Vitamin A and Measles

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Introduction

Measles is nearly forgotten in many western countries where immunization against the disease is universal. In developing countries, it is still a major problem not only because of incomplete immunization coverage, but also because of case-fatality rates as high as 30%. The high impact of the disease can be attributed to poor nutrition status, particularly with respect to vitamin A. In a previous Marabou Symposium, Alfred Sommer[1] addressed the effect of vitamin A on sight and life, whereas in this paper attention will be directed toward the consequences of measles, the relationship between measles and nutrition status with particular reference to vitamin A, the effect of vitamin A supplementation on morbidity and mortality from measles, and the effect of vitamin A supplementation on the efficacy of measles vaccination. The use of an animal model in which vitamin A–deficient chickens are infected with Newcastle disease virus will then be discussed with respect to vitamin A status, morbidity, and host defense mechanisms, both nonspecific and specific.

Characteristics of Measles Infection

Measles virus infects epithelial tissues, particularly of the respiratory tract where it results in the characteristic Koplik's spots of the buccal mucosa, cough, and pneumonia involving the lungs. Infection also affects the gastrointestinal tract resulting in stomatitis and diarrhea, whereas in the genitourinary tract it produces general irritation. In the skin, measles virus infection produces generalized maculopapular eruption and, if the infection is severe, gross desquamation. Infection of the eye results in measles-eye disease and infection of the middle ear causes otitis media. Beyond the epithelial tissues, measles infection results in a general viremia that involves a wide range of tissues and organs. In fact, the principal reason that measles immunization was introduced in developed countries was to counteract encephalomyelitis, which becomes clinically apparent 5 days after the maculopapular eruption. This occurs in 1 in 1000 of those developing measles and is fatal in 10% of such cases. The viremia is also responsible for reduced hepatic protein synthesis, fever, and reduced immunocompetance leading to secondary viral and bacterial infections.

Before the general nutrition status of European children reached the high level it is today, measles infection was something to be feared. As reviewed by Morley and colleagues,[2] measles accounted for 11% of all deaths in Glasgow in the years 1807–1812. Case fatality rates were high. For example, during the years 1867–1872 in a Paris orphanage, the Hospice des Enfants Assistés, 612 of the 1256 (49%) children who developed measles died. However, in London fever hospitals during the years 1911–1914, the measles case fatality rate in children younger than 5 years of age was 14% (Table 1). Hospital admissions peaked in the second year of life when the case fatality rate was greater than 20%. Fifty years later in Ilesha, an area in the rain forest of Nigeria 120 km northeast of Ibadan, hospital admissions during the years 1958–1961 also peaked in the second year of life (Table 1), possibly because maternal antibodies against measles last for 6 months postpartum. Case fatality rates in Ilesha remained greater than 20% for children throughout the first 5 years of life. During the last century, the burden of measles has dropped remarkably in Europe. In Glasgow, 14.2% of children younger than 5 years of age contracted measles in 1908 and had a mortality rate of 5.8% (Table 1). Even in the absence of a vaccine, by 1960, notification of childhood measles in England and Wales was only 2.4% and mortality fell to 0.030%, which is 1:200 of the 1908 Glasgow mortality rate. With virtually universal measles vaccination, measles is a disease that many young doctors have never seen in developed countries. It remains a problem, however, in many developing countries, particularly in Africa, where vaccination rates are in need of improvement.

One consequence of measles that has received particular attention is postmeasles eye lesions. In a study carried out in children admitted to five hospitals in the Dar es Salaam area of Tanzania with measles, Pepping[3] observed measles keratitis, or superficial punctate keratitis,

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Table 1. Comparative Mortality from Measles in the United Kingdom and West Africa

Age (years)	London Fever Hospitals 1911–1914		Ilesha, Nigeria 1958–1961		Glasgow, 1908		England and Wales, 1960	
	Hospital Admissions	Case Fatality Rate (%)	Hospital Admissions	Case Fatality Rate (%)	Notifications (%)	Mortality (%)	Notifications (%)	Mortality (%)
<1	1080	22.9	276	19.9	7.3	11.7	0.78	0.104
1–	2728	21.3	521	29.9	14.2	14.2	2.13	0.045
2–	2125	11.8	217	26.7	17.4	4.2	2.74	0.046
3–	1874	7.1	138	21.7	16.6	1.7	3.04	0.010
4–5	1470	5.2	72	23.6	16.6	1.2	3.24	0.005
Under 5	9277	13.9	1224	25.8	14.2	5.8	2.36	0.030

