EFFECT OF VITAMIN A SUPPLEMENTATION ON MORBIDITY OF LOW-BIRTH-WEIGHT NEONATES

Anna Coutsoudis, Miriam Adhikari, Kibendran Pillay, Louise Kuhn, Hoosen M Coovadia

Background. Low-birth-weight (LBW) infants (< 2 500 g) are at increased risk of respiratory infection in the first few months of life and have low liver stores of vitamin A. As retinol is essential for respiratory epithelial cell differentiation, deficiency could result in pathological changes in the respiratory epithelium, with respiratory problems.

Objective. A randomised, double-blind, placebo-controlled trial to investigate the effect of vitamin A supplementation on the incidence and severity of respiratory infections in LBW infants during their first year of life.

Method. One hundred and thirty LBW infants (gestational age < 36 weeks and birth weight 950 - 1 700 g) were enrolled in the study. The infants were randomly allocated to a vitamin A or placebo group. Infants in the vitamin A group received 25 000 IU of vitamin A (retinyl palmitate, Arovit drops, Roche, Basle, Switzerland) on study days 1, 4 and 8. Study day 1 was between 36 and 60 hours after delivery. Infants in the placebo group received a placebo (formulated by Roche) with a similar appearance and packed in the same dropper bottles as the vitamin A drops.

Results. Vitamin A supplementation markedly improved serum retinol levels. After the last vitamin A dose, the vitamin A group had higher mean serum retinol concentrations than the placebo group (45.77 ± 17.07 μg/dl v. 12.88 ± 6.48 μg/dl, $P = 0.0001$). There was no evidence of improvement in neonatal or post-neonatal respiratory problems associated with vitamin A supplementation. Vitamin A and placebo groups did not differ in the occurrence or duration of respiratory distress or the need for head-box oxygen. There were also no significant differences in the cumulative probability of developing lower or upper respiratory tract infection through the first year of life. There was a slight suggestion of an increase in the risk of

Department of Paediatrics and Child Health, University of Natal, Durban
Anna Coutsoudis, PhD
Miriam Adhikari, MD
Kubendran Pillay, FCP (Paed)
Hoosen M Coovadia, MD

Gertrude H Sergievsky Centre, University of Columbia, New York
Louise Kuhn, PhD

hospitalisation with pneumonia associated with vitamin A supplementation. The cumulative probability of being hospitalised with pneumonia by 6 months of age was 24.6% (7 hospitalisations) in the vitamin A group compared with 7.4% (2 hospitalisations) in the placebo group (log rank test $P = 0.04$). After adjusting for risk factors this difference was no longer significant.

Conclusion. Vitamin A supplementation in LBW neonates may not reduce incidence or severity of respiratory infections. These results do not negate the importance of improving vitamin A status in children as an important public health measure to reduce morbidity and mortality from other childhood infections.

S Afr Med J 2000; **90:** 730-736.

Developing countries have a high incidence of low-birth-weight (LBW) deliveries (< 2 500 g), which make a major contribution to perinatal mortality.[1] These infants are at increased risk of respiratory infections in the first few months of life.[2] It has been well established that infants born prematurely have low liver stores of vitamin A[3,4] as well as low serum retinol concentrations.[5-8] As retinol is essential for epithelial cell differentiation and may play a role in the integrity of the epithelial lining of alveoli and airways,[5] it is hypothesised that vitamin A deficiency in these infants could result in pathological changes in the respiratory epithelium, with resultant respiratory problems.[9,10] Three studies have investigated the role of vitamin A therapy in bronchopulmonary dysplasia (BPD);[11-13] however, in developed[14] and developing countries, the major problem for LBW infants is not BPD but rather pneumonia and other respiratory infections, both in the immediate neonatal period and during the first year of life (MA — personal communication). We therefore undertook a randomised, double-blind, placebo-controlled trial to investigate the effect of vitamin A supplementation on the incidence and severity of respiratory infections in LBW infants during their first year of life. This study was undertaken at King Edward VIII Hospital (KEH), Durban, which mainly serves a black, disadvantaged community where childhood infections[15] and pregnancy complications are common.[16,17]

