 10), transaminases 5–10× ULN (grade 3; n = 4),

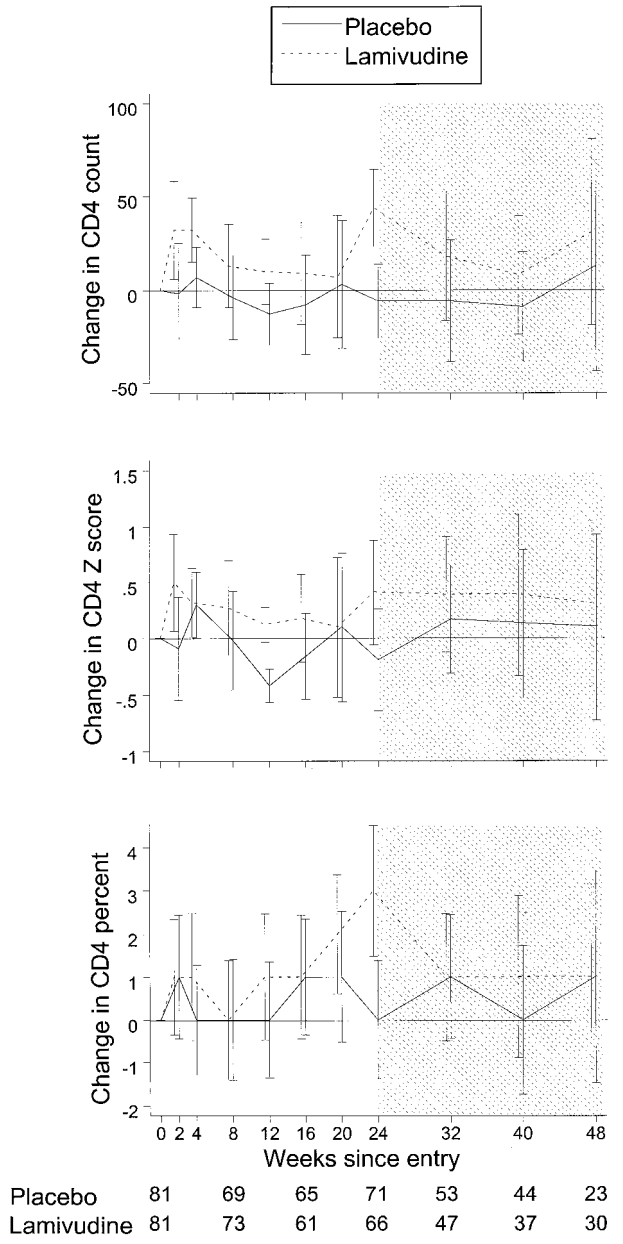

Fig. 1. Median (95% confidence interval) change in CD4 cell count, CD4 cell z-score and CD4 cell percentage from baseline. The shaded area indicates that children (on placebo) were switching to open-label lamivudine.

low NRTI doses (n = 6), or less than 3 months on stable NRTI therapy (n = 1). These children were included in the analysis.

Baseline characteristics

One hundred and fifty two (94%) children were infected by mother-to-child transmission, seven (4%) from infected blood products, and in three (2%) the route of infection was uncertain. The treatment groups were well balanced for these and other baseline characteristics, including duration of previous therapy and current NRTI therapy at baseline (Table 1).

Table 1. Baseline characteristics.

	Lamivudine (n = 81)	Placebo (n = 81)
Age (years)		
≤ 2	7	6
2–4	13	12
4–6	22	15
6–10	20	26
≥ 10	19	22
Sex		
Male	31	39
Female	50	42
Ethnicity		
White	68	68
Black African	10	10
Other	3	3
AIDS	14	18
Median (IQR) CD4 cell count ($\times 10^6$/l)	350 (151–696)	299 (126–694)
Median (IQR) CD4 percentage (%)	17 (9–24)	15 (7–28)
CD4 z-score		
≤ −3	47	49
−3 to −2	10	9
−2 to 0	22	19
≥ 0	1	2
Median (IQR) HIV-1 RNA ($\log_{10}$ copies/ml)	4.9 (4.3–5.3)	4.9 (4.4–5.5)
NRTI therapy at baseline		
ZDV	24	28
ddI	19	20
ZDV–ddI	26	28
ZDV–ddC	7	10
Median (IQR) months on prior NRTI	32 (12–50)	30 (14–52)

Data are numbers of patients, unless otherwise indicated. IQR, Interquartile range; ZDV, zidovudine; ddI, didanosine; ddC, zalcitabine; NRTI, nucleoside analogue reverse transcriptase inhibitors.