Source: Morley et al.[2]

in 34% of children and xerophthalmia-associated lesions in 8.5% (Table 2). The children with xerophthalmia-associated lesions had serum retinol concentrations (0.23 µmol/L) that were significantly lower than those with no eye lesions (0.38 µmol/L) or measles-associated eye lesions (0.39 µmol/L). The mean serum retinol concentrations in all children with measles were particularly low when compared with the cutoff value for vitamin A deficiency (0.35 µmol/L). This indicated that measles infection reduced serum vitamin A concentrations. It is interesting to note that in the Dar es Salaam study, in which the case fatality rate was 7.5%, 63% of children had respiratory tract infections and 65% had diarrhea, including 32% who had both conditions simultaneously. Foster and Sommer[4] further investigated the etiology of corneal ulceration following measles in 48 children in Tanzania. They found that 6 (12.5%) of the children had measles keratitis, whereas 24 (50%) had serum retinol concentrations <0.35 µmol/L, again indicating a role of measles in depressing serum retinol concentrations. Herpes simplex infection was evident in 10 (20.8%) of the children indicating that measles compromised the immune system and allowed secondary infection. Unfortunately, the remaining 8 (16.7%) children were administered traditional eye medicines that caused corneal ulceration.

The particular effect of measles on serum retinol concentrations was investigated by Inua et al.[5] in a cross-sectional study in children under the age of 3 in Zaria, the guinea savannah area of northern Nigeria (Table 3). Measles infection depressed serum retinol concentrations by more than 30% and the effect was more pronounced in malnourished children. Malnutrition per se had a more pronounced effect on serum albumin concentrations. Similar effects were observed by Reddy et al.[6] in a cross-sectional study in Hyderabad, India. Whereas control children had serum retinol concentrations of 0.63 µmol/L, during infection values of 0.40 µmol/L were recorded. After recovery, however, the mean serum retinol concentration was 0.69 µmol/L. At all times, serum albumin concentrations remained in the range 34–36 g/L. In a 6-month follow-up of children in the same area, Reddy et al.[6] observed that children who contracted measles had a higher incidence of bronchopneumonia (34%) and diarrhea (77%) than those who did not (6% and 49%, respectively), thus reemphasizing involvement of the respiratory and gastrointestinal tracts in measles infection. It is interesting to note that low levels

Table 2. Eye Lesions Observed in Children Hospitalized for Measles in Dar es Salaam, Tanzania (n=479)

	Eye Lesions	
	Total/Bilateral, n	%
No eye lesions	230	48.0
Measles-associated lesions		
Measles keratitis	136/120	28.4
Superficial punctate keratitis	25/21	5.2
Broken tear film	10/9	2.1
Absence of watery tears	118/118	26.7
Xerophthalmia-associated eye lesions		
Corneal ulceration/keratomalacia	15/6	3.1
Corneal xerosis	21/16	4.4
Conjunctival xerosis	4/4	0.8
Bitot's spots	1/0	0.2
Pigmented lateral triangle	18/18	3.8

Source: Data from Pepping.[3]

Table 3. Measles Compromises Nutrition Status: Cross-sectional Data from Children Under 3 Years of Age in Zaria, Nigeria

	n	Retinol[a]	Albumin
Well-nourished			
No measles	34	100[b]	100[b]
Measles	29	69±4[c]	88±8[c]
Malnourished			
No measles	24	78±9[d]	70±18[d]
Measles	33	62±6[e]	68±12[d]

[a]Concentrations in serum expressed as proportion (%) of values in well-nourished children (>80% weight-for-age) without measles. Malnourished children had weight-for-age <70%. Mean ± SE. Values in the same column not sharing the same superscript are significantly different from one another: P<0.001 (Students' t-test).