SUBJECTS AND METHODS

Subjects and treatment

One hundred and thirty consecutive LBW infants were enrolled in the intervention trial. The infants delivered at KEH between June and November 1993 were enrolled in the study. The infants (of gestational age < 36 weeks and birth weight 950 - 1 700 g) were enrolled with parental consent into this double-blind study. Relevant details about each baby, including sex, birth weight, gestational age and clinical problems, were noted. Clinical problems were those common conditions that may occur at the time of birth or in the first few days of life and included asphyxia, intraventricular haemorrhage (IVH), anaemia, septicaemia, jaundice, hypothermia, hyaline membrane disease (HMD) and congenital syphilis. Obstetric problems included pregnancy-induced hypertension and abruptio placentae. Cranial ultrasound was performed as soon as possible after birth and daily thereafter for 3 days, and weekly if an IVH was diagnosed.

The infants were randomly allocated to a vitamin A or placebo group. Infants in the vitamin A group received 25 000 IU of vitamin A (retinyl palmitate, Arovit drops, Roche, Basle, Switzerland) on study days 1, 4 and 8. Study day 1 was between 36 and 60 hours after delivery. This period of time was allowed for the baby to adapt to extra-uterine life and to be able to tolerate oral feeds. Infants in the placebo group received a placebo (formulated by Roche) with a similar appearance and packed in the same dropper bottles as the vitamin A drops. The dropper bottles were number coded and vitamin A/placebo was administered by one research assistant directly into the nasogastric tube; care was taken to ensure that the oral preparation was flushed down the tube with mother's breast-milk immediately after administration.

Infants born by normal vaginal delivery were all fed expressed breast-milk. Infants born by caesarean section whose mothers were in high care for the first few days were fed with formula feed (S26 Preemie). All the infants in the study were fed by nasogastric tube for the first week of life, after which some continued to be fed nasogastrically while others were breast-fed.

Outcome variables

The outcome variables included the development of upper respiratory tract infections (URTIs) defined as rhinitis, ear and throat infections and/or cough with fever; lower respiratory tract infections (LRTIs) defined as cough, fever, stridor, wheeze, tachypnoea, chest wall retractions and/or crackles; diarrhoeal disease and thrush.

This information was recorded weekly while the infant was in hospital and at 1, 3, 6, 9 and 12 months (chronological age) thereafter. At each visit any infection in the interim period following the previous visit was recorded.

Exclusion criteria

Any child who developed severe asphyxia (grade III - IV IVH), congenital abnormalities, meningitis or septicaemia (with haemodynamic instability) or who was placed on a ventilator within the first 60 hours of life was not eligible for entry into the trial. In addition, any child not established on nasogastric feeding within the first 60 hours of life was considered ineligible.

Ethical considerations

Written informed consent was obtained from the mothers of the infants involved in the study. The study was approved by the Ethics Committee of the University of Natal, Faculty of Medicine.

Blood sampling

Baseline blood samples were obtained from a sub-sample of 35 of the infants within 48 hours of delivery and before any vitamin A had been administered. A repeat blood sample (to determine whether the oral vitamin A had been absorbed) was taken at approximately 5 hours after vitamin A/placebo had been administered.

Vitamin A concentrations

One millilitre of venous blood was obtained and centrifuged within 5 hours. The serum was separated and stored at –20°C until analysis. Precautions were taken to protect the serum from light during separation, storage and analysis. Vitamin A (serum retinol) was measured by normal phase high-pressure liquid chromatography using fluorescence detection. The method used was a modification of a previously reported method.[18] The instrument used was a Hewlett-Packard HP 1090, which was attached to a programmable fluorescence detector (HP 1046). The column was a normal phase microbore column (Spherisorb S3W, Phase Sep, UK). The method was validated by using standard reference material for retinol (SRM 968a) from the National Institute for Standards and Technology (Gaithersburg, Maryland, USA). All samples were analysed in duplicate within 3 months of collection. Approximately equal numbers of controls and patients were analysed in each batch and the technician was blinded to their treatment regimen.

Monitoring for side-effects of vitamin A

Each infant was assessed before administration of each vitamin/placebo dose; after each dose infants were monitored at 4-hourly intervals for 48 hours for vomiting, bulging fontanelle, drowsiness and irritability. A bulging fontanelle was defined as a fontanelle that was tense to the touch and protruded from the skull; diagnosis was made only when the child was not crying. All nursing staff and research personnel were blinded to the treatment group of the infants. A proviso had been set that if more than 5 children exhibited vomiting or bulging fontanelle a blinded interim analysis would be conducted by a team consisting of a statistician and an epidemiologist to determine if the side-effects were vitamin A-related and who would then, if necessary, advise on discontinuing the trial.

