ROLE OF MICRONUTRIENTS IN HIV INFECTION

G Hussey, *MB ChB, MMed, DTM&H, MSc Clin Trop Med*
H Buys, *MB ChB, FCP, MRCP*
C Cowburn, *MB ChB, Dip HIV Med*
B Eley, *MB ChB, FCP*
M Hendricks, *MB ChB, MMed, MTropMed*
Institute of Infectious Diseases and Molecular Medicine and School of Child and Adolescent Health, University of Cape Town

More than 60% of the estimated 40 million persons with HIV/AIDS worldwide live in sub-Saharan Africa, where poverty, social insecurity, food shortages and malnutrition are major problems.[1] In children under the age of 5 years, who live in developing countries, malnutrition has been associated with 50% of the 10.8 million deaths mainly caused by neonatal disorders, diarrhoea, pneumonia, malaria and HIV/AIDS.[2] Likewise micronutrient deficiencies are widespread and are associated with increased morbidity and mortality particularly in relation to infectious diseases.[3] This review focuses on the interaction between micronutrients and HIV/AIDS and discusses recent research findings that may have important public health implications in terms of the case management of persons with HIV/AIDS.

IMPORTANCE OF MICRONUTRIENTS IN GENERAL

Micronutrients are trace elements and vitamins essential to all micro-organisms, plants and animals. They play an important role in gene expression, cellular differentiation and immune function. In addition many exhibit antioxidant properties. They are therefore essential for human health and development.

Of all the micronutrients vitamin A and zinc deserve special mention. Deficiencies of both are widespread in developing countries and contribute to the morbidity and mortality from infectious diseases. In children a pooled analysis of randomised controlled trials of prophylactic zinc supplementation has been associated with a 41% reduction in the incidence of pneumonia and an 18% reduction in the incidence of diarrhoeal disease.[4] Zinc was also found to have a therapeutic benefit in sick children through promoting recovery from acute and persistent diarrhoea, and from severe pneumonia.[5] Zinc supplementation was also associated with a two-thirds reduction in mortality in a randomised, double-blind, controlled trial of small-for-gestational age infants in India who were given zinc supplementation.[6] Vitamin A supplementation has resulted in a 23% reduction in all-cause mortality in children aged 6 - 60 months[7] and more specifically in children with severe measles.[8]

MICRONUTRIENTS AND HIV/AIDS

The hallmark of AIDS is immune suppression resulting in acute, recurrent and chronic opportunistic infections. While HIV is the main cause of the immune suppression, other factors contribute. These include concomitant malnutrition, opportunistic infections, in particular tuberculosis, and micronutrient deficiencies.

Micronutrient deficiencies are common in persons with HIV infection and AIDS in both developing and developed countries.[9] They occur as a consequence of a number of factors including reduced intake as a result of the anorexia that occurs with AIDS and opportunistic infections, excessive losses in the stools in patients with diarrhoea, malabsorption and parasitic infestations.[10] These deficiencies are more pronounced in individuals with advanced disease and in situations where diets are inadequate in meeting the recommended daily requirements of micronutrients. Recent studies have highlighted significant multiple micronutrient deficiencies in South Africans. A study of stable, HIV-infected children in Cape Town revealed that 62% had two or more trace element or vitamin deficiencies.[11] Most children were vitamin A deficient. In addition another study reported significant vitamin A and zinc deficiency in adults. Levels tended to be lower in individuals with stage 3 and 4 disease and there was a positive correlation with CD4 counts.[12]

EFFECTS OF MICRONUTRIENT DEFICIENCIES IN HIV/AIDS

Micronutrients are essential to immune function and their deficiencies may act as co-factors in HIV disease progression. Vitamins A, B_6, B_{12}, C and E, beta-carotene, zinc, copper, selenium, magnesium and iron deficiencies have all been described in association with HIV infection.[13,14] In general vitamin A deficiency has been associated with increased morbidity and mortality and with increased transmission of HIV from mother to child.[15,16]

