Background & objectives: Information on nutritional status of HIV infected children from India is lacking and is required before taking up nutritional supplementation trials. Thus, the aim of the present study was to assess the growth and morbidity status of HIV infected children over a period of one year in a city in southern India.

Methods: This was an observational study carried out between July 2009 and February 2011, at two orphanages in Hyderabad, India. Seventy seven HIV-positive children aged between 1 and half and 15 years, both on and not on antiretroviral therapy (ART) were included. Nutritional status was assessed longitudinally for one year by weight gain, linear growth and body composition. Serum samples were analyzed for haemoglobin, micronutrients, CD4 and CD8 counts. Dietary intakes were assessed by institutional diet survey and morbidity data were recorded every day for 12 months.

Results: Mean energy intakes were less than recommended dietary allowance (RDA) in all age groups. Iron and folate intakes were less than 50 per cent of RDA; 46 (59.7%) children were stunted, 36 (46.8%) were underweight and 15 (19.5%) had low BMI for age. Anaemia was observed in 35 (45.5%) children. Micronutrient deficiencies such as vitamin D (40/77; 51.9%), vitamin A (11/77; 14.3%), folate (37/77; 48.1%), iron (38/77; 49.3%) were widely prevalent. HIV viral load was higher in children not on ART and those with morbidity. Respiratory (36.6%) and dermatological illnesses (18.8%) were the commonest presentations.

Interpretation & conclusions: Acute, chronic malnutrition and micronutrient deficiencies were common in HIV infected children, especially in those not on ART and having morbidity. With severe malnutrition being an alarming consequence of HIV, prophylactic nutritive care should be considered for integration into HIV care strategies besides initiation of ART to improve the nutritional status and quality of life of these children.

Key words HIV - India - malnutrition - micronutrient status - morbidity

The 2014 UNAIDS report estimates about 2.89 million people living with HIV in India, of whom about 7 per cent are children under the age of 15 yr. It has been shown that undernourished children with HIV have relatively decreased lean body mass (fat free mass- FFM) compared to fat mass, a feature distinct from undernutrition due to food inadequacy. Reduced FFM has been shown to be associated with high HIV
viral load and disease progression, and high risk of death. Infants and under five children with their immature immune systems and small protein reserves, are more susceptible to malnutrition and wasting.

Most of the HIV infected people live in resource poor countries of Africa and Asia where food insecurity is widespread and the diagnosis of HIV is often made only in the advanced stage. Malnutrition has been shown to be an important co-morbid condition, as these populations are vulnerable to high prevalence of food insecurity. HIV infected adults in India have higher rates of malnutrition, anaemia and hypoalbuminemia compared with HIV uninfected individuals, despite similar caloric intake. While it is known that protein energy malnutrition is the commonest manifestation of HIV in India, there are only a few reports on growth pattern and nutritional deficiencies in asymptomatic HIV infected children in the Indian context.

HIV Infected children with severe weight loss, in particular, have been shown to have energy requirements 50-100 per cent above normal. These could be secondary to the impaired absorption or utilization of micronutrients translating as poor appetite or increased needs. Nutritional supplements given to HIV infected children or adults have shown conflicting results where most nutrients have been shown to be beneficial, while the effect of some such as vitamin A has remained controversial. Data on nutritional supplementation in HIV-positive individuals, however, are limited and current evidence on their benefits need to be further substantiated, especially in the Indian context. It is also important to know the baseline nutritional status of the HIV infected individuals before supplementation may be planned. We undertook this study to assess the health, nutritional status and growth pattern of HIV infected children from select orphanages in Hyderabad, India.

**Material & Methods**

*Study population:* Five orphanages for children with HIV in Hyderabad were visited to assess nutritional status of children infected with HIV in this longitudinal study. Of the five orphanages only two gave consent for the study. There were 77 children in these two orphanages, aged between 1.5 and 15 yr, all of whom were HIV positive (37 boys and 40 girls). All the 77 HIV-positive children were followed up from July 2009 to February 2011. Of these, 44 children were receiving antiretroviral therapy (ART) and 33 children were not on ART. All the children were routinely receiving B complex, vitamins A and D, zinc and calcium supplements. Informed consent was taken from the caregivers of the children and assent was taken from children aged eight and above. Immunization history of all the children was collected. Children were assessed clinically, examined for physical and mental development, nutritional status, and evidence of any opportunistic infections including tuberculosis at baseline and thereafter every month for a period of one year after recruitment. The study protocol was approved by the Institutional Ethics Committee of the National Institute of Nutrition, Hyderabad.

