

## Vitamin A and immunity to viral, bacterial and protozoan infections

R. D. Semba

Department of Ophthalmology, Johns Hopkins University School of Medicine, Baltimore, MD 21205, USA

Studies in animal models and cell lines show that vitamin A and related retinoids play a major role in immunity, including expression of mucins and keratins, lymphopoiesis, apoptosis, cytokine expression, production of antibody, and the function of neutrophils, natural killer cells, monocytes or macrophages, T lymphocytes and B lymphocytes. Recent clinical trials suggest that vitamin A supplementation reduces morbidity and mortality in different infectious diseases, such as measles, diarrhoeal disease, measles-related pneumonia, human immunodeficiency virus infection and malaria. Immune responses vary considerably during different infections, and the available data suggest that the modulation of immune function by vitamin A may also vary widely, depending on the type of infection and immune responses involved.

### Vitamin A: Immunity: Infection: Measles: Malaria

In the last two decades, considerable evidence has emerged that vitamin A plays an important role in immunity to infectious diseases. Perhaps more is known about how vitamin A modulates immune function than for any other micronutrient, yet there are still major gaps in our knowledge. Community- and hospital-based clinical trials have demonstrated that oral vitamin A supplementation or fortification can reduce preschool child mortality in developing countries by about one-third (Beaton *et al.* 1993). Recent experiments using animal and various cell lines suggest that vitamin A and related retinoids modulate many different immune response elements, including expression of keratins and mucins, lymphopoiesis, apoptosis, cytokine production, function of neutrophils, natural killer cells, monocytes or macrophages, T lymphocytes and B lymphocytes, and production of immunoglobulin (Ig; Semba, 1998). Regulation of immune responses by vitamin A and its active metabolites has been shown to occur via nuclear retinoic acid receptors at the level of the genome as well as via a pathway involving retinoids (Semba, 1998).

Although vitamin A and related retinoids have been shown to have important effects on immune function, the therapeutic value of vitamin A supplementation in enhancing immunity to different types of infections varies considerably. The purpose of the present paper is to gain an insight into the effects of vitamin A in modulating immune responses by comparing and contrasting its action during different types of infections, i.e. measles, diarrhoeal disease, respiratory disease, malaria, human immunodeficiency virus

(HIV) infection and tuberculosis. These comparisons suggest that the modulation of immune function by vitamin A is complex and involves many different arms of the immune system (Table 1).

### Measles

Measles virus is a member of the *Morbillivirus* genus of the Paramyxoviridae, a family which includes rinderpest, peste des petits ruminants and canine distemper viruses (Bellini *et al.* 1994). Human subjects are generally considered to be the only natural host of this virus, but other primates can be infected through contacts with infected human subjects. Measles virus is spread from person-to-person via the respiratory route, and viral replication occurs in macrophages in the lymphoid tissue of the respiratory mucosa and lungs (Griffin, 1995). A viraemia allows measles virus to spread to multiple organs, including the skin, liver and conjunctiva, and a prodrome of fever, cough and conjunctivitis occurs approximately 14 d after infection. Antibody responses against measles virus proteins are detectable at the onset of the rash. Infants are protected against measles virus infection by passively acquired maternal antibody to measles, and this is probably the strongest evidence that antibody is involved in protective immunity to measles virus. T-cell-driven cellular immune responses, including activation of CD4+ and CD8+ lymphocytes, occur during measles (Griffin, 1995). Delayed-type hypersensitivity skin test responses and *in*

**Abbreviations:** HIV, human immunodeficiency virus; Ig, immunoglobulin; IL, interleukin; RE, retinol equivalents; Th, T-helper.  
**Corresponding author:** Dr R. D. Semba, fax +1 410 955 0629, email rdsemba@jhmi.edu

**Table 1.** Vitamin A and immunity to infectious diseases

Disease	Immune response	Clinical effect of vitamin A	Effects of vitamin A on immunity	
			Human subjects	Animal models
Measles	Primarily Th2-like response	↓ Morbidity, mortality	↑ IgG against measles ↑ Lymphocyte counts ↓ Measles replication ?	↓ Morbidity, ↑ cytotoxic T-cell responses to NDV in chickens
Diarrhoeal disease	slgA response important; immune response variable	↓ Morbidity, mortality	Unknown	↓ Morbidity, ↑ slgA against rotavirus in mice
Respiratory disease	Motility of cilia; alveolar macrophages	↓ Morbidity, mortality of measles-associated pneumonia, little effect on RSV, ALRI	Unknown	↓ Morbidity, ↑ slgA against influenza A virus in mice
Malaria	Antibody and cell-mediated immune responses, protective immunity poorly defined	↓ Morbidity	Unknown	↓ Morbidity to <i>P. berghei</i> in rats
HIV or AIDS	Antibody and cell-mediated immune responses	↓ Morbidity, mortality	↑ CD4+, NK cell counts	Unknown
Tuberculosis	Primarily Th1-like response	Unknown	Unknown	↓ Morbidity to <i>M. bovis</i> in mice

Th2, T-helper cells type 2; slgA, secretory immunoglobulin A; RSV, respiratory syncytial virus, ALRI, acute lower-respiratory-tract infection, NDV, Newcastle disease virus, *P. berghei*, *Plasmodium berghei*; HIV, human immunodeficiency virus; AIDS, acquired immune deficiency syndrome; NK, natural killer; Th2, T-helper cells type 2; *M. bovis*, *Mycobacterium bovis*; ↑, increase; ↓, decrease.

*in vitro* proliferation of lymphocytes to viral antigens are often minimal or absent in measles infection (Griffin, 1995). Immune suppression often accompanies measles infection and is thought to increase the susceptibility to secondary infections. The immune response to measles is thought to be consistent with T-helper (Th) 2-like immune responses in which antibody responses predominate and are driven by interleukin-(IL) 4, IL-6 and IL-10. Measles virus infection of primary human monocytes has been shown to suppress cell-mediated immunity through down-regulation of IL-12 production (Karp *et al.* 1996).

Some determinants of clinical outcome during measles infection include vitamin A status (Markowitz *et al.* 1989), overcrowding and virus dose (Aaby, 1988), and malnutrition (Alwar, 1992). Clinical trials show that high-dose vitamin A supplementation reduces morbidity and mortality in children with acute measles infection (Barclay *et al.* 1987; Hussey & Klein, 1990; Coutsooudis *et al.* 1991; Ogaro *et al.* 1993). In acute complicated measles, high-dose vitamin A supplementation (60 mg retinol equivalents (RE) on admission and the following day) has been shown to reduce mortality by up to 80 % in Cape Town, South Africa (Hussey & Klein, 1990). Vitamin A supplementation seems to reduce the infectious complications associated with measles immune suppression, such as pneumonia and diarrhoeal disease.