Source: Data from Inua et al.[5]

of serum retinol are also associated with increased severity of measles in the United States.[7–9]

Vitamin A Supplementation and Measles Morbidity and Mortality

A number of trials were published regarding the effect of vitamin A supplementation on measles morbidity and mortality. The first of the recent trials, published in 1987, was carried out by Barclay et al.[10] in Mvumi, Tanzania. The relative risk of mortality in children less than 2 years of age was reduced significantly by a factor of 8 when supplemented with vitamin A (200,000 IU) upon hospital admission for measles and again the following day when compared with children who received a placebo on the 2 days. Mortality in those less than the age of 5 years was reduced from 13.0% to 6.8%, although this difference did not reach statistical significance.

Two hospital-based trials were subsequently carried out in South Africa and India. The first of these from Cape Town was reported in 1990.[11] The design was similar to the Tanzanian study with the following exclusion criteria: receipt of a vitamin A dose, xerophthalmia (no cases seen), or a measles rash for more than 4 days. The study population was young (two-thirds of the subjects younger than one year of age) and severely vitamin A–deficient (approximately 50% of children having serum vitamin A concentrations <0.35 μmol/L). Mortality was reduced significantly from 10.3% to 2.2% and morbidity from pneumonia, diarrhea, postmeasles croup, and herpes stomatitis was also markedly reduced in the vitamin A–supplemented group (Table 4). Significant reductions in morbidity and in mortality were also observed in studies carried out in Durban, South Africa[12] and Ahnedabad, India[13] but not in Zambia[14] or Ghana.[15] In the Zambian study, measles cases received just one dose of vitamin A, whereas in the Gambia, the incidence and severity of measles were examined after children were dosed with vitamin A or a placebo. It is therefore important to provide children who develop measles with vitamin A upon the first contact and on the subsequent day.

When the results of these studies were published, a 1932 London study by Ellison[16] came to light. Children admitted to the Grove Hospital were assigned to receive routine therapy or routine therapy plus one ounce (28 g) of cod-liver oil, which is particularly rich in vitamins A and D. Under-five mortality was reduced from 8.3% to 3.7% by vitamin A supplementation and, as reported in later studies, the effect was even more pronounced in children younger than 2 years of age. Ellison attributed the effect of cod-liver oil to vitamin A and not to vitamin D because of the previously reported effects of vitamin A. The long-established practice of daily dosing of young children with cod-liver oil has disappeared from most households in Europe, although it still persists in Norway. In developing countries where vitamin A deficiency is widespread, WHO and UNICEF recommend that children with measles be given a massive dose of vitamin A.[17] Indeed, WHO has expanded the indications for use of vitamin A to "all cases of severe measles," not just to children from populations where vitamin A deficiency is known to exist or where measles case fatality rates exceed 1%.[18]

Table 4. Reduction in Morbidity and Mortality from Measles Infection in Children in Cape Town, South Africa, Supplemented with Vitamin A

	Vitamin A (*n*=92)	Placebo (*n*=97)
Deaths, *n*	2	10
Pneumonia		
Duration of illness, days	5(3–8.5)[a]	8(5–17)
Children with symptoms >10 days, *n*	12	29
Diarrhea		
Duration of illness, days	5(3–7)	7(5–10)
Children with symptoms >10 days, *n*	8	21
Post-measles croup, *n*	13	27
Herpes stomatitis, *n*	2	9

Note: Children received either a massive dose of vitamin A (200,000 IU) or placebo upon admission and the following day.
[a]Median (25th and 75th percentiles).
Source: Data from Hussey and Klein.[11]

Vitamin A Supplementation and Measles Immunization

There is much interest in combining immunization, such as that against measles, with massive doses of vitamin A. This is for two reasons. First, immunization provides a contact opportunity with the child, which, for measles immunization, comes at a time when the child's vitamin A reserves are declining in the second 6 months of life. Second, measles immunization and vitamin A supplementation can be regarded as complimentary because of the interrelationship between the severity of measles disease and vitamin A deficiency. It is important to know, however, whether giving a massive dose of vitamin A (100,000 IU for children ages 6–12 months) will reduce seroconversion of the vaccine, the most common of which is the Schwarz vaccine. Thus the 1995 report by Semba et al.[19] that vitamin A supplementation significantly reduced seroconversion from 79% to 66% in 6-month-old infants in Indonesia came as a disappointment (Table 5). Not only was seroconversion, defined as a fourfold increase in antibody titer, adversely affected but so was protection, defined as a titer of >120. In the placebo group, this protection was 77%, whereas, in the vitamin A–supplemented group, protection was 62%. However, because measles immunization is usually given at 9 months, it was necessary to repeat the study with older infants. When this was