Statistical methods

The vitamin A and placebo groups were compared on baseline characteristics — differences between continuous variables (e.g. age) were tested using *t*-tests and categorical variables (e.g. sex) were tested using chi-squared tests. Multivariate analysis of possible differences between vitamin A and placebo groups in terms of respiratory distress or need for oxygen, taking into account other neonatal characteristics, was done using multivariate logistical regression models. Morbidity and mortality over the first year of life were analysed using Kaplan-Meier lifetable methods and tested using log rank tests. In addition to survival, time taken to develop each illness of interest (LRTI, URTI, diarrhoea and thrush) was calculated. Children who were lost to follow-up were censored at the age when they were last seen. This method provides an estimate of the proportion of children developing the various illnesses of interest by specific ages. It does not, however, take into account multiple episodes of the condition, nor does it describe the duration of these episodes. Therefore these parameters were also described. Taking into account baseline neonatal characteristics, Cox Proportional Hazards models were used to investigate whether or not there were differences between the groups in terms of developing these illnesses.

RESULTS

Study population

Of the 130 infants, 116 were randomly assigned. Of the 14 who were excluded, 3 died before receiving their assigned treatment, and 11 were placed on a ventilator and/or nasogastric feeding was not established. All infants were breast-fed for the first 3 months. No infant was exclusively breast-fed for the full 3 months.

Comparability of groups at baseline

The characteristics of the infants at baseline are shown in Table I and both groups had similar baseline characteristics. Mothers' characteristics (shown in Table I) were similar. A notable exception, however, was the significantly higher number of mothers in the vitamin A-treated group with positive syphilis serology.

Serum retinol levels

The mean serum retinol concentration before administration of the supplement was similar in the two subgroups (9.83 ± 5.45 μg/dl in the placebo group v. 10.36 ± 6.50 μg/dl in the vitamin A group). However, 5 hours after the last vitamin A dose the vitamin A supplemented group had higher mean serum retinol concentrations than the placebo group (45.77 ± 17.07 μg/dl v. 12.88 ± 6.48 μg/dl, $P = 0.0001$). This difference between the groups was also present when the mean increase in serum retinol for each individual was determined (35.41 ± 15.72 μg/dl v. 3.04 ± 4.89 μg/dl, $P = 0.0001$).

Table I. Baseline characteristics of 116 LBW infants randomised to vitamin A or placebo groups

	Placebo ($N = 60$)	Vitamin A ($N = 56$)
Mother's age (yrs) (mean (SD))	24.7 (6.59)	26.0 (6.17)
Gestational age (wks) (mean (SD))	31.8 (2.21)	32.3 (2.54)
Birth weight (g) (mean (SD))	1 351 (188)	1 330 (217)
Apgar 1-minute score (mean (SD))	7.1 (1.42)	7.2 (1.60)
Apgar 2-minute score (mean (SD))	9.3 (0.99)	9.2 (0.86)
Male (%)	46.7	42.9
Asphyxia (%)	3.3	5.4
Intraventricular haemorrhage (%)	23.2	17.9
Anaemia (%)	6.7	7.1
Septicaemia (%)	6.8	5.5
Jaundice (%)	58.3	60.7
Fever (%)	8.3	9.1
Hypothermia (%)	26.7	30.4
Hyaline membrane disease (%)	13.3	10.7
Vaginal delivery (%)	84.7	82.1
Born before arrival (%)	20.0	16.1
Pregnancy-induced hypertension (%)	5.0	8.9
Congenital syphilis (%)	3.3	3.6
Positive maternal screening test for syphilis (WR) (%)	5.0	16.1*

* $P = 0.05$. For all other comparisons $P > 0.05$.

Side-effects

No infant in either the placebo or vitamin A group was found to have feeding difficulties (failure to feed or vomiting), a bulging fontanelle, or neurological signs either before or after administration of vitamin A.