There have been no published animal studies (such as in experimentally-infected primates) which provide directly relevant information on how vitamin A status influences immune responses to measles virus. Canine distemper virus shares some sequences and similar proteins to measles virus, and there is potential for examining the effects of vitamin A status on canine distemper virus infection in dogs. Newcastle disease virus, another virus in the family Paramyxoviridae, is a well-studied animal model which might provide useful inferences for the effects of vitamin A status on immune responses during measles. Vitamin A-deficient chickens infected with Newcastle disease virus show immunological abnormalities such as increased virus shedding, decreased numbers of T and B lymphocytes in

peripheral blood, and impaired cytotoxic T lymphocyte responses against Newcastle disease virus-infected cells (Sijtsma *et al.* 1990; West *et al.* 1991).

There are some findings which suggest how vitamin A supplementation modulates immune responses during measles virus infection. Children with acute measles infection who received high-dose vitamin A supplementation (60 mg RE on admission and the following day) had higher IgG responses to measles virus and higher circulating lymphocyte counts during follow-up compared with children who received placebo (Coutsooudis *et al.* 1992). These results are consistent with the observation that vitamin A supplementation may enhance lymphopoiesis (Göttgens & Green, 1995). In a clinical trial in Indonesia, high-dose vitamin A supplementation increased circulating CD4+ lymphocytes, especially naive CD4+ cells (Semba *et al.* 1993).

Perhaps a clue as to how vitamin A may modulate immunity to measles comes from three studies in which a high-dose vitamin A supplement was administered with live attenuated measles vaccine. In a clinical trial in Indonesia, high-dose vitamin A supplementation (60 mg RE 2 weeks before immunization) was shown to increase IgG responses to tetanus toxoid in preschool children, aged 3–6 years, who received diphtheria–pertussis–tetanus immunization (Semba *et al.* 1992). This observation provided the rationale that high-dose vitamin A supplementation given with measles vaccination might enhance antibody responses to measles virus. In 6-month-old infants in Indonesia, administration of vitamin A (30 mg RE) at the time of immunization with standard titre Schwarz measles vaccine interfered with seroconversion to measles in infants who had maternal antibody present, and reduced the incidence of measles vaccine-associated rash (Semba *et al.* 1995). A separate clinical trial also showed that vitamin A (30 mg RE) reduced antibody responses to measles virus in 9-month-old infants who had maternal antibody present, but did not interfere with overall seroconversion rates to measles (Semba *et al.* 1997).

In a study in Guinea-Bissau, vitamin A supplementation (30 mg RE) was found to enhance geometric mean titres to

measles when given simultaneously with standard titre Schwarz measles vaccine in 9-month-old infants (Stabell Benn *et al.* 1997). In a two-dose measles immunization schedule at ages 6 and 9 months, simultaneous vitamin A supplementation did not interfere with seroconversion to measles when measured at 18 months of age (Stabell Benn *et al.* 1997). It is not possible to determine whether vitamin A supplementation interfered with seroconversion rates after measles vaccine in 6-month-old infants in the study in Guinea-Bissau (Stabell Benn *et al.* 1997) as it did in the study in Indonesia (Semba *et al.* 1995), because antibody titres were not measured until after two doses of measles vaccine. Although the results of the studies in Indonesia and Guinea-Bissau involving 6-month-old infants have been viewed as contradictory (Ross & Cutts, 1997; Stabell Benn *et al.* 1997), the differences in the design of measles vaccine studies lends little validity to making direct comparisons of these two studies. The findings may be complementary (Semba, 1997).

How does vitamin A supplementation enhance immunity to measles? Is there an increase in Th2-like responses with enhancement of antibody titres to measles? Such an effect might be predicted on the basis of a murine model which suggests that vitamin A status influences the balance between Th1- and Th2-like responses (Cantorna *et al.* 1994), and this effect is consistent with the findings of a study of children in South Africa (Coutsoudis *et al.* 1992). Do vitamin A and related retinoids enhance natural killer cell activity against measles virus, as has been suggested in a rat model of vitamin A deficiency (Zhao & Ross, 1995)? Alternatively, does vitamin A have an impact directly on the replication of measles virus? If a retinoic acid response element were present in the measles genome, it might provide a clue as to potential regulation of measles virus replication by vitamin A and related retinoids.

### Diarrhoeal disease

In developing countries, diarrhoeal diseases among children are caused by a wide variety of pathogens, including rotavirus, *Escherichia coli*, and *Shigella*, *Vibrio cholerae*, *Salmonella* and *Entamoeba histolytica*. The epidemiology, clinical features, immunology and pathogenesis of diarrhoea may differ according to characteristics of the pathogen, such as production of toxins, tissue invasion, fluid and electrolyte loss, and location of infection. In general, host defences in the gut include gastric acidity, the presence of normal microflora, gut motility, mucus production, integrity of microvilli, local secretion of antibody and cell-mediated immunity. The development of protective immunity to rotavirus involves the generation of secretory IgA antibodies against rotavirus in the gut and serum (Grimwood *et al.* 1988; Bernstein *et al.* 1989; Coulson, 1998).

Clinical vitamin A deficiency is associated with diarrhoeal disease in children (Brilliant *et al.* 1985; DeSole *et al.* 1987; Gujral *et al.* 1993; Schaumberg *et al.* 1996). A large longitudinal study in Indonesia has shown that children with pre-existing xerophthalmia are at a higher risk of having a subsequent episode of diarrhoeal disease (Sommer *et al.* 1984). Recent large community-based clinical trials of vitamin A supplementation in Tamil Nadu,

Nepal and Ghana show that vitamin A has a major impact on the overall mortality from diarrhoeal disease (Sommer & West, 1996). In a clinical trial in Brazil the severity of diarrhoeal disease was shown to be reduced by vitamin A supplementation (Barreto *et al.* 1994). Urinary losses of vitamin A during *Shigella* infection may be substantial in some children (Mitra *et al.* 1998), and vitamin A supplementation (60 mg RE) has recently been shown to reduce morbidity in children with acute shigellosis (Hossain *et al.* 1998).