Table 5. Effect of Vitamin A Supplementation on Seroconversion Following Measles Vaccination

	Seroconversion, %	
	Vitamin A	Placebo
Vaccination at 6 months		
Semba et al.[19] (Indonesia)	66	79
Vaccination at 9 months		
Semba et al.[20] (Indonesia)	100	100
Benn et al.[21] (Guinea-Bissau)	97	93

done, Semba and colleagues[20] found that 100% of children seroconverted. (Table 5)

Similar results were reported by Benn and colleagues[21] from Guinea-Bissau. (Table 5)They also showed that there was no difference in seroconversion and protection at 18 months when children were given the measles vaccine at 6 and 9 months. The results obtained by Semba and colleagues,[19] when immunization was combined with vitamin A supplementation at 6 months, could be explained by the fact that at this age, maternal antibodies against measles are still present in some children (in their study, the proportion of such children was 67%). Thus, they observed no difference in seroconversion in those with baseline titers of >8. Studies were also carried out to see whether massive dosing with vitamin A would reduce seroconversion of other vaccines. No effect was demonstrated with vaccines against petussis, tetanus, and polio, whereas seroconversion was enhanced with the diphtheria vaccine. Combining massive vitamin A dosing with vaccination should therefore be actively encouraged: For measles, this generally means when vaccination takes place at 9 months. For high-risk groups, however, such as refugee populations and HIV-infected infants, the 'early' two-dose measles vaccination regimen (6 and 9 months) should be adopted.[22]

Mechanism of Interaction Between Measles Infection and Vitamin A Deficiency

In many human and animal studies of the relationship between infection and nutrition, it was difficult to separate the specific effects of the nutrient in question from a generalized deficiency of a variety of nutrients and energy. With respect to vitamin A deficiency, many of the experimental models also used animals that were so severely vitamin A–deficient it was not possible to extrapolate the results to humans.

We therefore applied an animal model, first introduced by the Bangs,[23] based on chickens with marginal vitamin A deficiency and infection with Newcastle disease virus[24,25] because this virus, and the disease process associated with it, has many characteristics in common with measles. Although measles virus does infect rabbits, the disease is milder in that species than in man. Measles virus is in the genus *Morbillivirus,* along with distemper that infects dogs and ferrets, rinderpest that infects cattle, and 'peste des petits ruminants' that infects sheep and goats. None of these virus-animal dyads can be used realistically as models. In other genera of the family Paramyxoviridae, *Paramyxovirus* and *Pneumovirus*, most of the viral species infect primates. One exception is Newcastle disease virus, a *Paramyxovirus* that infects poultry, including chickens. In the Netherlands, all chickens are immunized successively with two viruses: first a lentogenic (almost avirulent) strain, La Sota, and subsequently a mesogenic (mildly virulent) strain. Thus the chicken infected with the La Sota strain of Newcastle disease virus provides an excellent and safe model.

Day-old chickens with limited vitamin A reserves, the progeny of marginally vitamin A–deficient hens, were fed purified diets containing either marginal (120 retinol equivalents/kg feed, ad libitum) or adequate (1200 retinol equivalents/kg feed, ad libitum or pair-fed) levels of vitamin A for a period of 10 weeks. At 4 weeks of age, half of the chickens in each group were infected intraocularly with virus. Infection resulted in increased rates of morbidity in the marginally vitamin A–deficient chickens compared with their nondeficient infected counterparts (Figure 1). Moreover, within 1 week of infection, plasma retinol levels in the infected marginally vitamin A–deficient chickens showed a significant and persistent decline compared with their noninfected counterparts fed the same diet (Figure 2). Thus infection with Newcastle disease virus exacerbates existing marginal vitamin A status and any effect of