*Morbidity assessment:* Morbidity was recorded every day by a clinician and a support staff. History of respiratory tract illness including upper respiratory infections with and without fever, lower respiratory infection; gastrointestinal manifestations such as gastroenteritis, parasitic infestations, loss of appetite, vomiting, constipation and pyrexia were recorded. Dermatological manifestations such as scabies, dermatitis, mumps, Herpes zoster, Herpes simplex, Molluscum contagiosum and lymphadenopathy were diagnosed clinically. Tuberculosis was diagnosed by Mantoux test and chest X-ray with sputum or gastric lavage examination and culture. Routine clinical monitoring included recording and treatment of minor infections, co-trimoxazole prophylaxis and ART administration. Nutritional supplementation was given to all the children by a physician designated for their care.

*Nutritional assessment:* Nutritional status of the children was assessed by anthropometry, body composition, serum biochemical parameters and diet survey. Anthropometric measurements included height, weight and skinfold measurements at four sites every month for one year. Weight was measured by SECA weighing scale, height by SECA stadiometer (USA) and skinfold measurements by Harpenden skinfold calipers (UK). Body mass index (BMI) was calculated from the height and weight as weight (kg)/height (m) squared. Body composition parameters such as body density, per cent body fat, body fat (kg) and lean body mass (LBM) were calculated by a standard procedure from skinfold measurements at four sites. The Z-scores for weight, height and BMI were computed based on the child’s age and gender using WHO standards. As per the WHO Global Database on Child Growth and Malnutrition recommendations, a cut-off Z score of < -2 was used to classify low weight-for-age (underweight), low height-for-age (stunting), and
low weight-for-height (wasting) as moderate and a Z score of <-3 to define severe undernutrition. None of the children had nutritional oedema.

**Diet survey:** Institutional diet survey with direct weighment method was conducted at baseline. Energy and nutrient intakes were derived using the Nutritive value of Indian foods.

**Serum markers:** Five ml of venous blood sample was collected from the children at three time points: at baseline, at 6th month and at the end of 1 year. Haemoglobin was assessed by cyanmethemoglobin method. Serum iron, zinc, and copper levels were assessed by high performance liquid chromatography (HPLC). Serum albumin was measured by using an autoanalyzer.

**HIV viral load and CD4, CD8 counts:** HIV RNA viral load (IU/ml) was measured using real-time quantitative polymerase chain reaction (RT-PCR) assay CD4 and CD8 cell counts (cells/µl) were determined using BD FACS Calibre Flowcytometer at baseline, 6 and 12 months. The Bioserve Biotechnologies (India) Pvt. Ltd. were outsourced to perform the above two investigations on payment.

**Statistical analysis:** Statistical analysis was performed using SPSS statistical software (SPSS Inc, Chicago, IL, USA) version 19. Correlations between nutritional deficiencies, CD4, CD8 and HIV viral load was done using Pearson’s correlation test. Z scores were calculated using WHO Anthro software. Median nutrient intakes were computed according to ages and expressed as percentage of recommended dietary allowances (RDA) of India.

**Results**

Between July 2009 and February 2011, 77 HIV positive children (37 boys and 40 girls) were screened. The ages ranged from 1.6 to 15.5 yr (mean age 9.12 yr, median 9 yr). There was no mortality or loss to follow up reported as all the children were institutionalized and under rehabilitative care.