Although vitamin A supplementation reduces morbidity and mortality from diarrhoeal disease in general, it is unclear whether vitamin A enhances immunity against all diarrhoeal pathogens or has effects specific for certain organisms. The effect of vitamin A on mucosal immunity in human subjects has not been addressed. The results of autopsy studies of children obtained from the older literature suggest that vitamin A deficiency is associated with alterations in the gut epithelia (Blackfan & Wolbach, 1933; Sweet & K'ang, 1935), but it is likely that other concomitant nutritional abnormalities were present at time of death. A mouse model using epizootic diarrhoea of infant mice rotavirus strain has been used to gain an insight into the effects of vitamin A status on immunity to infection. Vitamin A-deficient mice were much more susceptible to rotavirus infection and had greater pathology in the gut, including destruction of microvilli (Ahmed *et al.* 1990). Following challenge with epizootic diarrhoea of infant mice rotavirus, serum antibody responses against the rotavirus were significantly lower in vitamin A-deficient mice than in control or pair-fed mice (Ahmed *et al.* 1991). The secretory IgA response against rotavirus was not specifically measured in this mouse study; however, studies in mice and human subjects suggest that serum IgA and IgG may be useful indicators of immunity to rotavirus (Grimwood *et al.* 1988; Bernstein *et al.* 1989; McNeal & Ward, 1995).

### Respiratory disease

Acute lower-respiratory-tract infections are a major cause of death among children in developing countries, and major causes of acute lower-respiratory-tract infections include respiratory syncytial virus infection, parainfluenza, *Haemophilus influenzae*, *Streptococcus pneumoniae* and *Bordetella pertussis*. Secondary bacterial infection with high case fatality may follow a primary viral infection in the lungs. Important components of the immune response in the respiratory tract include motility of cilia, mucin production and the activity of alveolar macrophages. The immune response to different respiratory pathogens may vary greatly. For example, immunity to respiratory syncytial virus infection in infants seems to be associated with a Th2-like response characterized by interferon  $\gamma$  and IL-2 production (Roman *et al.* 1997), whereas immunity to whooping cough seems to be associated with a Th1-like response (Ryan *et al.* 1997a, b).

Vitamin A deficiency causes pathological alterations in the epithelium of the respiratory tract, including keratinization and loss of ciliated cells, mucus, and goblet cells. Clinical vitamin A deficiency is associated with lower-respiratory-tract infections (DeSole *et al.* 1987; Tielsch

*et al.* 1986). A large longitudinal study in Indonesia suggested that preschool children with mild xerophthalmia are at an increased risk of having a subsequent episode of respiratory infection (Sommer *et al.* 1984). These natural history studies suggested that vitamin A supplementation might reduce morbidity and mortality from respiratory disease. Community-based clinical trials have attempted to determine whether periodic high-dose vitamin A supplementation reduces the incidence and/or severity of acute respiratory infections in children. Hospital-based clinical trials have addressed whether high-dose vitamin A supplementation can reduce morbidity when given at the time of an acute attack of respiratory disease.

The community-based trials of vitamin A supplementation did not seem to reduce the incidence of acute lower-respiratory-tract infections, although there was an overall beneficial effect in reducing child mortality (The Vitamin A and Pneumonia Working Group, 1995). Vitamin A supplementation, when delivered through childhood immunization programmes, may reduce the duration of acute respiratory infections among infants (Rahman *et al.* 1996). Hospital-based studies have shown that high-dose vitamin A supplementation has no therapeutic effect on the morbidity of acute lower-respiratory-tract infections in children (Kjohede *et al.* 1995; Nacul *et al.* 1997; Fawzi *et al.* 1998a). In Chile and the USA, hospital-based trials showed that vitamin A supplementation had little impact on respiratory syncytial virus infection among infants and young children (Bresee *et al.* 1996; Dowell *et al.* 1996; Quinlan & Hayani, 1996).

Although vitamin A status has been shown to be related to the severity of acute respiratory infection in children (Dudley *et al.* 1997), it is unclear why in some trials vitamin A therapy has no apparent effect on the morbidity of acute respiratory infections among preschool children. Young age might be one contributing factor to the lack of an effect, as large community-based studies suggest that vitamin A supplementation has little effect on morbidity and mortality of infants (West *et al.* 1995; WHO/CHD Immunisation-Linked Vitamin A Supplementation Study Group, 1998). Studies have been conducted also in populations where vitamin A deficiency is not considered a public health problem. In the recent clinical trials involving respiratory syncytial virus infection, the apparent lack of impact of vitamin A supplementation on respiratory syncytial virus infection might be due to the young age of the subjects and the lack of vitamin A deficiency in the population. Other micronutrient deficiencies, such as that of vitamin D and Zn, may have limiting roles in immunity to respiratory infections (Muhe *et al.* 1997; Ruel *et al.* 1997).

An animal model of influenza A virus infection in mice has been used to study the effects of vitamin A deficiency on immune responses. Vitamin A-deficient mice had impaired regeneration of normal respiratory epithelium following infection, and the secretory IgA response in the respiratory tract was decreased by vitamin A deficiency (Stephensen *et al.* 1993). Vitamin A deficiency was associated with decreased salivary IgA responses but increased serum IgG responses against influenza A virus in severely-infected mice (Stephensen *et al.* 1996). The decreased secretory IgA responses against influenza A virus appear to be due to

lower numbers of IgA-secreting plasma cells in vitamin A-deficient mice compared with control mice (Gangopadhyay *et al.* 1996). The studies in this murine model suggest that vitamin A deficiency would impair immunity against viral infections in the respiratory tract, and further research is needed to determine the effects of vitamin A deficiency against respiratory pathogens in human subjects. It would be erroneous to consider vitamin A as ineffective in increasing immunity to acute lower-respiratory-tract infections; vitamin A supplementation has been shown to reduce the life-threatening complication of pneumonia after acute measles infection (Barclay *et al.* 1987; Hussey & Klein, 1990).

### Malaria

Malaria is a mosquito-borne protozoan disease which is caused by *Plasmodium falciparum*, *P. vivax*, *P. malariae* and *P. ovale* in human subjects, of which *P. falciparum* is the most serious, causing an estimated one to two million deaths worldwide each year (Murphy & Oldfield, 1996). In the life cycle of *P. falciparum*, sporozoites are inoculated with the saliva of a biting mosquito into the blood of the human host, and within minutes the sporozoites reach the liver and invade hepatocytes. During the pre-erythrocytic cycle in the liver, a sporozoite can develop into a mature liver schizont, which can burst, releasing thousands of merozoites into the blood. The merozoites infect human erythrocytes and initiate the erythrocytic cycle. Merozoites mature into blood schizonts which burst, producing eight to thirty-two new merozoites that can invade more erythrocytes. Symptoms such as fever and malaise are produced during the infection, rupture and destruction of erythrocytes. Within 10–12 d sexual forms of *P. falciparum* (male and female gametocytes) appear in the peripheral blood and can be taken up by the mosquito during a blood meal. In the mosquito, gametocytes undergo development into zygotes, ookinetes, and oocysts, and eventually sporozoites.