Selenium is an important antioxidant and deficiency has been reported to reduce immune function in animal studies.[17] Low plasma selenium levels have been described early in the disease process and are linked to faster disease progression and higher morbidity in HIV-positive children.[18] It is known that cardiac muscle dysfunction in HIV-infected children is associated with a higher mortality.[19] The contribution of copper and manganese to immune function is not clear but children with overt copper deficiency (Menkes' syndrome) are prone to infection.[17] Copper deficiency has been documented in adults with HIV infection. The equilibrium in iron homeostasis is delicate – deficiency or excess both negatively affect immune function. As in other groups of HIV-infected subjects, anaemia is a common finding in HIV-infected South African children[20,21] and is known to enhance disease progression and increase mortality.[22]

There is growing evidence to support zinc supplementation as a possible public health intervention strategy that could enhance the health status of HIV-infected children and undernourished children in developing countries. Zinc is best derived from animal rather than cereal proteins. Bioavailability is good from breast-milk in the first 4 - 6 months of life, but this is markedly reduced by high-phytate diets. Excessive zinc losses occur in diarrhoeal disease leading to loss of the gastrointestinal epithelial integrity and absorptive power.[23] HIV-infected children in Zaire were found to have an 11-fold higher risk than uninfected controls of dying from diarrhoeal disease.[24] The other consequence of zinc deficiency is impaired thymolymphoid integrity and reversible immune dysfunction, particularly of T-lymphocyte cell-mediated immunity, and hence deficiency may contribute to infectious morbidity. Zinc deficiency appears to be relatively common in adult HIV-infected individuals even at the asymptomatic stage but is not well documented in children[13,25] and may be more common than is currently appreciated.

SINGLE MICRONUTRIENT SUPPLEMENTS IN HIV/AIDS

A number of studies have demonstrated the beneficial effects of supplementation with single micronutrients. Most studies, excluding those evaluating the role of vitamin A, have been small ones involving less than 50 individuals.

There is no doubt that vitamin A supplementation is beneficial in children. Vitamin A supplementation in HIV-infected children has been shown to improve immune function,[26] reduce morbidity from diarrhoeal disease[27] and reduce mortality in HIV-infected children.[28] However, a number of clinical trials in Africa indicated that vitamin A supplementation in pregnant women was not associated with reduced mother-to-child transmission of HIV.[29-31]

Vitamin B_6 supplementation in HIV-positive adults has been associated with improved survival.[32] Supplementation with vitamins B_1 or B_6 or vitamin E has been associated with reduced progression to AIDS, and vitamin B_{12} supplementation has been associated with improvement of AIDS dementia. Selenium supplementation in a small cohort of HIV-infected adults with low selenium levels and dilated cardiomyopathy resulted in improved left ventricular function in 2 out of 3 patients.[33] Zinc supplementation in HIV-positive adults showed potential benefit in reducing candida- and pneumocystis-related infections, increasing CD4 lymphocyte counts and improving weight gain.[34]

MULTIPLE MICRONUTRIENT INTERVENTIONS

While a number of studies have shown potential benefit following supplementation with individual micronutrients, there have been few randomised clinical trials evaluating the impact of multiple micronutrients in combination.

Multivitamin supplementation in pregnant HIV-positive women in Tanzania was shown to reduce progression of HIV to AIDS or death from AIDS-related causes significantly by 59% over the first 2 years and by 29% over the whole 4 - 8-year supplementation and follow-up period. The authors of this study suggest that multivitamins could be a low-cost means of delaying initiation of antiretroviral therapy in HIV-infected women.[35]

In a study from Zimbabwe no effect on diarrhoeal morbidity or mortality was found from supplementing HIV-positive adults with high doses of vitamins A, C and E, zinc and selenium for 2 weeks.[36] However, the patients had advanced HIV disease with persistent diarrhoea and may not have absorbed the micronutrients.

In Thailand a study of a commercial supplement containing 18 micronutrients and taken for 48 weeks was associated with a reduction in mortality (mortality hazard ratios = 0.37 (95% confidence interval (CI) 0.13, 1.06) particularly in those with a CD4 count $<100 \times 10^6/l$.[37]

SAFETY OF MICRONUTRIENTS

While in most instances single micronutrient supplements have been associated with benefits, a few studies have indicated possible adverse effects in association with excessive supplementation, particularly of vitamin A and zinc.