Of the 77 children, 46 (59.7%) were stunted, 36 (46.8%) were underweight and 15 (19.5%) had low BMI for age. In terms of BMI for age, boys (29.7%, 11/37) appeared to be more undernourished when compared to girls (10%, 4/40) and had significantly (P<0.05) lesser per cent body fat, but higher lean body mass than girls. Underweight was comparable across all three age groups. Anaemia (Hb<11 g/dl) was prevalent in 35 of 77 children (45.5%) at baseline. Micronutrient deficiencies were widely prevalent, with vitamin D, iron and folate deficiencies being the most common. Vitamin D deficiency (< 20 ng/ml) was observed in 51.9 per cent (40/77) of children with mean serum 25 (OH) vitamin D being comparable between boys and girls. Nearly 50 per cent of children had low iron (38/77) and folate levels (37/77). Vitamin A deficiency was observed in 14.3 per cent (11/77) children. Overall, zinc deficiency was observed in 16.9 per cent (13/77) children, with the extent of zinc deficiency being higher in boys (8/37, 21.9%) compared to girls (5/40, 12.5%). Only one boy with HIV had low serum copper concentration, and serum calcium and serum albumin concentrations were low in 26 per cent (20/77) and 36.4 per cent (28/77) children respectively (Table I).

The mean energy intakes were less than the recommended dietary allowances (RDA) in children >9 yr but were normal in the other age groups (Table II). Fat intakes were more than 100 per cent RDA in children of all age groups (Figure). Iron and folic acid intakes were less than 50 per cent of RDA. In the >9-12 yr age group, Iron intakes were lower in girls compared to boys while intake of other nutrients was comparable between boys and girls of >9-12 yr and >12-15 yr.

HIV viral load was higher in the children not on ART compared to those on ART. Moderate underweight was prevalent in 54.5 per cent (24/44) children on ART, while it was 39.6 per cent (13/33) in those not on treatment. Children on ART had a high proportion of stunting (65.9%, 29/44) than those not on ART (51.5%, 17/33). Severe stunting was observed in 36.4 per cent (16/44) children on ART compared to 9 per cent (3/33) children who were not on ART. A higher proportion of children on ART (20.5%, 9/44) had low BMI compared to children not on ART (12.1%, 4/33). The serum nutritional profile was comparable between both the groups and equal proportion of children on and not on ART were anaemic. However, a significantly (P<0.05) higher proportion (66.0%, 29/44) of children who were on ART were vitamin D deficient compared to 40.0 per cent (13/33) not on ART. Though not significant, more children not receiving ART (6/33) were zinc deficient compared to those on ART (6/33) were zinc deficient.
Table I. Demographic, immune- and nutritional profile of the children

| Parameter                          | Males (n=37)       | Females (n=40)     | Total (n=77)      |
|------------------------------------|--------------------|--------------------|-------------------|
| Age (yr)                           | 8.96 ± 0.56        | 9.30 ± 0.48        | 9.13 ± 0.36       |
| WAZ                                | -1.29 ± 0.07       | -1.80 ± 0.18       | -1.72 ± 0.23      |
| HAZ                                | -1.47 ± 0.47       | -2.64 ± 0.22       | -2.12 ± 0.20      |
| BMI for age mean Z scores          | -1.48 ± 0.18       | -0.84 ± 0.14       | -1.24 ± 0.11      |
| Fat %                              | 13.05 ± 0.49       | 21.10 ± 0.52       | 16.70 ± 5.05      |
| Total fat (kg)                     | 2.98 ± 0.25        | 4.61± 0.27         | 3.66 ± 1.72       |
| Total LBM (kg)                     | 19.01 ± 0.88       | 16.91 ± 0.70       | 17.84 ± 4.6       |
| Absolute count CD4 (cells/µl)      | 1021.89 ± 110.37   | 1092.34 ± 161.76   | 1058.97 ± 866.16  |
| Absolute count CD8 (cells/µl)      | 1415.20 ± 125.09   | 1554.74± 290.69    | 1488.64 ± 1423.2  |
| HIV viral load (IU/ml)             | 104935.72 ± 48726.41 | 76212.25 ± 38648.96 | 89818.11 ± 266769.38 |
| Hb (g/dl)                          | 11.95 ± 0.50       | 11.80 ± 0.58       | 11.87 ± 3.37      |
| Vitamin A (µg/dl)                  | 34.52 ± 2.65       | 33.68 ± 2.52       | 34.04 ± 15.32     |
| Vitamin D (ng/ml)                  | 19.31 ± 1.34       | 20.30 ± 1.12       | 19.83 ± 7.46      |
| Zinc (µg/dl)                       | 97.38 ± 6.98       | 88.88 ± 6.37       | 93.05 ± 40.83     |
| Iron (µg/dl)                       | 91.30 ± 0.22       | 83.54 ± 7.85       | 87.42 ± 48.06     |
| Copper (µg/dl)                     | 121.10 ± 5.74      | 121.48 ± 4.95      | 121.29 ± 52.58    |
| Vitamin B₁₂ (pmol/l)               | 666.52 ± 40.02     | 763.75 ± 68.93     | 717.19 ± 344.23   |
| Folic acid (nmol/lit)              | 11.02 ± 1.23       | 15.35 ± 1.96       | 13.3 ± 10.26      |
| Serum albumin (g/dl)               | 3.62 ± 0.07        | 3.63 ± 0.09        | 3.63 ± 0.52       |