Immunity to malaria occurs over several years of exposure to repeated infection, and it may take years to develop immunity to the full range of antigenic variation of the parasite (Reeder & Brown, 1996; Baird, 1998). Antibody-mediated immunity, cell-mediated responses and non-specific immunity comprise the immune response against *P. falciparum*, and complete immunity against re-infection is not known. Antibody responses are directed against many stages in the life cycle of *P. falciparum*, including circumsporozoite protein, merozoite surface antigens 1 and 2, apical membrane antigen 1 and Pf155/ring-infected erythrocyte surface antigens. The results of a recent cohort study from Ghana suggest that the level of maternal antibody may help protect young infants from infection (Wagner *et al.* 1998); these findings may help to explain the increasing prevalence of clinical *P. falciparum* infection with increasing age during the first year of life. Lymphocyte proliferation responses to various malarial antigens have been observed, but their significance is unclear (Høgh, 1996).

Natural history studies suggest an association between indicators of poor vitamin A status and malaria (Stürchler

*et al.* 1987; Galan *et al.* 1990; Friis *et al.* 1997). A recent randomized placebo-controlled clinical trial was conducted in Papua New Guinea to examine the effects of vitamin A supplementation (60 mg RE every 3 months) on malarial morbidity in preschool children (Shankar *et al.* 1997). Children between 6 and 60 months of age were randomized to receive vitamin A or placebo every 3 months. Weekly morbidity surveillance and clinic-based surveillance were established to monitor acute malaria, and children were followed for 1 year. Vitamin A significantly reduced the incidence of malaria attacks by about 20–50 % for all except extremely high levels of parasitaemia. Similarly, vitamin A supplementation reduced ‘clinic-based’ malaria attacks, which consisted of self-solicited visits to the clinic by mothers who thought that their children should be seen because of fever. Vitamin A supplementation had little impact in children under age 12 months and greatest effect from 13 to 36 months of age.

Although the effects of vitamin A deficiency on susceptibility to malaria have been examined in a rat model (Stoltzfus *et al.* 1989), it is unclear how vitamin A status influences immune responses to malaria. The clinical trial of vitamin A supplementation in Papua New Guinea involved laboratory analyses of antibody responses to malaria antigens, T-cell subsets and lymphocyte proliferation responses to mitogens and malaria antigens, and these studies are currently in progress. Clearly, it will be important to identify underlying immunological mechanisms which might explain the observations of reduced malarial morbidity in children who received vitamin A.

### Human immunodeficiency virus infection

During HIV infection, microenteropathy, malabsorption, steatorrhoea and anorexia are not uncommon, and low plasma or serum vitamin A levels have been described in all stages of HIV infection and among many different risk groups (Semba & Tang, 1999). Plasma or serum levels of vitamin A or intake of vitamin A has been associated with increased disease progression, mortality and higher mother-to-child transmission of HIV (Semba *et al.* 1994; Semba & Tang, 1999). Periodic high-dose vitamin A supplementation seems to reduce morbidity among children born to HIV-infected mothers (Coutsoudis *et al.* 1995), but vitamin A supplementation itself does not appear to influence HIV load in the blood (Semba *et al.* 1998). Providing multi-vitamin supplementation to HIV-infected women during pregnancy reduces fetal mortality and low birth weight by about 40 %, but vitamin A supplementation alone had no significant impact (Fawzi *et al.* 1998c). High-dose vitamin A supplementation appeared to reduce mortality among HIV-infected children who presented with acute lower-respiratory tract infection (Fawzi *et al.* 1998b).

Low plasma vitamin A levels have been associated with T-cell subset abnormalities such as decreased CD4+ lymphocytes and decreased CD3+ CD28+ lymphocytes in injection drug users (Semba *et al.* 1996). In children with acquired immune deficiency syndrome in Cape Town vitamin A supplementation was shown to increase circulating CD4+ lymphocytes and natural killer cells 1 month following high-dose vitamin A supplementation (60 mg RE

on consecutive days; Hussey *et al.* 1996). Again, these observations are consistent with the hypothesis that vitamin A status influences lymphopoiesis. Studies of the effects of vitamin A status on functional immunity have not been conducted in subjects with HIV infection. Although retrovirus infection in mice has been used as a model for HIV infection (Watson *et al.* 1988), this infection is not similar in that the virus infects both B and T lymphocytes in mice rather than CD4+ lymphocytes as it does in HIV infection.

### Tuberculosis

*Mycobacterium tuberculosis* infection is usually spread by the respiratory route, and infection by tubercle bacilli can result in asymptomatic infection or clinical tuberculosis. The vast majority of primary infections do not result in clinical infection; however, in individuals with poor nutritional status, the risk of developing pulmonary or miliary tuberculosis is greatly increased.

Although malnutrition and vitamin A deficiency seem to be major risk factors for the progression of tuberculosis, clinical management usually involves chemoprophylaxis and chemotherapy alone, rather than improvement of host nutritional status. Cod-liver oil, a rich source of vitamins A and D, was used as treatment for tuberculosis for over 100 years. The role of nutrition in tuberculosis remains a major area of neglect, despite the promise that micronutrients have shown as therapeutic agents for other types of infections, and the long record of the use of vitamins A and D for treatment of pulmonary and miliary tuberculosis in both Europe and the USA. A recent clinical trial suggests that high-dose vitamin A supplementation does not influence the morbidity of tuberculosis in children (Hanekom *et al.* 1997). Studies which address the use of multivitamins and minerals or vitamins A plus D as adjunct therapy for tuberculosis have not yet been conducted.

In animal models immunity against *M. tuberculosis* is most consistent with a Th1-like response, with predominance of interferon  $\gamma$  and IL-12 (Hernández-Pando *et al.* 1996; Cooper *et al.* 1997). Cytotoxic T lymphocytes (Lalvani *et al.* 1998) and natural killer cells (Yoneda & Ellner, 1998) also appear to play an important role in resistance to *M. tuberculosis*. Macrophages are a key element in host resistance to tubercle bacilli. Activated macrophages destroy tubercle bacilli, and whether a tuberculous lesion progresses or regresses is largely dependent on the ability of macrophages which enter the granulation tissue to ingest and destroy bacilli (Dannenberg, 1989).