A study done on HIV-infected pregnant and lactating women in Tanzania who were randomly assigned to vitamin A or multivitamins (excluding vitamin A) showed an increased risk of HIV-1 vertical transmission in women receiving vitamin A supplementation. Multivitamin supplementation, however, significantly reduced HIV-1 transmission in immunologically and nutritionally compromised mothers as well as child mortality at 24 months.[38] Similar studies from Malawi and South Africa reported no such effect.[20,29] Observational studies have also suggested an increased rate of progression in individuals consuming large amounts of vitamin A. Experimental data also suggest that vitamin A increases viral replication *in vitro*. It is reassuring to note that a number of clinical studies have not demonstrated such an effect.

There is also a concern that zinc may potentiate HIV replication since the HIV-Tat protein and the HIV nucleocapsid NCp7 proteins are strongly zinc dependent.[39] Evidence from one observational study suggested that high-dose zinc supplementation was associated with increased HIV/AIDS disease progression and mortality in HIV-positive adults.[32]

UNANSWERED QUESTIONS?

Should we provide routine micronutrient supplements to persons with HIV/ AIDS? If we do, what dose do we use and for how long? What is the role of micronutrient supplementation in individuals treated with antiretrovirals? These are important questions to address given concerns that have been raised about possible adverse events, particularly in relation to zinc.

For these reasons we have been involved in studies evaluating the safety and efficacy of micronutrient supplementation in HIV-positive children. The first study was designed to assess the safety of low- and high-dose zinc supplementation in HIV-positive children. This study also provided information that informed our second study, which was designed to evaluate the efficacy of zinc and a micronutrient mixture in HIV-positive children.

SAFETY OF ZINC SUPPLEMENTATION IN HIV-POSITIVE CHILDREN[40]

The objective of this study was to evaluate the safety and tolerability of zinc supplementation in HIV-infected children. We conducted a double-blind, randomised, placebo-controlled clinical trial in 39 clinically stable HIV-infected children aged 6 months to 6 years attending the Infectious Diseases Clinic at Red Cross Children's Hospital, Cape Town. Children excluded from the study included those who had an intercurrent infection or an axillary temperature of > 38°C, had recently been hospitalised, had any opportunistic infection, tuberculosis or chronic lung disease or had received high-dose vitamin A, trace elements or zinc supplements within the preceding 8 weeks. The patients were randomly assigned to one of three treatment arms: placebo, a low-dose zinc sulphate supplement (0.5 mg/kg/day elemental zinc), and a high-dose zinc supplement (3 mg/kg/day to a maximum of 45 mg elemental zinc). The trial drugs were given orally daily over 6 weeks and the children were seen weekly for 12 weeks from start to end of the study. Children were clinically evaluated at baseline and at monthly intervals. Investigations including CD4 counts and viral loads were done at baseline, and at 6 and 12 weeks' follow-up. The baseline characteristics of the three groups were similar. Of note, the median age and CD4 counts were 30 months and 20% respectively. Zinc supplementation was not associated with any significant clinical adverse events and did not affect CD4 counts or viral loads. High-dose zinc supplementation in HIV-infected children was safe and generally well tolerated. The study concluded that clinical trials to assess the efficacy of zinc to reduce infectious morbidity and mortality in HIV-positive children need to be done.

EFFICACY OF ZINC SUPPLEMENTATION COMPARED WITH MULTIPLE MICRONUTRIENTS[41]

The objective of this randomised, placebo-controlled clinical trial was to determine the efficacy of zinc and a multiple micronutrient mixture in reducing morbidity and mortality in 6 - 72-month-old HIV-positive children at Red Cross Hospital, Cape Town. Children were randomised to one of three groups: group A received multivitamins and placebo, group B received multivitamins and trace elements, and group C received multivitamins and zinc. Children with a history of a significant acute illness within the last 6 weeks before the trial, who had received high-dose vitamin A or mineral supplements within the past 3 months or who were suspected of having tuberculosis or another invasive opportunistic infection were excluded from the study.