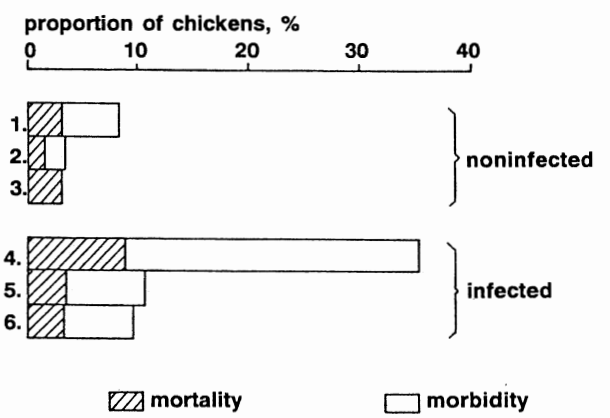

Figure 1. Effect of Newcastle disease virus infection on morbidity and mortality within 14 days of infection in chickens fed diets from the time of hatching containing marginal or adequate amounts of vitamin A as described in Figure 1. Groups 1–3, noninfected; groups 4–6, infected; groups 1 and 4, vitamin A–deficient; groups 2 and 5, vitamin A–adequate, pair-fed; groups 3 and 6, vitamin A–adequate, ad libitum–fed. Morbidity is defined as the appearance of clinical signs of disease including respiratory problems, diarrhea, or general weakness. Chi-square analysis in a log-linear model combining groups fed adequate amounts of vitamin A, which did not differ: Morbidity: vitamin A nutriture, P=0.001; infection, P=0.001. Mortality: vitamin A nutriture, P=0.188; infection, P=0.319. Source: reference 24. Reprinted with permission of the American Society for Nutritional Sciences.

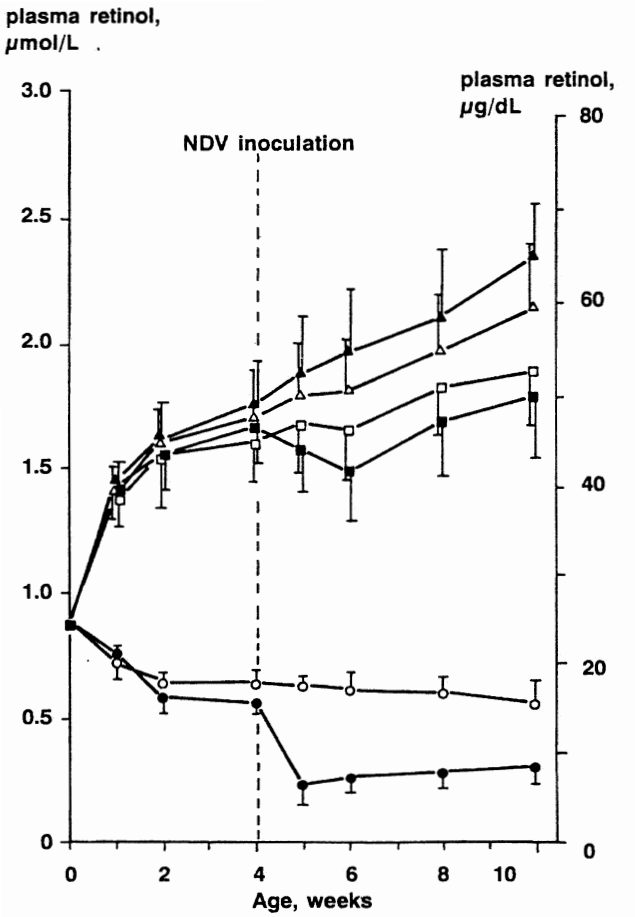

Figure 2. Effect of Newcastle disease virus infection on plasma retinol concentrations in chickens fed diets from the time of hatching containing marginal (120 RE/kg feed; circles) or adequate amounts of vitamin A (1200 RE/kg feed) either ad libitum (triangles) or pair-fed (squares) to those fed diets containing marginal amounts of vitamin A (squares). Open symbols represent noninfected chickens whereas closed symbols represent chickens inoculated intraocularly with La Sota strain of Newcastle disease virus. Values are means with the vertical bars representing one SD (n=11–15). Source: reference 24. Reprinted with permission of the American Society for Nutritional Sciences.

vitamin A deficiency.