Vitamin A-deficient animals have decreased resistance to experimental infection with *Mycobacterium* spp. Among animals inoculated with virulent tubercle bacilli, mortality was higher among vitamin A-deficient rats than rats on a normal diet (Gloyne & Page, 1921). In another study of experimental tuberculosis infection, vitamin A-deficient rats had more severe infection and more extensive lesions on histopathology than control rats (Sriramachari & Gopalan, 1958). In addition, rats given adequate vitamin A but restricted in protein or energy were not more susceptible to experimental tuberculosis infection, suggesting that increased susceptibility to tuberculosis was attributable to lack of vitamin A rather than protein–energy malnutrition.

Vitamin A-deficient mice have a more severe infection with *M. bovis* compared with mice supplemented with dietary  $\beta$ -carotene (Finkelstein, 1932). Chicks fed on a high-vitamin A diet had 26 % increased survival compared with chicks fed on a normal diet following experimental infection with *M. tuberculosis* (Solotorovsky *et al.* 1961). Production of IL-1 by macrophages is dependent on vitamin A status in experimental animal models (Moriguchi *et al.* 1985). Dietary vitamin A enhances T-cell proliferation responses and antibody responses to *M. tuberculosis* antigen in the chick model (Sklan *et al.* 1994). Vitamin A-supplemented mice challenged with *M. bovis* and treated with IL-2 have strong delayed-type hypersensitivity responses and proliferation responses to purified protein derivative compared with control mice without supplementary vitamin A (Ferraro *et al.* 1988).

### Conclusions

The available information suggests that immune modulation by vitamin A during infections may depend on the pathogen or pathogens involved. The type of antigens, the level of T-cell activation and the local cytokine environment may all influence the modulation of immune response following vitamin A supplementation. As seems apparent from examination of different diseases, the modulation of immune responses by vitamin A does not seem to fit a clear Th1 v. Th2 type of pattern. During measles infection vitamin A seems to be enhancing Th2-like responses with potentiation of antibody responses to measles virus. The use of cod-liver oil for treatment of tuberculosis suggests that this therapy may have an effect on Th1-like immune responses against *M. tuberculosis*, but it may be difficult to attribute these effects to vitamin A alone, as cod-liver oil is a rich source of vitamins A and D and other nutrients. One study suggests that vitamin A supplementation enhances delayed-type hypersensitivity skin responses in infants (Rahman *et al.* 1997), consistent with modulation of Th1-like responses. In human subjects, it has been somewhat difficult to study immune modulation by vitamin A in certain disease states, such as respiratory and diarrhoeal disease, given the wide variety of pathogens involved in such infections. Measles is probably the most well-defined illness for the study of immune modulation by vitamin A. Further studies are needed to elucidate the ways in which vitamin A enhances immunity to malaria. A large gap remains between studies of immune modulation by vitamin A and its metabolites in cell lines and studies of immune modulation by vitamin A in human subjects (Semba, 1998).

### Acknowledgements

This work was supported in part by the National Institute of Health (HD30042, HD32247, AI41956), the Fogarty International Center and the United States Agency for International Development (Cooperative Agreement HRN-A-00-97-00015-00).

### References

- Aaby P (1988) Malnutrition and overcrowding/intensive exposure in severe measles infection: review of community studies. *Reviews of Infectious Disease* **10**, 478–491.
- Ahmed F, Jones DB & Jackson AA (1990) The interaction of vitamin A deficiency and rotavirus infection in the mouse. *British Journal of Nutrition* **63**, 363–373.
- Ahmed F, Jones DB & Jackson AA (1991) Effect of vitamin A deficiency on the immune response to epizootic diarrhoea of infant mice (EDIM) rotavirus infection in mice. *British Journal of Nutrition* **65**, 475–485.
- Alwar AJE (1992) The effect of protein energy malnutrition on morbidity and mortality due to measles at Kenyatta National Hospital, Nairobi, Kenya. *East African Medical Journal* **69**, 415–418.
- Baird JK (1998) Age-dependent characteristics of protection v. susceptibility to *Plasmodium falciparum*. *Annals of Tropical Medicine and Parasitology* **92**, 367–390.
- Barclay AJG, Foster A & Sommer A (1987) Vitamin A supplements and mortality related to measles: a randomised clinical trial. *British Medical Journal* **323**, 160–164.
- Barreto ML, Santos LMP, Assis AMO, Purificação M, Araújo N, Farenzena GG, Santos PAB & Fiaccone RL (1994) Effect of vitamin A supplementation on diarrhoea and acute lower-respiratory-tract infections in young children in Brazil. *Lancet* **344**, 228–231.
- Beaton GH, Martorell R, L'Abbe KA, Edmonston B, McCabe G, Ross AC & Harvey B (1993) *Effectiveness of Vitamin A Supplementation in the Control of Young Child Morbidity and Mortality in Developing Countries*. ACC/SCN State-of-the-Art Nutrition Policy Discussion Paper no. 13. Geneva: UN.
- Bellini WJ, Rota JS & Rota PA (1994) Virology of measles virus. *Journal of Infectious Diseases* **170**, Suppl. 1, S15–S23.
- Bernstein DI, McNeal MM, Schiff GM & Ward RL (1989) Induction and persistence of local rotavirus antibodies in relation to serum antibodies. *Journal of Medical Virology* **28**, 90–95.
- Blackfan KD & Wolbach SG (1933) Vitamin A deficiency in infants: a clinical and pathological study. *Journal of Pediatrics* **3**, 679–706.
- Bresee JS, Fischer M, Dowell SF, Johnston BD, Biggs VM, Levine RS, Lingappa JR, Keyserling HL, Petersen KM, Bak JR, Gary HE, Sowell AL, Rubens CE & Anderson LJ (1996) Vitamin A therapy for children with respiratory syncytial virus infection: a multicenter trial in the United States. *Pediatric Infectious Diseases Journal* **15**, 777–782.
- Brilliant LB, Pokhrel RP, Grasset NC, Lepkowski JM, Kolstad A, Hawks W, Pararajasegaram R, Brilliant GE, Gilbert S, Shrestha SR & Kuo J (1985) Epidemiology of blindness in Nepal. *Bulletin of the World Health Organization* **63**, 375–386.
- Cantorna MT, Nashold FE & Hayes CE (1994) In vitamin A deficiency multiple mechanisms establish a regulatory T helper cell imbalance with excess Th1 and insufficient Th2 function. *Journal of Immunology* **152**, 1515–1522.
- Cooper AM, Magram J, Ferrante J & Orme IM (1997) Interleukin 12 (IL-12) is crucial to the development of protective immunity in mice intravenously infected with *Mycobacterium tuberculosis*. *Journal of Experimental Medicine* **186**, 39–45.
- Coulson BS (1998) Longitudinal studies of neutralizing antibody responses to rotavirus in stools and sera of children following severe rotavirus gastroenteritis. *Clinical and Diagnostic Laboratory Immunology* **5**, 897–901.
- Coutsoudis A, Bobat RA, Coovadia HM, Kuhn L, Tsai WY & Stein ZA (1995) The effects of vitamin A supplementation on the morbidity of children born to HIV-infected women. *American Journal of Public Health* **85**, 1076–1081.