All children received the standard multivitamin syrup (5 ml/day): vitamin A (3 000 U), vitamin D (300 IU), vitamin C (30 mg), vitamin B_1 (1.65 mg), vitamin B_2 (1.32 mg), nicotinamide (11 mg) and vitamin B_6 (1 mg). The trace element solution contained the following (1 ml): zinc acetate 0.51 mg (providing 506.28 µg/ml elemental zinc), copper sulphate 61 µg (elemental copper), manganese sulphate (0.024 µg/ml manganese), selenium selenite 8 µg/ml, magnesium chloride 71.7 µg/ml, potassium iodide 0.08 µg/ml, chromic chloride 0.70 µg/ml, citric acid, sodium citrate, raspberry flavouring, and hydroxybenzoate. The daily dose was 1 ml/kg/day with a maximum of 20 ml/day. The dose of zinc was 3 mg/kg/day, with a maximum of 45 mg daily. The trial drugs were given orally daily over 6 months and the children were seen monthly for a year. Children were clinically evaluated at baseline and at monthly intervals. Investigations including CD4 counts and viral loads were done at baseline, and at 3-monthly and 6-monthly intervals, respectively.

The primary endpoint was the reduction in significant adverse events including hospitalisation and death. A total of 124 children were enrolled in the study (mean age 32 months). Baseline characteristics were similar for all children with the exception that there were significantly more immuno-compromised children in the group receiving zinc supplements (group C): 43% versus 25% in the placebo group (group A). Compared with the placebo and trace element groups (groups A and B) the zinc-treated group reported significantly fewer episodes of hospitalisation and diarrhoeal disease. In addition there was a tendency towards reduced episodes of pneumonia in the zinc-treated group. There was no difference in the groups receiving trace elements and placebo. No significant interaction between micronutrients and antiretrovirals were reported. In conclusion, zinc supplementation was associated with reduced infective episodes and hospitalisations in HIV-positive children.

CONCLUSION

In summary, micronutrients may be involved in the pathogenesis of HIV disease. Micronutrient deficiencies are

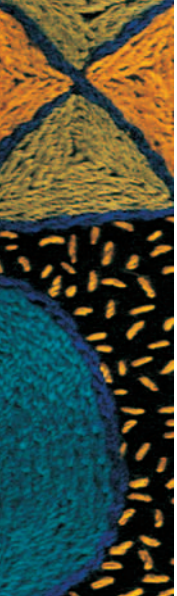
common in persons with HIV/AIDS, are more pronounced in persons with severe disease, and contribute to morbidity and mortality. Micronutrient supplementation, in particular zinc and vitamin A, are beneficial in HIV-positive persons. A daily dose of 3 mg zinc for 6 months has been shown to be safe and effective, and 6-monthly supplements of high-dose vitamin A likewise (50 000 IU before 6 months; a single dose of 100 00 IU between 6 and 11 months; and a single dose of 200 000 IU every 6 months from 12 months onward). Multivitamins have been found to be effective in delaying disease progression and in reducing vertical transmission in immunologically and nutritionally compromised HIV-infected women. However, more work on multiple micronutrients needs to be done; in particular, optimal formulations and dosage regimens have to be defined. Nevertheless, in situations where micronutrient deficiencies are endemic, these nutrients should be provided through food fortification or micronutrient supplements, containing at least 1 - 2 times the recommended dietary allowance, where available.

Nutritional interventions to improve the health and well-being of persons living with HIV/AIDS, including micronutrient supplementation, need to be optimised and research into identifying optimal interventions and operational strategies is encouraged. However, this should not be done to the detriment of antiretroviral strategies, the one intervention to date that has consistently been shown to be associated with a reduction in the disease burden associated with AIDS.

We thank the mothers and children who so willingly participated in these studies, the outpatient staff and the Pharmacy Department at Red Cross Children's Hospital, and BMS Secure the Future for funding the research.