Neonatal respiratory problems

There were no significant differences between the vitamin A and placebo groups in terms of the occurrence or duration of respiratory distress or head box oxygen (Table II). No infants in the vitamin A group were on mechanical ventilation, compared with 4 in the placebo group, a difference that was statistically significant but based on very small numbers. Among those with respiratory distress, the mean duration was 2.4 days in the vitamin A group, and 3.3 days in the placebo group. Among those requiring oxygen, the mean duration was 2.6 and 2.5 days among the vitamin A and placebo groups respectively.

The occurrence of respiratory distress was significantly associated with birth weight, gestational age, low Apgar scores, asphyxia, IVH, hypothermia, mechanical ventilation, and HMD in univariate analysis. In multivariate analysis, none of the co-morbid conditions remained significantly associated with respiratory distress after adjusting for either birth weight or gestational age. Vitamin A supplementation was not significantly associated with respiratory distress (odds ratio (OR) 0.82, 95% confidence interval (CI) 0.38 - 1.78) after adjusting for birth weight. Similarly, the need for head box oxygen was associated with birth weight, gestational age, anaemia, jaundice, IVH, and HMD in univariate analysis. In multivariate analysis, vitamin A supplementation was not significantly associated with the need for head box oxygen (OR 0.84, 95% CI 0.38 - 1.86) after adjusting for birth weight.

Table II. Neonatal respiratory problems (occurrence and duration of respiratory distress or head box oxygen and mechanical ventilation) in vitamin A and placebo groups

	Placebo ($N = 60$)	Vitamin A ($N = 56$)	P-value
Respiratory distress (%)			
None	41.7	44.6	
One or 2 days	31.7	35.7	
Three or more days	26.7	19.6	0.67
Head box oxygen (%)			
None	36.7	35.7	
One or 2 days	40.0	37.5	
Three or more days	23.3	26.8	0.91
Mechanical ventilation (%)			
Yes	6.7	0.0	0.05

Follow-up rates

Of the 116 children in the study, 89 (77%) had at least one follow-up visit during the first year of life: 50% were followed to at least 3 months of age, 39% to at least 6 months, and 21% to 12 months. There were no differences in follow-up rates between vitamin A and placebo groups. Cumulatively, the 56 children in the vitamin A group contributed 148 follow-up visits (an average of 3.4 visits per child in the 43 followed up) and the 60 children in the placebo group contributed 173 follow-up visits (an average of 3.8 visits per child in the 46 followed up).

Morbidity and mortality in the first year of life

There were 2 deaths in the vitamin A group and 2 deaths in the placebo group during the follow-up period. Cumulatively, by 6 months of age 6.5% of the infants in the vitamin A group had died compared with 5.7% in the placebo group (Table III).

There were no apparent differences between the groups in terms of the cumulative probability of developing LRTI, URTI, diarrhoea, or thrush through the first year of life (Table III).

There was a suggestion that LRTI observed in the vitamin A group may have been more severe than in the placebo group. Of the 10 children who developed LRTI in the vitamin A group, 7 were hospitalised, compared with 2 of 10 in the placebo group. In the vitamin A group there were 14 reported episodes of LRTI (a rate of 9.5 episodes per 100 visits (2 children reported more than 1 episode each, 1 child 2 episodes, 1 child 4 episodes)) with a mean duration of 9.6 days (duration of 50% of episodes was 7 or more days). In the placebo group there were 16 reported episodes of LRTI (a rate of 9.2 episodes per 100 visits (4 children reported more than 1 episode each, 2 had 2 episodes, 1 had 3 and 1 had 4 episodes)), with a mean duration of 7.1 days (duration of 31% of episodes was 7 or more days).

The cumulative probability of being hospitalised with pneumonia by 6 months of age was 24.6% (7 hospitalisations) in the vitamin A group compared with 7.4% (2 hospitalisations) in the placebo group (log rank test $P = 0.04$). After adjusting for birth weight and other neonatal characteristics associated with pneumonia hospitalisation in univariate analysis (hypothermia, low Apgar scores, maternal Wassermann reaction result), children in the vitamin A group retained about a threefold increased risk of hospitalisation with pneumonia, but the increase was no longer significant ($P = 0.19$ from proportional hazards model).