- Coutsoudis A, Broughton M & Coovadia HM (1991) Vitamin A supplementation reduces measles morbidity in young African children: a randomized, placebo-controlled, double-blind trial. *American Journal of Clinical Nutrition* **54**, 890–895.
- Coutsoudis A, Kiepiela P, Coovadia HM & Broughton M (1992) Vitamin A supplementation enhances specific IgG antibody levels and total lymphocyte numbers while improving morbidity in measles. *Pediatric Infectious Diseases Journal* **11**, 203–209.
- Dannenberg AM Jr (1989) Immune mechanisms in the pathogenesis of pulmonary tuberculosis. *Reviews of Infectious Diseases* **11**, Suppl. 2, S369–S378.
- DeSole G, Belay Y & Zegeye B (1987) Vitamin A deficiency in southern Ethiopia. *American Journal of Clinical Nutrition* **45**, 780–784.
- Dowell SF, Papis Z, Bresee JS, Larrañaga C, Mendez M, Sowell AL, Gary HE, Anderson LJ & Avendaño LF (1996) Treatment of respiratory syncytial virus infection with vitamin A: a randomized, placebo-controlled trial in Santiago, Chile. *Pediatric Infectious Diseases Journal* **15**, 782–786.
- Dudley L, Hussey G, Huskissen J & Kessow G (1997) Vitamin A status, other risk factors and acute respiratory infection morbidity in children. *South African Medical Journal* **87**, 65–70.
- Fawzi WW, Mbise RL, Fataki MR, Herrera MG, Kawau F, Hertzmark E, Spiegelman D & Ndossi G (1998a) Vitamin A supplementation and severity of pneumonia in children admitted to the hospital in Dar es Salaam, Tanzania. *American Journal of Clinical Nutrition* **68**, 187–192.
- Fawzi W, Mbise FL, Kawau F, Herrera MG, Hertzmark E, Fataki M & Spiegelman D (1998b) Vitamin A supplements and mortality among HIV positive and negative children in Tanzania. *Abstracts of the 12<sup>th</sup> World AIDS Conference, Geneva, June 28–July 3, 1998*, p. 840, Abstr. 170/42331, Geneva: 12th World Aids Conference Committee.
- Fawzi WW, Msamanga GI, Spiegelman D, Urassa EJN, McGrath N, Mwakagile D, Antelman G, Mbise R, Herrera G, Kapiga S, Willett W, Hunter DJ & Tanzania Vitamin and HIV Infection Trial Team (1998c) Randomised trial of effects of vitamin supplements on pregnancy outcomes and T cell counts in HIV-1-infected women in Tanzania. *Lancet* **351**, 1477–1482.
- Ferraro F, Mattei M & Colizzi V (1988) Low doses of recombinant interleukin-2 enhanced delayed type hypersensitivity to PPD in mice infected with *Mycobacterium bovis*-BCG and fed a diet supplemented with vitamin A acetate. *Zeitschrift für Erkrankungen der Atmungsorgane* **171**, 45–49.
- Finkelstein MH (1932) Effect of carotene on course of *B. tuberculosis* infection of mice fed on a vitamin A deficient diet. *Proceedings of the Society for Experimental Biology and Medicine* **29**, 69–71.
- Friis H, Mwaniki D, Omondi B, Muniu E, Magnussen P, Geissler W, Thiong'o F & Michaelsen KF (1997) Serum retinol concentrations and *Schistosoma mansoni* intestinal helminths, and malarial parasitemia: a cross-sectional study in Kenyan preschool and primary school children. *American Journal of Clinical Nutrition* **66**, 665–671.
- Galan P, Samba C, Luzeau R & Amedee-Manesme O (1990) Vitamin A deficiency in preschool age Congolese children during malaria attacks. Part 2: impact of parasitic disease on vitamin A status. *International Journal of Vitamin and Nutrition Research* **60**, 224–228.
- Gangopadhyay NN, Moldoveanu Z & Stephensen CB (1996) Vitamin A deficiency has different effects on immunoglobulin A production and transport during influenza A infection in BALB/c mice. *Journal of Nutrition* **126**, 2960–2967.
- Gloyne SR & Page DS (1921) A preliminary note on the relationship between deficiency in vitamin A and tuberculosis. *Tubercle* **3**, 577–579.
- Göttgens B & Green AR (1995) Retinoic acid and the differentiation of lymphohaemopoietic stem cells. *Bioessays* **17**, 187–189.
- Griffin DE (1995) Immune responses during measles virus infection. *Current Topics in Microbiology and Immunology* **191**, 117–134.
- Grimwood K, Lund JC, Coulson BS, Hudson IL, Bishop RF & Barnes GL (1988) Comparison of serum and mucosal antibody responses following severe acute rotavirus gastroenteritis in young children. *Journal of Clinical Microbiology* **26**, 732–738.
- Gujral S, Abbi R & Gopaldas T (1993) Xerophthalmia, vitamin A supplementation and morbidity in children. *Journal of Tropical Pediatrics* **39**, 89–92.
- Hanekom WA, Potgieter S, Hughes EJ, Malan H, Kessow G & Hussey GD (1997) Vitamin A status and therapy in childhood pulmonary tuberculosis. *Journal of Pediatrics* **131**, 925–927.
- Hernández-Pando R, Orozco H, Sampieri A, Pavón L, Velasquillo C, Larriva-Sahd J, Alcocer JM & Madrid MV (1996) Correlation between the kinetics of Th1/Th2 cells and pathology in a murine model of experimental pulmonary tuberculosis. *Immunology* **89**, 26–33.
- Høgh B (1996) Clinical and parasitological studies on immunity to *Plasmodium falciparum* malaria in children. *Scandinavian Journal of Infectious Diseases* **102**, Suppl., 1–53.
- Hossain S, Biswas R, Kabir I, Sarker S, Dibley M, Fuchs G & Mahalanabis D (1998) Single dose vitamin A treatment in acute shigellosis in Bangladeshi children: randomised double blind controlled trial. *British Medical Journal* **316**, 422–426.
- Hussey G, Hughes J, Potgieter S, Kessow G, Burgess J, Beatty D, Keraan M & Carlesle E (1996) Vitamin A status and supplementation and its effects on immunity in children with AIDS. In *Abstracts of the XVII International Vitamin A Consultative Group Meeting, Guatemala City*, p. 6. Washington, DC: International Life Sciences Institute.
- Hussey GD & Klein M (1990) A randomized, controlled trial of vitamin A in children with severe measles. *New England Journal of Medicine* **323**, 160–164.
- Karp CL, Wysocka M, Wahl LM, Ahearn JM, Cuomo PJ, Sherry B, Trinchieri G & Griffin DE (1996) Mechanism of suppression of cell-mediated immunity by measles virus. *Science* **273**, 228–231.
- Kjølhed CL, Chew FJ, Gadowski AM & Marroquin DP (1995) Clinical trial of vitamin A as adjuvant treatment for lower respiratory tract infections. *Journal of Pediatrics* **126**, 807–812.
- Lalvani A, Brookes R, Wilkinson RJ, Malin AS, Pathan AA, Andersen P, Dockrell H, Pasvol G & Hill AVS (1998) Human cytolytic and interferon  $\gamma$ -secreting CD8<sup>+</sup> T lymphocytes specific for *Mycobacterium tuberculosis*. *Proceedings of the National Academy of Sciences USA* **95**, 270–275.
- McNeal MM & Ward RL (1995) Long-term production of rotavirus antibody and protection against reinfection following a single infection of neonatal mice with murine rotavirus. *Virology* **211**, 474–480.
- Markowitz L, Nzilambi N, Driskell WJ, Sension MG, Rovira EZ, Nieburg P & Ryder RW (1989) Vitamin A levels and mortality among hospitalized measles patients, Kinshasa, Zaire. *Journal of Tropical Pediatrics* **35**, 109–112.
- Mitra AK, Alvarez JO, Guay-Woodford L, Fuchs GJ, Wahed MA & Stephensen CB (1998) Urinary retinol excretion and kidney function in children with shigellosis. *American Journal of Clinical Nutrition* **68**, 1095–1103.
- Moriguchi S, Werner L & Watson RR (1985) High dietary vitamin A (retinyl palmitate) and cellular immune functions in mice. *Immunology* **56**, 169–177.
- Muhe L, Lulseged S, Mason KE & Simoes EAF (1997) Case-control study of the role of nutritional rickets in the risk