Values are mean ± SD  
WAZ, weight-for-age Z score (under weight); HAZ, height-for-age Z score (stunting); BMI, body mass index; LBM, lean body mass

ART (7/44). Serum vitamin A, iron and calcium levels were relatively higher in children on ART, though not significant, however, serum copper was significantly (P<0.01) higher in children on ART compared to those not on ART. Serum albumin and vitamin B₁₂ were also higher in children on ART.

The mean absolute CD4 and CD8 counts were comparable amongst males and females. The mean HIV viral load was higher in males than in females. The CD4:CD8 ratio was significantly (P<0.05) lesser in children receiving ART, however, the absolute CD4 counts were comparable between those on and not on ART (Table I). When viral load was quantified based on nutrition status, only children with vitamin D deficiency (74148± 46564 IU/ml) had significantly (P<0.05) higher viral load compared to those without vitamin D deficiency (>20ng/ml, 41560±34539 IU/ ml), and children with or without wasting or anaemia had comparable viral load.

The most common co-morbidities recorded were respiratory illnesses which included, upper and lower respiratory tract infections and pneumonia. Pneumonia was the commonest lower respiratory infection and was reported in 19 (24.6%) children while upper respiratory tract infection with fever was seen in 28 (36.6%) children. Pulmonary tuberculosis was the commonest opportunistic infection seen in 13 (16.8%) children. Dermatological manifestations such as scabies, dermatitis, Herpes simplex, Herpes zoster, Molluscum contagiosum, mumps, etc. were seen in 14 (18.8%) children and other non specific morbidities were reported in 27 (35%) children in the study.

When children were categorized based on presence or absence of co-morbidity at baseline, of the children with co-morbidity (no=32), 13 (40.6%) were undernourished, 20 (62.5%) were stunted and low BMI for age was observed in 6 (18.8%) children. More children without co-morbidity (n=45) were however, more undernourished (n=23, 51%). LBM and fat % were comparable between both the groups. As expected absolute CD4, CD8 counts and CD4:CD8 ratio were lower and HIV viral load was significantly
### Table II. Mean dietary intakes of children with HIV (n=66)