Follow-up and time on allocated treatment

Total follow-up to the data freeze in June 1997 was 1550 child-months with a median follow-up of 40 weeks (IQR, 29–49 weeks). One child did not return after the week 0 assessment. All other children were seen within 8 weeks of the data freeze date. A total of 147 children (73 on 3TC, 74 on placebo) were still on blinded therapy at week 24, but by week 48 all but 16 (eight on 3TC, eight on placebo) had switched to open-label 3TC. A total of 597 child-months in the placebo group and 577 child-months in the 3TC group were spent on blinded trial therapy equivalent to 75.6 and 76.0% of the total time in the trial, respectively. Among 52 children on ZDV monotherapy at randomization, one (2%) switched to ddI and two (4%) added ddI to ZDV monotherapy while remaining on blinded trial drug.

Adverse events

A total of 23 serious adverse events (12 3TC, 11 placebo) were reported in 18 children (nine 3TC, nine placebo) on blinded treatment (Table 2). Eleven events (five events in four children on 3TC, and six in five children on placebo) were considered to be probably or possibly related to trial therapy and all resulted in inter-

Table 2. Adverse events.

	No. events (no. children)					
	Serious adverse events on blinded therapy		All adverse events leading to discontinuation of blinded trial therapy		All serious adverse events throughout follow-up	
	Lamivudine (n = 81)	Placebo (n = 81)	Lamivudine (n = 81)	Placebo (n = 81)	Lamivudine (n = 81)	Placebo (n = 81)
Clinical						
Rash	–	1 (1)	–	1 (1)	–	1 (1)
Cardiomyopathy	–	1 (1)	–	1 (1)	1 (1)	1 (1)
Vomiting and diarrhoea	1 (1)	–	–	–	1 (1)	–
Nausea*	–	–	1 (1)	–	–	–
Pancreatitis	–	–	–	–	1 (1)	–
Convulsions	–	–	–	–	–	1 (1)
Haematuria	–	–	–	–	1 (1)	–
Laboratory						
Neutropenia	5 (4)	4 (2)	–	–	5 (4)	6 (4)
Leukopenia	1 (1)	–	–	–	2 (2)	1 (1)
Anaemia	1 (1)	1 (1)	–	–	1 (1)	3 (3)
Thrombocytopenia	–	–	–	–	–	1 (1)
Autoimmune haemolytic anaemia	–	–	–	–	1 (1)	–
Raised transaminases	2 (2)	3 (3)	–	1 (1)	2 (2)	3 (3)
Raised alkaline phosphatase	1 (1)	1 (1)	1 (1)	–	1 (1)	1 (1)
Raised amylase	1 (1)	–	–	–	1 (1)	–
Total	12 (9)	11 (9)	2 (2)	3 (3)	17 (13)	18 (9)

*Classified as a minor adverse event.

ruption or modification of the dose. Of these, two children had clinical events (cardiomyopathy and eczematous rash, both in children on placebo), six (three 3TC, three placebo) had neutropenia below 0.75×10^9/l (grade 3) or below 0.5×10^9/l (grade 4), two (one 3TC, one placebo) had transaminase levels more than 5× ULN (grade 3), and one (on 3TC) had alkaline phosphatase more than 5× ULN. One child (on 3TC) had amylase 2–5× ULN (grade 3), which resolved without modification of trial drug. Only five children (two 3TC, three placebo) stopped trial drug during the blinded phase, four for serious (one 3TC, three placebo) and one (on 3TC) for minor adverse events (nausea; Table 2). However, four children (two 3TC, two placebo) subsequently took open-label 3TC.

A total of 35 serious adverse events occurred on blinded and open-label 3TC combined (17 events in 13 children randomized to 3TC and 18 in nine children on placebo; Table 2). One child developed a first episode of pancreatitis 15 months after randomization to 3TC, and while on open-label 3TC, ZDV, indinavir and ganciclovir. The child had an AIDS diagnosis prior to entering the trial and developed disseminated *Mycobacterium avium-intracellulare* infection at the same time as pancreatitis. The child received no ddI during the trial. Pancreatic amylase was normal until the onset of clinical pancreatitis. Only one other child stopped 3TC while on open-label therapy after having a convulsion.