What is the mechanism of the reduction in plasma retinol concentrations with measles infection? Retinol is carried in plasma as a complex with retinol-binding protein and transthyretin. We demonstrated that the concentration of retinol-binding protein in vitamin A–deficient chickens infected with Newcastle disease virus is lower than in noninfected counterparts, which indicates that infection reduces synthesis of the protein.[25] This correlates with the acute phase character of retinol-binding protein. A second reason for lowered plasma retinol concentrations following measles infection is the loss of vitamin A in urine associated with fever as was first reported by Moore and colleagues[26] in 1941. Stephensen and colleagues[27] showed that in children with fever in Bangladesh, daily urinary excretion of vitamin A can exceed 50% of the dietary reference intake. In a later study,[28] the same group showed that in children with Shigella dysentery tubular reabsorption of low-molecular-weight proteins including retinol-binding protein were impaired. Third, vitamin A can be lost in stools during measles diarrhea, not only because of rapid transit time but also because of leakage through the epithelial layer of the intestine. Fourth, damage to epithelial tissues as a result of vitamin A deficiency increases the demand for vitamin A for repair.

Host Defense: Nonspecific Mechanisms

Host defense begins at the epithelial surface. Thus, the integrity of the mucosal cell layer and its protective mucous covering could play an important role in defense. In 1925, Wolbach and Howe[29] showed that in vitamin A deficiency, mucous-producing epithelial cells were replaced by keratin-producing cells. These changes are seen particularly in the mucosa of the respiratory, gastrointestinal, and genitourinary tracts, in the conjunctival-corneal epithelium, and the lining of the taste buds. In cellular differentiation, suppression of the genome occurs as characterized by the disappearance of proteins specific to the undifferentiated state and appearance of other specific proteins. The role of vitamin A and its retinoid derivatives in cellular differentiation are now becoming understood in terms of their effects on gene expression.[30] The synthesis of glycoproteins, components of epithelial mucin and the surface of epithelial cells, also requires vitamin A. In vitamin A deficiency, the number of goblet cells responsible for mucin production are reduced in epithelial tissues in the conjunctiva[31] and the duodenum.[32] Therefore, it is somewhat surprising that vitamin A supplementation of children with vitamin A deficiency does not reduce the incidence but does reduce the severity of disease.[33]

After entering the body via epithelial surfaces, microbes spread to other tissues. This is facilitated by the circulatory system and hindered by macrophages and monocytes.[34] The increased microbiocidal capacity of macrophages in response to infection is largely nonspecific because activated macrophages that arise in response to infection with one microorganism tend to show increased activity against other microorganisms.[35] We examined the effect of vitamin A deficiency on the activity of peritoneal macrophages in our chicken–Newcastle disease virus model.[36] The activity of macrophages against microorganisms can be divided into an attachment and ingestion phase (phagocytosis), which was assessed by counting the uptake of fluorescein isothiocyanate–labeled yeast cells, and an oxygen-dependent killing phase, which was assessed by measuring the reduction of nitroblue tetrazolium (NBT), both with and without additional stimulation in vitro with zymosan A particles isolated from yeast. Vitamin A deficiency had no significant effect on phagocytosis in peritoneal macrophages isolated from both infected

Table 6. Effect of Vitamin A Intake and Newcastle Disease Virus Infection in Chickens on Phagocytosis and Nitroblue Tetrazolium (NBT) Reduction by Chicken Peritoneal Macrophages

	Phagocytosis (cells ingested/PM)	NBT Reduction (absorbance/mg protein)	
		Without zymosan A	With zymosan A
Noninfected			
Adequate	3.7±0.3[a]	11.6±1.1[a]	32.9±1.6[a]
Deficient	3.4±0.3[a]	7.0±1.2[b]	20.9±1.8[b]
Infected			
Adequate	4.6±0.2[b]	14.9±2.1[a]	41.8±3.6[c]
Deficient	3.8±0.3[ab]	13.1±1.6[a]	26.2±2.2[ab]

Note: Mean±SE (n=6–8 per group). Values in the same column not sharing the same superscript are significantly different from one another (Tukey): $P<0.05$.
Source: From Sijtsma et al.[36]