Severity of the other conditions was similar between the two groups. The 32 reported episodes of URTI (incidence rate of 21.6 episodes per 100 visits) in the vitamin A group had a mean duration of 6.8 days, while the 42 episodes (incidence rate 24.3 per 100) in the placebo group had a mean duration of 6.9 days. The 6 episodes of diarrhoea (incidence rate 4.1 per 100 visits) in the vitamin A group had a mean duration of 3.8 days, while the 5 episodes (incidence rate 2.9 per 100) in the placebo group had a mean duration of 3 days. One child in each group had a repeat episode of thrush.

Anthropometry

There were no differences in any of the growth parameters between the two groups, and height and head circumference for age were similar in the two groups.

Table III. Morbidity (URTI, LRTI, diarrhoea, and thrush) and mortality through the first year of life in vitamin A and placebo groups

	Number of children	Number with condition	Cumulative % of children developing the condition by each age*			
			3 months	6 months	12 months	*P*-value
All-cause mortality						
Vitamin A	43	2	0.0	6.5	6.5	
Placebo	46	2	2.2	5.7	5.7	0.89
LRTI						
Vitamin A	43	10	29.8	35.2	35.1	
Placebo	46	10	13.4	31.0	31.0	0.52
URTI						
Vitamin A	43	21	30.2	55.1	70.1	
Placebo	46	18	41.6	53.5	73.8	0.69
Diarrhoea						
Vitamin A	43	4	0.0	0.0	25.6	
Placebo	46	5	0.0	0.0	33.7	0.47
Thrush						
Vitamin A	43	7	15.0	15.0	25.6	
Placebo	46	6	19.8	19.8	19.8	0.93

* Cumulative percents calculated from Kaplan-Meier lifetable methods which adjust for the duration of follow-up.
URTI = upper respiratory tract infection; LRTI = lower respiratory tract infection.

DISCUSSION

This study attempted to define the role of vitamin A in the LBW infant who is susceptible to respiratory infections in the first year of life and who may require rehospitalisation.[14] We had a low follow-up rate; however, the pattern of follow-up rates is typical of a developing country in which the cost of travelling and access to health care facilities pose major problems for mothers. In our experience most mothers attend the neonatal follow-up 3 - 4 times a year.

Vitamin A supplementation has been shown to be of benefit in childhood — having a significant influence on the outcome of measles,[19] it has helped to reduce childhood mortality.[20,21] The success of vitamin A supplementation in the newborn is restricted to the prevention of BPD; one study showed the benefit of vitamin A,[9] while others[12,13] have shown less success.[9-11] Respiratory tract infections (RTIs) occur more commonly in the first few months of life of the LBW infant. At the weekly neonatal follow-up clinic at KEH RTIs comprised 66% of all infections experienced by the LBW infants.

Although there is consensus on the dose of vitamin A to be administered to children in both measles and community studies, there are no clear guidelines on what dose of vitamin A to use in LBW infants. We hypothesised that high doses of vitamin A should be used as soon as possible, preferably within the first week, the most vulnerable period for the LBW infant, during which time measures to promote the integrity of the respiratory epithelium and to strengthen the immune response are likely to produce maximum benefits. As 50 000 IU has been shown to be safe in full-term neonates,[12] we chose to use half this dose, viz. 25 000 IU, and to administer it on study days 1, 4 and 8.

A further consideration in vitamin A therapy for LBW infants is the route of administration. Generally, it has been assumed that the absorption and bio-availability of enteral vitamin A is reduced relative to that of intramuscular vitamin A, although no study has systematically studied the kinetics of vitamin A metabolism after absorption in LBW infants. The previous supplementation trials in LBW neonates used intramuscular vitamin A on alternate days[9,10] or 3 times a week[11] for 28 days. The use of repeated intramuscular injections in such tiny infants has many disadvantages, including the risk of poor absorption.[22] We decided to use oral vitamin A in this intervention study. We have already described a preliminary study in which 3 oral doses of 25 000 IU were well absorbed and had no toxic effects.[23] The mean serum retinol level in the treated group was adequate for the physiological effect of the drug.

The factors associated with respiratory distress and oxygen administration are common to the newborn and the analyses at univariate and multivariate levels are not unexpected.