REFERENCES

1. Joint United Nations Programme on HIV/AIDS (UNAIDS), World Health Organization (WHO). *Aids Epidemic Update*: 2004. Geneva: UNAIDS/WHO, 2004.
2. Black RE, Morris SS, Bryce J. Where and why are 10 million children dying every year? *Lancet* 2003; **361**: 2226–2234.
3. Tomkins A, Watson F. *Malnutrition and Infection. A review.* Geneva: ACC/SCN, 1989.
4. Zinc Investigators' Collaborative group. Prevention of diarrhoea and pneumonia by zinc supplementation in children from developing countries: pooled analysis of randomised controlled trials. *J Pediatr* 1999; **135**: 689-697.
5. Brooks WA, Yunus M, Santosham M, *et al.* Zinc for severe pneumonia in very young children: double-blind placebo-controlled trial. *Lancet* 2004; **363**: 1683–1688.
6. Black RE. Zinc deficiency, infectious disease and mortality in the developing world. *J Nutr* 2003; **133**: 1485S-1489S.
7. Beaton GH, Martorell R, Aronson KJ, *et al.* Effectiveness of vitamin A supplementation in the control of young child morbidity and mortality in developing countries. ACC/SCN state-of-the-art series nutrition policy discussion paper No. 13, December 1993, International Nutrition Program, Faculty of Medicine, University of Toronto, Ontario, Canada.
8. Hussey GD, Klein M. A randomised, controlled trial of vitamin A in children with severe measles. *N Engl J Med* 1990; **323**: 160-164.
9. Buys H, Hendricks M, Eley B, Hussey G. The role of nutrition and micronutrients in paediatric HIV infection. *South African Dental Journal* 2002; **57**: 454-456.
10. Winter H. Gastrointestinal tract function and malnutrition in HIV-infected children. *J Nutr* 1996; **126**: 2620s.
11. Eley BS, Sive AA, Abelse L, Kossew G, Cooper M, Hussey GD. Growth and micronutrient disturbances in stable, HIV-infected children in Cape Town. *Ann Trop Paed* 2002; **22**: 19-23.
12. Visser ME, Maartens G, Kossew G, Hussey GD. Plasma vitamin A levels in HIV infected adults in Cape Town, South Africa. *Br J Nutr* 2003; **89**: 475-482.
13. Beach R, Mantero-Atienza E, Shor-Posner G, *et al.* Specific nutrient abnormalities in asymptomatic HIV-1 infection. *AIDS* 1992; **6**: 701-708.
14. Periquet B, Jammes N, Lambert W, *et al.* Micronutrient levels in HIV-1-infected children. *AIDS* 1995; **9**: 887-893.
15. Sommer A, Hussaini G, Tarwotjo I, Susanto D. Increased mortality in children with mild vitamin A deficiency. *Lancet* 1983; **3**: 588-589.
16. Semba RD, Miotti PG, Chiphangwi JD, *et al.* Maternal vitamin A deficiency and mother-to-child transmission of HIV-1. *Lancet* 1994; **343**: 1593-1597.
17. Beisel W. Single nutrients and immunity. *Am J Clin Nutr* 1982; **35**; 417-468.
18. Campa A, Shor-Posner G, Indacochea F, *et al.* Mortality risk in selenium-deficient HIV-positive children. *J Acquir Immune Defic Syndr Hum Retrovirol* 1999; **20**: 508-513.
19. Lipshutz S. Dilated cardiomyopathy in HIV-infected patients. Editorial. *N Engl J Med* 1998; **339**: 1153-1154.
20. Friedland I, Snipelisky M. Vertically transmitted HIV-1 infection in children. *S Afr Med J* 1991; **79**: 157-159.
21. Eley BS, Sive AA, Shuttleworth M, Hussey GD. A prospective, cross-sectional study of anaemia and peripheral iron status in antiretroviral-naive, HIV-1 infected children in Cape Town, South Africa. *BMC Infectious Diseases* 2002; **2:** 3.
22. Mocroft A, Kirk O, Barton S, *et al.* Anaemia is an independent predictive marker for clinical prognosis in HIV-infected patients from across Europe. *AIDS* 1999; **13**: 943-950.
23. Shankar A, Prasad A. Zinc and immune function: the biological basis of altered resistance to infection. *Am J Clin Nutr* 1998; **68**: 447S-463S.
24. Thea D, St Louis M, Atido U, *et al.* A prospective study of diarrhoea and HIV-1 infection among 429 Zairian infants. *N Engl J Med* 1993; **329**: 1696-1701.
25. Bogden J, Baker H, Frank O, *et al.* Micronutrient status and HIV infection. *Ann NY Acad Sci* 1990; **587**: 189-195.
26. Hussey G, Hughes J, Potgieter S, *et al.* Vitamin A status and supplementation and it's effects on immunity in children with acquired immune deficiency syndrome. Department of Paediatrics, UCT, 1995.
27. Coutsoudis A, Bobat R, Coovadia H, Huhn L, Tsai W, Stein Z. The effects of vitamin A supplementation on the morbidity of children born to HIV infected women. *Am J Public Health* 1995; **85**: 1076-1081.
28. Fawzi W, Mbise R, Hertzmark E, *et al.* A randomised trial of vitamin A supplements in relation to mortality among HIV-infected children in Tanzania. *Pediatr Infect Dis J* 1999; **18**: 127-133.
29. Coutsoudis A, Pillay K, Spooner E, Kuhn L, Coovadia HMI. Randomized trial testing the effect of vitamin A supplementation on pregnancy outcomes and early mother-to-child HIV-1 transmission in Durban, South Africa. *AIDS* 1999; **13:** 1517-1524.
30. Kumwenda N, Miotti PG, Taha TE. Antenatal vitamin A supplementation increases birth weight and decreases anemia among infants born to human immunodeficiency virus-infected women in Malawi. *Clin Infect Dis* 2002: **35**: 618-624.
31. Fawzi WW, *et al.* Randomized trial of vitamin supplements in relation to vertical transmission of HIV-1 in Tanzania. *J Acquir Immune Defic Syndr* 2000: **23**: 246-254.
32. Tang A, Graham N, Saah A. Effects of micronutrient intake on survival in HIV-1 infection. *Am J Epidemiol* 1996; **143**(12): 1244-1256.
33. Chariot P, Perchet H, Monnet I. Dilated cardiomyopathy in HIV-infected patients. *N Engl J Med* 1999; **340**: 732.
34. Mocchegiani E, Veccia S, Ancarani F, Scalise G, Fabris N. Benefit of oral zinc supplementation as an adjunct to AZT therapy against opportunistic infections in AIDS. *Int J Immunopharmacol* 1995; **17**: 719-727.