Table 7. Effect of Vitamin A Intake and Newcastle Disease Virus Infection in Chickens on Lymploid Organ Weights 9 Days After Virus Inoculation at the Age of 37 Days

	Organ Weight, g		
	Bursa	Thymus	Spleen
Noninfected			
Adequate	1.84±0.23[a]	1.75±0.47[a]	0.90±0.23[a]
Deficient	1.46±0.22[bc]	1.55±0.21[ab]	0.81±0.21[a]
Infected			
Adequate	1.67±0.19[c]	1.85±0.27[a]	1.06±0.22[a]
Deficient	1.24±0.16[b]	1.31±0.26[b]	0.90±0.22[a]

Note: Mean±SE (n=10 per group). Values in the same column not sharing the same superscript are significantly different from one another (Tukey): $P<0.05$.
Source: Sijtsma et al.[39]

and noninfected chickens (Table 6). However, deficiency did significantly depress NBT reduction in peritoneal macrophages isolated from both noninfected and infected chickens. The effect in infected chickens was only seen after additional stimulation in vitro with zymosan A particles isolated from yeast, which suggests that differences in NBT reduction could be attributed partially to the decrease in uptake. This difference was not observed in a study in Indian children with serum retinol levels greater than or less than 0.7 µmol/L in either phagocytic activity or oxygen-dependent killing.[37] It is difficult, however, to assess vitamin A status in infected children based only on serum retinol levels.

In addition to phagocytic cells, antiviral substances such as the enzyme lysozyme, transferrins, precipitins, natural cytotoxins, and complement play a crucial role in preventing the spread of infections. Lysozyme is a glycoprotein dependent on vitamin A for its synthesis. This dependence on vitamin A was demonstrated in a further study in children in India[38] in which the activity of lysozyme in leucocytes was significantly lower in children with ocular signs of vitamin A deficiency compared with control children. Following vitamin A therapy, the levels of enzyme in the deficient children returned to normal.

Host Defense: Specific Mechanisms

In addition to the nonspecific host defense system, the immune system is comprised of a number of organs and cell types that have evolved to accurately and specifically recognize nonself antigens such as those present on microbes and microbe-infected cells. These ultimately aid in the elimination of such microbes and cells. In the early work of the Bangs[23] using the chicken–Newcastle disease virus model, vitamin A deficiency or Newcastle disease virus infection alone was found to have only moderate effects on lymphoid systems. However, together they caused substantial or even total loss of lymphocytes from primary lymphoid organs, as well as rapid weight loss. Using our model, in which there was no loss of body weight,[39] we demonstrated that vitamin A deficiency resulted in lower weights of the bursa of Fabricius in both noninfected and infected chickens and of thymus in infected chickens, indicating changes in production and/or recruitment of B and T cells, respectively (Table 7). However, no effect of vitamin A deficiency on spleen weight was observed. These changes were accompanied by changes in the number of circulating lymphocytes.

Vitamin A deficiency resulted in a significant lymphopenia that was even more pronounced in Newcastle disease virus–infected chickens during the acute phase of infection (Table 8). Using flow cytometry with a monoclonal antibody specific for chicken immunoglobulin (Ig) light chain, we were able to characterize the circulating lymphocyte population into surface Ig–positive cells (B cells) and surface Ig–negative cells (mainly T cells). We also showed that vitamin A deficiency significantly low-

Table 8. Effect of Vitamin A Intake and Newcastle Disease Virus Infection in Chickens on Peripheral Blood Lymphocytes Numbers 5 and 11 Days After Infection.

	Lymphocytes ($\times 10^7$/nL)	
	4 days	11 days
Noninfected		
Adequate	1.7±0.1[a]	1.8±0.2[a]
Deficient	1.3±0.3[b]	1.2±0.1[b]
Infected		
Adequate	1.3±0.1[b]	2.1±0.1[c]
Deficient	1.0±0.1[c]	1.5±0.1[d]

Note: Mean±SE (10–12 per group). Values in the same column not sharing the same superscript are significantly different from one another (Tukey): $P<0.05$.
Source: Sijtsma et al.[39]

ered the number of circulating B cells and, in Newcastle disease virus–infected chickens, the number of T cells.[39] Vitamin A–induced lymphopenia is probably a result not only of impaired development of primary lymphoid organs, as indicated by the reduced weight of the bursa of Fabricius with consequent impaired proliferation and differentiation of lymphoid cells, but also of changes in homing patterns.[40]