The statistical analyses included the incidence of disease at follow-up and used three methods to measure these. One method included the incidence rates, i.e. giving the rate of the disease per 100 child visits. This takes into account the fact that a child can have more than one event, but does it not indicate the number of children who are disease-free. A second method was to calculate the proportion of children that develop the condition; this is less satisfactory since in this study there was a variable follow-up between the children. The third option was a lifetable or survival analysis, calculating time to the first development of the outcome of interest. Children who are lost to follow-up are censored when last seen. The advantage of this approach is that it allows an estimate of the proportion of children developing the condition of interest by certain age cut-offs, but it does not take into account multiple episodes of the condition.

One of the limitations of the study was that the follow-up rates were suboptimal. In order to calculate, *post hoc*, the power of the study to detect reductions in the incidence of LRTI by 6 months of age (our main outcome of interest), we used estimates of the probability of developing LRTI (and estimates of the standard error of this probability) from the lifetable estimates observed. Since at 6 months the placebo group had a probability of developing LRTI of 31% with a 95% CI 15 - 47%, a reduction in LRTI due to vitamin A supplementation stronger than about 0.5 (i.e. halving the incidence of LRTI or a reduction from 31% to 15%) can be ruled out. Weaker associations could not have been detected.

Not unexpectedly, RTIs were by far the commonest infections experienced in both the treated and placebo groups. There were no significant differences between the placebo and vitamin A-treated groups with regard to maternal data, weight, gestational ages and early neonatal problems.

The total number of hospitalisations was similar in the two groups; however, more babies in the vitamin A group were hospitalised for LRTI than those in the placebo group. Considering the overall results (for example, 4 children in the placebo-treated group needed mechanical ventilation and none in the vitamin A group), it is not clear that this necessarily implies that vitamin A may in fact be harmful, and it may have been fortuitous that the vitamin A-treated group were more ill, requiring hospitalisation.

URTIs occurred with equal frequency and duration in both groups. It could be anticipated that if vitamin A supplementation was a possible risk factor, then these infections would have been aggravated.

A vitamin A effect on mortality did not manifest in this study as deaths occurred equally in both groups. The study did not, however, set out to determine the effect on mortality — such a study would require a much larger sample size.

Other conditions that were monitored in this study, namely diarrhoeal disease and thrush, occurred in relatively few patients and about equally in both groups.

In conclusion, this study shows that vitamin A supplementation in LBW neonates does not reduce incidence or severity of respiratory infections, suggesting either that the other factors associated with LBW have a greater influence on respiratory outcome, or that vitamin A does not play a significant role in improving respiratory infections. Our study results are consistent with a meta-analysis of data from intervention trials that failed to indicate any consistent impact of vitamin A supplementation on the incidence of LRTIs in older infants and preschool children, a conclusion supported by a World Health Organisation *ad hoc* review panel.[24]

The results of this study obviously do not negate the importance of improving vitamin A status in children as an important public health measure to reduce morbidity and mortality from childhood infections.[25]

We thank Thembi Ngubane for her assistance in dispensing the medications and monitoring patients; the doctors and nursing staff of the prem nursery for their co-operation; Eleanor Gouws of the Medical Research Council Department of Biostatistics for assistance in data analysis; Inge Elson of the Analytical Unit, Department of Physiology, University of Natal for vitamin A analysis and Dr Mike Brown, Roche Products, South Africa for the generous donation of Arovit drops and placebo.

The study was supported by grants from Gerber-Purity, the Medical Research Council and the Faculty of Medicine, University of Natal.