There was no evidence of a difference in the time to first serious adverse event between the 3TC and placebo groups (log-rank test at 24 weeks, $P = 0.56$). There were no significant differences in the adverse event rates by age, disease stage or NRTI therapy at trial entry.

Minor adverse events

A total of 109 minor adverse events were reported on blinded trial therapy (42 in 29 children on 3TC and 67 in 42 children on placebo). Of these, 19 (nine 3TC, 10 placebo) events in 15 children were considered possibly or probably related to trial therapy. Eleven minor adverse events (six 3TC, five placebo) in seven children resulted in temporary interruption or modification of trial drug dose. Most minor adverse events were mild clinical symptoms including gastrointestinal symptoms (three 3TC, three placebo), headache (two placebo), and rash (one 3TC, three placebo). Only one child with nausea on 3TC stopped trial therapy permanently.

Tolerability and compliance

Both 3TC and placebo appeared to be well-tolerated. A total of 159 (98%) children were prescribed trial drug as a syrup and three received tablets. During the trial, 37 (14 3TC, 23 placebo) children (23% of those on syrup) switched from syrup to tablets, the majority at the time of starting open-label 3TC, and subsequently tolerated the tablets well.

During the blinded phase, interruptions or discontinuations because of difficulty taking the medication, illness, or the parents' wishes were reported in six children (0.055 interrupted days per child-week) in the 3TC group and in nine children (0.013 interrupted days per

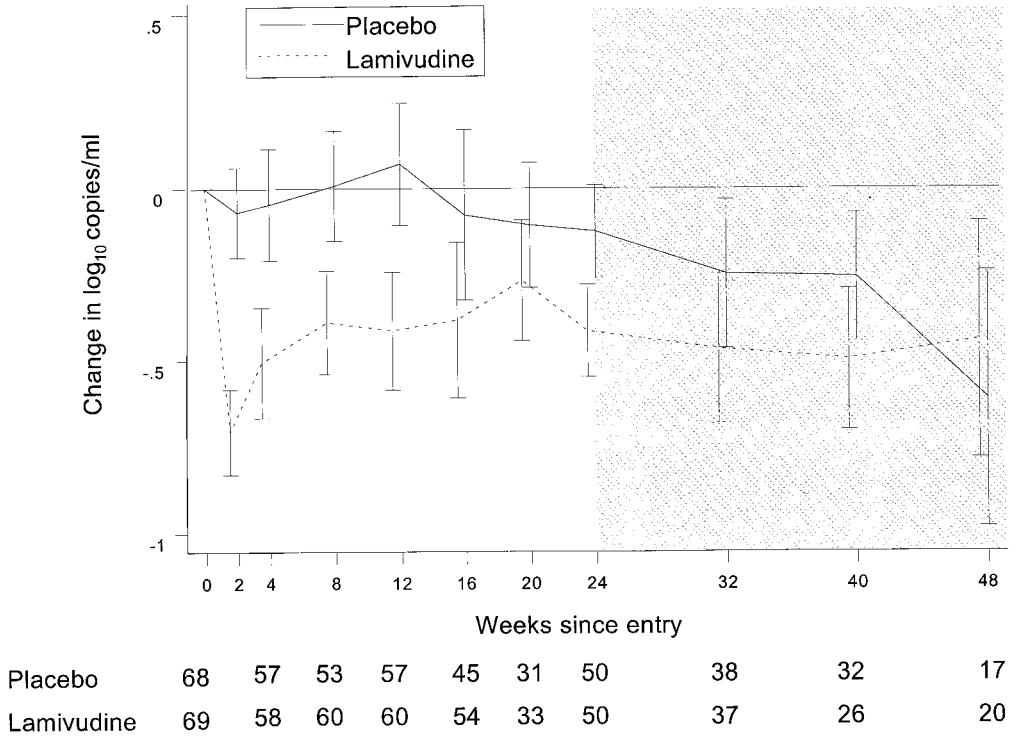

Fig. 2. Median (95% confidence interval) change in HIV-1 RNA viral load (log_{10} copies/ml) from baseline. The shaded area indicates that children (on placebo) were switching to open-label lamivudine.

child-week) in the placebo group. Interruptions or discontinuations because of serious or minor adverse events were reported in 10 children (0.09 interrupted days per child-week) in the 3TC group and in eight children (0.07 interrupted days per child-week) in the placebo group.