The increase in the number of circulating peripheral blood lymphocytes following the period of Newcastle disease virus–induced lymphopenia might be attributed to a change in migration pattern rather than increased production of new lymphocytes.[41] Both the lymphopenic effect and the subsequent increase in the number of circulating lymphocytes owing to Newcastle disease virus infection were less pronounced in vitamin A–deficient chickens than in their counterparts pair-fed the control diet. This potentially suggests that the recruitment of cells has been impaired. The results from the present study indicate that vitamin A deficiency affects lymphoid cell systems and that this is aggravated by concomitant Newcastle disease virus infection.

Natural measles infection causes prolonged depression of cell-mediated immunity. This was shown to be associated not only with depletion of T cells but also with the activity of an inhibitor of lymphocyte proliferation in serum, and possibly with defective antigen processing.[42]

Depressed cellular response to mitogens is characteristic of vitamin A deficiency.[43] We showed that vitamin A deficiency reduces cytotoxic T lymphocyte activity in Newcastle disease virus infection.[44] This impairment is an important part of the cell-mediated defense system and could have important implications for recovery from viral infection. In studies in children with marginal vitamin A deficiency, no difference in in vitro mitogenic stimulation of T cells has been found,[37] but as mentioned above, it is difficult to determine vitamin A status during infection. Various other effects of vitamin A on cellular immune response were discussed by Hanson.[45]

With respect to humoral immunity, vitamin A deficiency was shown to be accompanied by low levels of immunoglobulins and impaired response to antigens.[43,45] We studied primary and secondary immunization with sheep red blood cells and bovine serum albumin as thymus-dependent antigens and *Brucella abortus* as a more or less thymus-independent antigen during and after the acute phase of disease produced by Newcastle disease virus infection.[46] Vitamin A deficiency did not affect the hemagglutination-inhibition antibody response to Newcastle disease virus. The level of the primary and secondary IgG response to bovine serum albumin and of the primary IgG response to sheep red blood cells and bovine serum albumin was reduced, whereas the secondary IgM response to sheep red blood cells and *Brucella abortus* was slightly elevated by vitamin A deficiency. Newcastle disease virus infection reduced primary IgM and IgG responses to sheep red blood cells and bovine serum albumin but not to *Brucella abortus*, indicating a defect in T–helper cell function, as was suggested earlier.[47] The combination of vitamin A deficiency and Newcastle disease virus infection resulted in the lowest IgG titers to thymus-dependent antigens. Because most pathogens are of the latter type, the risk for secondary infection appears to be increased in vitamin A deficiency and Newcastle disease virus infection.

With respect to immunologic protection on mucosal surfaces, as opposed to the nonspecific protection referred to above, the basic component is secretory immunoglobulin A (sIgA). In our model, vitamin A deficiency lowered the concentration of sIgA in bile and this effect was even more pronounced in chickens infected with Newcastle disease virus.[48] There was, however, no effect on the IgM concentration in bile or on the number of IgA- or IgM-containing plasma cells in mucosal cells, indicating that hepatobiliary transport of sIgA is impaired in vitamin A deficiency and that this effect is exacerbated by Newcastle disease virus infection. Similar results were found in noninfected vitamin A–deficient rats.[49] Thus the host's ability to defend itself at the mucosal level against both primary and secondary infections would be impaired.

Concluding Remarks

Measles infection was shown to compromise nutrition status and vice versa. The nutrient that appears to play the most important role in this respect is vitamin A. Children in developing countries are at particular risk because they are exposed to low rates of measles immunization and to vitamin A deficiency, both primary[50] and secondary. Therefore, measles infection and vitamin A deficiency need to be considered parallel problems. Current advice from WHO and UNICEF is to provide massive doses of vitamin A to children with measles and at the time of measles vaccination; both procedures are considered safe and effective. We are beginning to understand some of the mechanisms involved in the interaction between measles infection and vitamin A deficiency but there is need for more fundamental work in this area.

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