References

1. Adhikari M, Moodley D, Desai PK, Moodley J. Trends in perinatal mortality and morbidity at a tertiary referral centre: 1980 - 1992. Implications for resource allocation. *International Child Health* 1995; **6:** 45-53.
2. McCracken GH. Bacterial and viral infections of the newborn. In: Avery GB, ed. *Neonatology, Pathophysiology and Management of the Newborn*. 2nd ed. Philadelphia: JB Lippincott, 1981: 723-747.
3. Olson JA, Gunning DB, Tilton RA. Liver concentrations of vitamin A and carotenoids, as a function of age and other parameters, of American children who died of various causes. *Am J Clin Nutr* 1984; **39:** 903-910.
4. Woodruff CW, Latham CB, Mactier H, Hewett JE. Vitamin A status of preterm infants: correlation between plasma retinol concentration and retinol dose response. *Am J Clin Nutr* 1987; **46:** 985-988.
5. Brandt RB, Schroeder JR, Guyer KE, Hutcher NE, Ehrlich FE. Serum vitamin A in premature and term neonates. *J Pediatr* 1978; **92:** 101-104.
6. Shenai JP, Chytil F, Jhaveri A, Stahlman MT. Plasma vitamin A and retinol binding protein in premature and term neonates. *J Pediatr* 1981; **99:** 302-305.
7. Hustead VA, Gutcher GR, Anderson SA, Zachman RD. Relationship of vitamin A (retinol) status to lung disease in the preterm infant. *J Pediatr* 1984; **105:** 610-615.
8. Coutsoudis A, Adhikari M, Coovadia HM. Serum vitamin A (retinol) concentrations and association with respiratory disease in premature infants. *J Trop Pediatr* 1995; **41:** 230-233.
9. Jetten AM, Smits H. Regulation of differentiation of tracheal epithelial cells by retinoids. In: *Retinoids, Differentiation and Disease* (Ciba Foundation Symposium 113). London: Pitman, 1985: 61-76.
10. Wong Y-C, Buck RS. An electron microscopic study of metaplasia of the rat tracheal epithelium in vitamin A deficiency. *Lab Invest* 1971; **24:** 55-56.
11. Shenai JP, Kennedy KA, Chytil F, Stahlman MT. Clinical trial of vitamin A supplementation in infants susceptible to bronchopulmonary dysplasia. *J Pediatr* 1987; **111:** 269-277.
12. Pearson E, Bose C, Snidow T, *et al.* Trial of vitamin A supplementation in very low birth weight infants at risk for bronchopulmonary dysplasia. *J Pediatr* 1992; **121:** 420-427.
13. Bental RY, Cooper PA, Cummins RR, Sandler DL, Wainer S, Rotschild A. Vitamin A therapy — Effects on the incidence of bronchopulmonary dysplasia. *South African Journal of Food Science and Nutrition* 1994; **6:** 141-145.
14. Van Zeben - van der Aa DM, Verloove-Vanhorick SP, Brand R, *et al.* The use of health services in the first 2 years of life in a nationwide cohort of very preterm and / or very low birth weight infants in the Netherlands. *Perinatal Epidemiology* 1991; **5:** 11-26.
15. Yach D, Gcinile B. Health status. In: *South African Health Review*. Durban: Health Systems Trust and the Henry J Kaiser Family Foundation, 1995: 27-52.
16. Moodley J, Raigopal M. Maternal and perinatal outcome associated with hypertensive crises of pregnancy. *Hypertension in Pregnancy* 1998; **17:** 43-53.
17. Moopanar D, Naidu S, Moodley J, Gouws E. Thromboelastography in abruptio placentae. *J Obstet Gynaecol* 1997; **17:** 229-233.
18. Rhys Williams AT. Simultaneous determination of serum vitamin A and E by liquid chromatography with fluorescence detection. *J Chromatogr Biomed Appl* 1985; **341:** 198-201.
19. Coutsoudis A, Broughton M, Coovadia HM. Vitamin A supplementation reduces measles morbidity in young African children: a randomised placebo-controlled, double-blind trial. *Am J Clin Nutr* 1991; **54:** 890-895.
20. Fawzi WW, Chalmers TC, Herrera MC, Mosteller F. Vitamin A supplementation and child mortality: A meta-analysis. *JAMA* 1993; **269:** 898-903.
21. Glasziou PP, Mackerras Dem. Vitamin A supplementation in infectious diseases: a meta-analysis. *BMJ* 1993; **306:** 366-370.
22. Roberts JR. Principles of neonatal pharmacology. In: Avery ME, Taeuscher HW. *Schaffer's Diseases of the Newborn*. 5th ed. Philadelphia: WB Saunders, 1984: 950-968.
23. Coutsoudis A, Adhikari M, Pillay K, Coovadia HM. Absorption of high-dose enteral vitamin A in low-birth-weight neonates. *S Afr Med J* 1996; **86:** 1337-1339.
24. The Vitamin A and Pneumonia Working Group. Potential interventions for the provision 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. *Bull World Health Organ* 1995; **73:** 609-619.
25. McQuire JM. *The Payoff From Improving Nutrition*. Draft Monograph. Washington: World Bank, 1990..

Accepted 25 Jan 2000.