| Dietary parameters (per day) | Age of children (yr) | 0-4 (n=2) | >4-6 (n=10) | >6-9 (n=18) | >9-12 (n=20) | >12-15 (n=16) |
|-----------------------------|----------------------|-----------|-------------|-------------|--------------|---------------|
|                             |                      | Male (9)  | Female (11) | Male (7)    | Female (9)   |               |
| Energy (kcal)               |                      | 1131.67 ± 526.78 | 1708.55 ± 249.69 | 1729.84 ± 157.32 | 1727.94 ± 248.83 | 1745.30 ± 304.35 | 2107.61 ± 214.59 | 1792.54 ± 278.26 |
| Protein (g)                 |                      | 34.83 ± 12.41 | 43.66 ± 5.13 | 45.56 ± 3.84 | 49.05 ± 7.26 | 49.42 ± 8.20 | 55.19 ± 7.09 | 48.92 ± 8.70 |
| Fat (g)                     |                      | 32.14 ± 4.97 | 35.36 ± 4.06 | 37.47 ± 4.67 | 41.86 ± 6.60 | 41.46 ± 7.24 | 42.61 ± 6.90 | 40.95 ± 7.78 |
| Calcium (mg)                |                      | 539.89 ± 115.30 | 629.20 ± 53.90 | 658.94 ± 88.65 | 684.62 ± 90.64 | 664.39 ± 84.94 | 674.90 ± 67.90 | 719.89 ± 161.53 |
| Iron (mg)                   |                      | 3.76 ± 1.63 | 6.29 ± 3.73 | 7.16 ± 4.17 | 9.86 ± 4.95 | 9.11 ± 3.81 | 8.38 ± 2.80 | 7.48 ± 2.80 |
| Vitamin A (µg)              |                      | 570.97 ± 226.34 | 657.43 ± 283.61 | 727.45 ± 298.65 | 1120.34 ± 486.65 | 1048.63 ± 426.64 | 1067.54 ± 433.10 | 870.74 ± 395.83 |
| Thiamine (mg)               |                      | 0.47 ± 0.14 | 0.56 ± 0.06 | 0.60 ± 0.07 | 0.67 ± 0.11 | 0.65 ± 0.13 | 0.72 ± 0.12 | 0.65 ± 0.13 |
| Riboflavin (mg)             |                      | 0.69 ± 0.31 | 0.81 ± 0.18 | 0.83 ± 0.17 | 0.96 ± 0.24 | 0.9 ± 0.24 | 1.07 ± 0.22 | 1.01 ± 0.24 |
| Nicotinic acid (mg)         |                      | 7.18 ± 3.39 | 9.30 ± 1.82 | 9.26 ± 1.60 | 8.29 ± 2.14 | 8.14 ± 2.36 | 10.59 ± 2.12 | 9.11 ± 2.09 |
| Ascorbic acid (mg)          |                      | 0.48 ± 0.06 | 17.31 ± 53.05 | 31.91 ± 63.49 | 74.89 ± 83.67 | 78.79 ± 73.73 | 75.21 ± 93.02 | 68.72 ± 84.69 |
| Folic acid total (µg)       |                      | 38.83 ± 11.60 | 53.74 ± 30.57 | 66.31 ± 41.82 | 98.63 ± 50.69 | 96.92 ± 42.64 | 91.38 ± 50.0 | 88.29 ± 52.52 |
| Vitamin B 12 (µg)           |                      | 74.91 ± 33.81 | 77.5 ± 30.32 | 68.25 ± 37.72 | 42.20 ± 49.05 | 34.21 ± 46.22 | 62.9 ± 58.59 | 50.65 ± 47.16 |

Values are mean ± SD
higher in children with co-morbidity. Nearly 50 per cent children with co-morbidity had anaemia as against 44.9 per cent among children not having any morbidity at baseline. A higher proportion of children with co-morbidity had vitamin D deficiency (62.5%) compared to those without co-morbidity (50.7%) and a significantly higher proportion of children with co-morbidity had zinc deficiency (50% vs 13%). Vitamin A deficiency was seen in 37.5 per cent children.
Body fat, fat per cent and LBM were seen to gradually increase from baseline to the end of one year. Significant increases in terms of LBM and body fat were also seen at 6 months from baseline. Mean absolute CD4 counts increased from 1058.97±866.16 to 1173.96 ± 659.55 cells/µl (P<0.01) and CD8 counts increased from 1488.64±1423.2 to 1511.37 ± 632.97 cells/µl (P<0.05) from baseline to 6th month with a concomitant decrease in HIV viral load from 89818.11 ± 266769.38 to 86395.44 ± 337401.08 IU/ml (P<0.05). Underweight and stunting seemed to decrease over time. Low BMI for age was seen in relatively lesser number of children, 16.9 per cent at baseline, however, it increased by the end of one year. When categorized based on viral load, of the children (n=39) with high viral load (> median) undernutrition was seen in eight (20.5%) children, which improved over time to 12.8 per cent (n=5) and 15.4 per cent (n=6) at 6 months and one year, respectively. However, the number of children with low BMI had increased from 10.3 per cent at baseline to 23.1 per cent at 6 months and again reduced to 15.4 per cent by 12th month. The proportion of stunting was 15.4 per cent at baseline, which increased to 35.9 per cent at 6th month and remained the same at the end of the 12th month.