- of developing pneumonia in Ethiopian children. *Lancet* **349**, 1801–1804.
- Murphy GS & Oldfield EC (1996) Falciparum malaria. *Infectious Disease Clinics of North America* **10**, 747–775.
- Nacul LC, Kirkwood BR, Arthur P, Morris SS, Magalhães M & Fink MCDS (1997) Randomised, double blind, placebo controlled clinical trial of efficacy of vitamin A treatment in non-measles childhood pneumonia. *British Medical Journal* **315**, 505–510.
- Ogato FO, Orinda VA, Onyango FE & Black RE (1993) Effect of vitamin A on diarrhoeal and respiratory complications of measles. *Tropical and Geographical Medicine* **45**, 283–286.
- Quinlan KP & Hayani KC (1996) Vitamin A and respiratory syncytial virus infection. Serum levels and supplementation trial. *Archives of Pediatric and Adolescent Medicine* **150**, 25–30.
- Rahman MM, Mahalanabis D, Alvarez JO, Wahed MA, Islam MA & Habte D (1997) Effect of early vitamin A supplementation on cell-mediated immunity in infants younger than 6 mo. *American Journal of Clinical Nutrition* **65**, 144–148.
- Rahman MM, Mahalanabis D, Alvarez JO, Wahed MA, Islam MA, Habte D & Khaled MA (1996) Acute respiratory infections prevent improvement of vitamin A status in young infants supplemented with vitamin A. *Journal of Nutrition* **126**, 628–633.
- Reeder JC & Brown GV (1996) Antigenic variation and immune evasion in *Plasmodium falciparum* malaria. *Immunology and Cell Biology* **74**, 546–554.
- Roman M, Calhoun WJ, Hinton KL, Avendano LF, Simon V, Escobar AM, Gaggero A & Diaz PV (1997) Respiratory syncytial virus infection in infants is associated with predominant Th-2-like response. *American Journal of Respiratory and Critical Care Medicine* **156**, 190–195.
- Ross DA & Cutts FT (1997) Vindication of policy of vitamin A with measles vaccination. *Lancet* **350**, 81–82.
- Ruel MT, Rivera JA, Santizo MC, Lonnerdal B & Brown KH (1997) Impact of zinc supplementation on morbidity from diarrhea and respiratory infections among Guatemalan children. *Pediatrics* **99**, 808–813.
- Ryan M, Gothefors L, Storsaeter J & Mills KH (1997a) Bordetella pertussis-specific Th1/Th2 cells generated following respiratory infection or immunization with an acellular vaccine: comparison of the T cell cytokine profiles in infants and mice. *Developments in Biological Standardization* **89**, 297–305.
- Ryan M, Murphy G, Gothefors L, Nilsson L, Storsaeter J & Mills KH (1997b) Bordetella pertussis respiratory infection in children is associated with preferential activation of type 1 T helper cells. *Journal of Infectious Diseases* **175**, 1246–1250.
- Schaumburg DA, O'Connor J & Semba RD (1996) Risk factors for xerophthalmia in the Republic of Kiribati. *European Journal of Clinical Nutrition* **50**, 761–764.
- Semba RD (1997) Vitamin A supplementation. *Lancet* **350**, 1031–1032.
- Semba RD (1998) The role of vitamin A and related retinoids in immune function. *Nutrition Reviews* **56**, S38–S48.
- Semba RD, Akib A, Beeler J, Munasir Z, Permaesih D, Muherdiyantiningsih, Komala, Martuti S & Muhilal (1997) Effect of vitamin A supplementation on measles vaccination in nine-month-old infants. *Public Health* **111**, 245–247.
- Semba RD, Lyles CM, Margolick JB, Caiaffa WT, Farzadegan H, Cohn S & Vlahov D (1998) Vitamin A supplementation and human immunodeficiency virus load in injection drug users. *Journal of Infectious Diseases* **177**, 611–616.
- Semba RD, Miotti PG, Chipphangwi JD, Saah AJ, Canner JK, Dallabetta GA & Hoover DR (1994) Maternal vitamin A deficiency and mother-to-child transmission of HIV-1. *Lancet* **343**, 1593–1597.
- Semba RD, Muhilal, Scott AL, Natadisastra G, Wirasasmita S, Mele L, Ridwan E, West KP Jr & Sommer A (1992) Depressed immune response to tetanus in children with vitamin A deficiency. *Journal of Nutrition* **122**, 101–107.
- Semba RD, Muhilal, Ward BJ, Griffin DE, Scott AL, Natadisastra G, West KP Jr & Sommer A (1993) Abnormal T-cell subset proportions in vitamin A-deficient children. *Lancet* **341**, 5–8.
- Semba RD, Munasir Z, Beeler J, Akib A, Muhilal, Audet S & Sommer A (1995) Reduced seroconversion to measles in infants given vitamin A with measles vaccination. *Lancet* **345**, 1330–1332.
- Semba RD, Park S, Royal W & Griffin DE (1996) Vitamin A deficiency and T-cell subpopulations in HIV-infected adults. *Nutrition Research* **16**, 915–923.
- Semba RD & Tang AM (1999) Micronutrients and the pathogenesis of human immunodeficiency virus infection. *British Journal of Nutrition* **81**, 181–189.
- Shankar AH, Genton B, Semba RD, Tielsch J & West KP Jr (1997) Vitamin A supplementation as a nutrient-based intervention to reduce malaria-related morbidity. In *Abstracts of the XVIII International Vitamin A Consultative Group Meeting, Cairo, Egypt*, p. 31. Washington, DC: The Human Nutrition Institute.
- Sijtsma SR, Rombout JHWM, van der Zijpp AJ & West CE (1990) Vitamin A deficiency impairs cytotoxic T lymphocyte activity in Newcastle disease virus-infected chickens. *Veterinary Immunology and Immunopathology* **26**, 191–201.
- Sklan D, Melamed D & Friedman A (1994) The effect of varying levels of dietary vitamin A on immune response in the chick. *Poultry Science* **73**, 843–847.
- Solotorovsky M, Squibb RL, Wogan GN, Siegel H & Gala R (1961) The effect of dietary fat and vitamin A on avian tuberculosis in chicks. *American Review of Respiratory Diseases* **84**, 226–235.
- Sommer A, Katz J & Tarwotjo I (1984) Increased risk of respiratory disease and diarrhea in children with preexisting mild vitamin A deficiency. *American Journal of Clinical Nutrition* **40**, 1090–1095.
- Sommer A & West KP Jr (1996) *Vitamin A Deficiency: Health, Survival, and Vision*. New York: Oxford University Press.
- Sriramachari S & Gopalan C (1958) Nutrition and tuberculosis: effect of some nutritional factors on resistance to tuberculosis. *Indian Journal of Medical Research* **46**, 105–112.
- Stabell Benn C, Aaby P, Balé C, Olsen J, Michaelsen KF, George E & Whittle H (1997) Randomised trial of effect of vitamin A supplementation on antibody response to measles vaccine in Guinea-Bissau, west Africa. *Lancet* **350**, 101–105.
- Stephensen CB, Blount SR, Schoeb TR & Park JY (1993) Vitamin A deficiency impairs some aspects of the host response to influenza A virus infection in BALB/c mice. *Journal of Nutrition* **123**, 823–833.
- Stephensen CB, Moldaveanu Z & Gangopadhyay NN (1996) Vitamin A deficiency diminishes the salivary immunoglobulin A response and enhances the serum immunoglobulin G response to influenza A virus infection in BALB/c mice. *Journal of Nutrition* **126**, 94–102.
- Stoltzfus RJ, Jalal F, Harvey PWJ & Nesheim MC (1989) Interactions between vitamin A deficiency and *Plasmodium berghei* infection in the rat. *Journal of Nutrition* **119**, 2030–2037.
- Stürchler D, Tanner M, Hanck A, Betschart B, Gautschi K, Weiss N, Burnier E, Del Giudice G & Degrémont A (1987) A longitudinal study on relations of retinol with parasitic infections and the immune response in children of Kikawwila village, Tanzania. *Acta Tropica* **44**, 213–227.
- Sweet LK & K'ang HJ (1935) Clinical and anatomic study of avitaminosis A among the Chinese. *American Journal of Diseases of Childhood* **50**, 699–734.