35. Fawzi WW, Msamanga GI, Spiegelman D, *et al.* 1998 Randomised trial of effects of vitamin supplements on pregnancy outcomes and T cell counts in HIV-1-infected women in Tanzania. *Lancet* 1988; **351**: 1477-1482.
36. Kelly P, Musonda R, Kafwembe E, Kaetano L, Keane E, Farthing M. Micronutrient supplementation in the AIDS diarrhoea-wasting syndrome in Zambia. *AIDS* 1999; **13**: 495-500.
37. Jiamton S, Pepin J, Suttent R, *et al.* A randomized trial of the impact of multiple micronutrient supplementation on mortality among HIV-infected individuals living in Bangkok. *AIDS* 2003, **17**: 2461-2469.
38. Fawzi WW, Msamanga GI, Hunter D, *et al.* Randomized trial of vitamin supplements in relation to transmission of HIV-1 through breastfeeding and early mortality. *AIDS* 2002, **16**: 1935-1944.
39. Baum MK, Shor-Posner G, Campa A. Zinc status in human immunodeficiency virus infection. *J Nutr* 2000; **130:** 1421S-1423S.
40. Buys H, Hendricks M, Eley B, Kossew G, Hussey G. Safety of zinc supplementation in HIV-infected children in Cape Town. School of Child and Adolescent Health Annual Research Day, University of Cape Town, Oct 2003.
41. Buys H, Cowburn C, Hendricks M, Eley B, Hussey G. Efficacy of micronutrient supplementation in HIV positive children. School of Child and Adolescent Health Annual Research Day, University of Cape Town, Oct 2004.