**Discussion**

High occurrence of stunting, underweight, and anaemia in children with HIV was seen in our study. Micronutrient deficiencies were common with vitamin D, iron and folate being most common followed by vitamin A and zinc. However, with improvement in the micronutrient status, a modest decrease in the prevalence of underweight, stunting and wasting was observed over the study period of one year. The prevalence of undernutrition among HIV infected children in India has been reported to range from 17-63 per cent\(^8\)\(^9\)\(^17\)\(^19\). These figures were higher than those reported among HIV infected children from Africa where undernutrition was 14 per cent and stunting was reported to be 31 per cent\(^20\)\(^21\). A large number of children were stunted in our study population, which was in line with a study from Tamil Nadu\(^19\). The stunting was seen more in children >9 yr, which could probably be due to disease progression with age compared to preschool children and children >4-9 yr. These children also had lower mean CD4 and CD8 counts also indicating disease progression. The HIV viral load in this age group was expectedly higher given the lower CD4 and CD8 counts. A similar trend was also reported by Padmapriyadarshini \cite{et al}\(^19\).

Apart from wasting, HIV infected people suffer from micronutrient deficiencies which can further compromise the immune system resulting in a poorer outcome\(^22\). The present study showed a high occurrence of anaemia similar to other studies on children and adults with HIV in India. Of the 35 children who were anaemic in our study, 51.4 per cent were deficient in serum folate and iron levels. These findings are in agreement with the low iron and folate dietary intakes recorded for these children. Castro \cite{et al}\(^22\) also reported that folate and iron deficiency were the most common causes of anaemia in HIV infected adults in Brazil. Low vitamin A levels were associated with lower CD4 counts and significantly higher HIV viral load in our study. A similar trend was noticed by Steenkamp \cite{et al}\(^23\) who observed low CD4 counts, high viral load with low levels of vitamins A, D, E and zinc in their study in HIV infected children in Manguang.

Slowing down of the disease progression was also observed during the course of study, with significant increase in CD4 cell count and decrease in HIV viral load. This is consistent with similar studies that have reported the benefits of nutritional support and micronutrient supplementation in delaying disease progression and the onset of ART\(^24\)\(^25\). Respiratory illnesses are a very common paediatric illness, but are persistent and refractory to treatment in children with HIV. The most common clinical presentations observed in this study were upper respiratory tract infections with or without fever and lower respiratory tract infections seen. Tuberculosis was reported in 16.8 per cent children, similar to Verghese \cite{et al}\(^26\) who reported 14 per cent, but was much lower than that reported by Madhivinan \cite{et al}\(^27\) (35%), and Pol \cite{et al}\(^28\) (38.8%). Skin lesions were seen in <20 per cent children in our study. The incidence of skin lesions in paediatric HIV has varied from 22.10 per cent to as high as 40.70\(^27\)\(^28\). Scabies was seen in 15.58 per cent children, a figure much higher than that reported by Shah \cite{et al}\(^8\). The cases of Molluscum contagiosum were low in our study (3.89%) compared to an earlier study from India\(^29\).

The strengths of our study included morbidity recall every day of all the children enrolled in the study for 12 months, besides monthly monitoring of weight and body composition parameters. Though anaemia in HIV has been widely studied, we have assessed serum micronutrients such as vitamin A, vitamin D, zinc, iron,
copper, vitamin B₁₂, and folic acid. This study also had a few limitations such as small sample size, and not having age and sex-matched non-HIV infected controls living in orphanages for comparison.

In conclusion, our results showed that micronutrient deficiencies were very common in paediatric HIV which should be addressed. It is known that morbidities and growth failure are rampant in HIV infected individuals, corrective measures need to be directed at micronutrient deficiencies which would compromise immunity and increase morbidity. With severe malnutrition being an alarming consequence of the HIV epidemic, prophylactic nutritive care could be considered for integration into HIV child care strategies to improve nutritional status and quality of life in such children. A judicious mix of macro- and micronutrient supplements is indicated for reducing growth failure and correcting the nutrient deficiencies which are widely prevalent in children infected with HIV.

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