- The Vitamin A and Pneumonia Working Group (1995) Potential interventions for the prevention of childhood pneumonia in developing countries: a meta-analysis of data from field trials to assess the impact of vitamin A supplementation on pneumonia morbidity and mortality. *Bulletin of the World Health Organization* **73**, 609–619.
- Tielsch JM, West KP, Katz J, Chirambo MC, Schwab L, Johnson GJ, Tizazu T, Swartwood J & Sommer A (1986) Prevalence and severity of xerophthalmia in southern Malawi. *American Journal of Epidemiology* **124**, 561–586.
- Wagner G, Koram K, McGuinness D, Bennett S, Nkrumah F & Riley E (1998) High incidence of asymptomatic malaria infections in a birth cohort of children less than one year of age in Ghana, detected by multicopy gene polymerase chain reaction. *American Journal of Tropical Medicine and Hygiene* **59**, 115–123.
- Watson RR, Yahya MD, Darban HR & Prabhala RH (1988) Enhanced survival by vitamin A supplementation during a retrovirus infection causing murine AIDS. *Life Sciences* **43**, 13–18.
- West CE, Rombout JHWM, van der Zijpp AJ & Sijtsma SR (1991) Vitamin A and immune function. *Proceedings of the Nutrition Society* **50**, 251–262.
- West KP Jr, Katz J, Shrestha SR, LeClerq SC, Khatri SK, Pradhan EK, Adhikari R, Wu LS, Pokhrel RP & Sommer A (1995) Mortality of infants < 6 mo of age supplemented with vitamin A: a randomized, double-masked trial in Nepal. *American Journal of Clinical Nutrition* **62**, 143–148.
- WHO/CHD Immunisation-Linked Vitamin A Supplementation Study Group (1998) Randomised trial to assess benefits and safety of vitamin A supplementation linked to immunisation in early infancy. *Lancet* **352**, 1257–1263.
- Yoneda T & Ellner JJ (1998) CD4+ T cell and natural killer cell-dependent killing of *Mycobacterium tuberculosis* by human monocytes. *American Journal of Respiratory and Critical Care Medicine* **158**, 395–403.
- Zhao Z & Ross AC (1995) Retinoic acid repletion restores the number of leukocytes and their subsets and stimulates natural cytotoxicity in vitamin A-deficient rats. *Journal of Nutrition* **125**, 2064–2073.