Data on mean corpuscular volume (MCV) of red blood cells were collected in all children to provide some information on compliance. In children on ZDV-containing regimens, median MCV was 93 fl (IQR, 87–100 fl) at baseline, 94 fl (IQR, 89–102 fl) at 24 weeks, and 95 fl (IQR, 89–102 fl) at 48 weeks, compared with median MCV of 76, 75 and 77 fl at 0, 24 and 48 weeks, respectively, in children on ddI monotherapy.

Changes in CD4 cell count

At 24 weeks, the median increase in CD4 lymphocyte count was 41×10^6/l (IQR, −55 to $+110 \times 10^6$/l) in the 3TC group compared with a decrease of 6×10^6/l (IQR, −91 to $+40 \times 10^6$/l) in the placebo group, a difference of 47×10^6/l ($P = 0.03$). By 48 weeks the difference was no longer significant but the majority had switched to open-label 3TC and the numbers were smaller (Fig. 1; Table 3). The changes from baseline to 24 weeks for CD4 cell percentage and z-score were also significantly higher in the 3TC group than in the

Table 3. Median change from baseline to 24 weeks for immunological, virological and clinical parameters.

	Median (IQR) change from baseline to 24 weeks			Median (IQR) AUCMB (0–24 weeks)		
	Lamivudine (n = 81)	Placebo (n = 81)	*P*	Lamivudine (n = 81)	Placebo (n = 81)	*P*
CD4 cells ($\times 10^6$/l)	41 (–55 to 110)	–6 (–91 to 40)	0.03	11.58 (–25.04 to 85.03)	–2.19 (–50.31 to 26.16)	0.02
CD4 percentage (%)	3 (–1 to 6)	0 (–2 to 3.5)	0.03	0.90 (–0.54 to 2.51)	0.29 (–1.88 to 1.99)	0.05
CD4 z-score	0.42 (–0.8 to 1.44)	–0.19 (–1.37 to 0.46)	0.01	0.22 (–0.52 to 1.03)	–0.21 (–0.72 to 0.27)	0.01
HIV RNA (log_{10} copies/ml)	–0.42 (–0.8 to –0.03)	–0.12 (–0.30 to 0.14)	0.002	–0.39 (0.23 to 0.16)	–0.11 (–0.71 to 0.15)	< 0.0001
Height (cm)	3.0 (2.0–4.0)	2.5 (1.5–3.2)	0.15	1.59 (0.80–2.30)	1.09 (0.57–1.74)	0.03
Height for age z-score	0.03 (–0.20 to 0.23)	–0.05 (–0.23 to 0.12)	0.30	0.04 (–0.11 to 0.13)	–0.03 (–0.15 to 0.06)	0.04
Weight (kg)	1.4 (0.7–2.0)	1.0 (0.5–1.8)	0.17	0.61 (0.16–1.03)	0.43 (0–0.92)	0.12
Weight for age z-score	0.02 (–0.17 to 0.23)	–0.06 (–0.20 to 0.14)	0.10	0.01 (–0.12)	–0.03 (–0.18 to 0.09)	0.12

IQR, Interquartile range; AUCMB, area under the curve minus baseline.

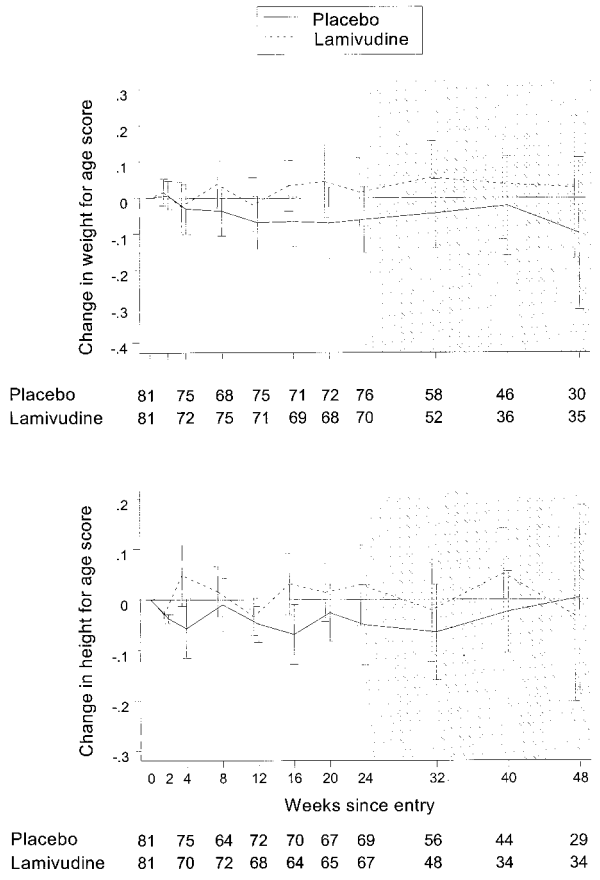

Fig. 3. Median (95% confidence interval) change in height for age and weight for age from baseline. The shaded area indicates that children (on placebo) were switching to open-label lamivudine.

placebo group. An on-treatment analysis (i.e., restricted to the period when each child was on allocated therapy) showed very similar results (data not shown).

Viral load measured by HIV-1 RNA

There were minimal changes in HIV-1 RNA in children allocated to placebo up to 24 weeks. In the 3TC group, the maximum decrease in median HIV-1 RNA was 0.71 $\log_{10}$ copies/ml at week 2, and by week 24, the median decrease was 0.42 $\log_{10}$ copies/ml (Table 3; Fig. 2), 0.30 $\log_{10}$ copies/ml greater than in the placebo group ($P = 0.002$). Mean reductions in HIV-1 RNA from 0 to 24 weeks and 0 to 48 weeks, as measured by AUCMB, were significantly greater in the 3TC group ($P < 0.0001$). Results to week 24 on allocated treatment were similar (data not shown).

No child had a viral load below 400 copies/ml at baseline. Subsequently, 23 samples from nine children on 3TC were reported to be below the limit of detection mostly in the first 12 weeks, but in only four (6% of children) at 24 weeks. Five children randomized to placebo had 11 samples with undetectable viral load; three had very low values at baseline, and in two children, decreases occurred after switching to open-label 3TC.

Compared with children on ZDV monotherapy, the difference in HIV-1 RNA reduction between 3TC and placebo was similar in children on ZDV–ddI or ZDV–ddC, but significantly less in those on ddI monotherapy. In an analysis combining children on ZDV-containing regimens at baseline, and after adjusting for AIDS, time on previous therapy, CD4 z-score, and HIV-1 RNA at baseline, the difference in reduction in HIV-1 RNA up to 24 weeks, as measured by AUCMB, between 3TC and placebo was 0.38 $\log_{10}$ copies/ml (95% confidence interval, 0.12–0.65) greater in children on ZDV-containing regimens at baseline than in those on ddI monotherapy ($P = 0.005$).

Clinical outcome

A total of 13 children (seven 3TC, six placebo) children developed a new AIDS event, of whom three (all on placebo) died. Two had AIDS at baseline and died of fungal pneumonia, and the third developed PCP and died. Six children developed their first AIDS event during the trial, four (on 3TC) failure to thrive, one (on placebo) had lymphoma with cytomegalovirus retinitis, and one (on placebo) had severe failure to thrive, tuberculous meningitis and progressive multifocal leukoencephalopathy. Four (two 3TC, two placebo) children with AIDS at randomization developed new AIDS events during the trial.

Analysis of growth responses showed that the AUCMB to 24 weeks was significantly greater in the 3TC group than in the placebo arm for height ($P = 0.03$) and height for age ($P = 0.04$), but not for weight or weight for age (Table 3; Fig. 3).

Discussion

There are few data on the use of 3TC in children and no previous studies have been placebo-controlled [7,12]. Lack of controlled data has resulted in concern about toxicity, particularly as pancreatitis was cited as a possible effect of toxicity of 3TC in children in the Phase I trial [7]. In our randomized trial of the addition of 3TC or matching placebo to current NRTI therapy including ddI over a maximum period of 72 weeks, serious adverse events were infrequent and similar in the 3TC and placebo groups. In particular, pancreatitis occurred in only one child who had very advanced disease and concurrent *M. avium-intracellulare* infection. In this child, serial monthly measurements of pancreatic amylase failed to predict the development of pancreatitis. Lack of predictive value has also been reported in

adult trials and suggests that measurement of serum amylase may not be helpful or cost-effective in children taking these combinations of antiretroviral therapies. Other adverse reactions reported in the Phase I/II paediatric study [7], including hyperactivity requiring medical intervention (two out of 89) and ataxia (one out of 89), were not observed in this trial.

Follow-up of children in this study involving 35 centres was excellent with only one child being lost to follow-up. 3TC syrup appeared to be well tolerated. We encouraged all children to start with syrup, but older children of appropriate weight could switch to tablets if they wished. Thirty seven older children switched during the course of the trial and tolerated the tablets well. The number of parents reporting that their child had interrupted therapy for adverse events or other reasons was also low. Although interruptions are likely to be underreported even with close follow-up, increases in MCV among children on the ZDV-containing regimens were maintained throughout the trial and were similar to those reported in the Phase II paediatric trial of ZDV therapy where follow-up was more intensive [13].

At entry to this trial, children had relatively advanced disease and had received nearly 3 years of antiretroviral therapy, mostly ZDV. The difference in CD4 cell counts at 24 weeks in favour of 3TC of 47×10^6/l was similar to that reported in adults where 3TC was added to ZDV-containing regimens in the CAESAR trial (34×10^6/l) [14], and the Phase II trial of Staszewski *et al.* (60×10^6/l) [6]. HIV-1 RNA, expressed as the change at 24 weeks or the AUCMB to 24 weeks, was significantly decreased in the 3TC arm compared with the placebo arm, similar to results in pretreated adults [6,14] and to those in the uncontrolled Phase I/II study in children [7]. Furthermore, as in adult trials [5,6], children switching from placebo to open-label 3TC at 24 weeks or later showed subsequent decreases in HIV-1 RNA. The trial was not powered to evaluate clinical efficacy. However, we did observe some benefit in terms of growth, although to a lesser degree to that observed in trials of treatment-naive children comparing ZDV–ddI versus ZDV alone [1] or ZDV–3TC versus ddI alone [2,15].

There appeared to be a difference in 3TC treatment effect depending on whether children entered the trial on ddI alone or on ZDV either alone or with ddI or ddC. Length of time on previous treatment, type of treatment (87% of ddI monotherapy group had received previous ZDV), and clinical status at baseline were similar in the ddI compared with the ZDV groups, and were controlled for in the analysis. These results must be interpreted with caution because this was a non-randomized comparison. However, they do suggest that 3TC and ddI may not be an ideal double combination to use in pretreated children. Studies of genotypic resistance and viral fitness are currently ongoing, which may throw more light on this observation.

There is optimism that combination antiretroviral regimens, particularly those including a protease inhibitor, can delay disease progression and prolong survival in HIV-infected adults. The natural history of HIV infection in vertically infected children differs from that in adults in some important respects, including faster progression to AIDS and death in a subgroup of children [16], and very high viral load levels in infancy, which gradually fall over the first 5 years in the absence of therapy [17]. US paediatric guidelines have recommended that children should commence triple therapy regimens early in disease [18]. However, problems remain, particularly with respect to the lack of availability of palatable formulations for young children of most of the protease inhibitor drugs, lack of data on toxicity and tolerability in the medium term for the protease inhibitor drugs that are available, and potential problems around long-term compliance with complicated regimens. The shorter term goal of achieving undetectable viral load in adults is being increasingly advocated. Only 6% of children reached undetectable HIV-1 RNA viral loads at 24 weeks in this trial. Thus, adding a single drug to current NRTI therapy is inadequate. Where possible, children who have been on antiretroviral therapy and are failing should now receive triple therapy regimens with at least two new drugs including a protease inhibitor. The results of this placebo-controlled trial add to current knowledge that 3TC is safe and well-tolerated in children when used in combination with other NRTI drugs and will contribute to HIV suppression when used in combination with other antiretroviral drugs.

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Appendix

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