Integration of Vitamin A Supplementation with the Expanded Program on Immunization Does Not Affect Seroconversion to Oral Poliovirus Vaccine in Infants[1]

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ABSTRACT Childhood immunization programs may provide infrastructure for delivering vitamin A supplements to infants in developing countries. The effect of giving vitamin A, an immune enhancer, on antibody responses to trivalent oral poliovirus vaccine (TOPV) is unknown. A randomized, double-blind, placebo-controlled clinical trial was conducted to determine the effect of giving vitamin A simultaneously with TOPV on antibody responses to poliovirus. Infants (*n* = 467) received oral vitamin A, 15 mg retinol equivalent (RE), 7.5 mg RE or placebo with TOPV at 6, 10 and 14 wk of age. Antibody responses to poliovirus types 1, 2 and 3 were measured by a microvirus neutralization assay at enrollment and at 9 mo of age. Seroconversion rates to poliovirus types 1, 2 and 3 ranged from 98 to 100% in the three treatment groups, and there were no differences in mean antibody titers to poliovirus types 1, 2 and 3 among treatment groups. This study demonstrates that oral vitamin A does not affect antibody responses to poliovirus vaccine when integrated with the Expanded Program on Immunization. J. Nutr. 129: 2203–2205, 1999.

KEY WORDS: • infants • vitamin A • immunization • poliovirus • vaccine

Vitamin A deficiency is a major cause of childhood morbidity and mortality in developing countries (Sommer and West 1996). Periodic high dose vitamin A supplementation has been shown to reduce child mortality by about one third (Beaton et al. 1993). The infrastructure of childhood immunization programs involves an estimated 500 million child contacts per year; it has been suggested that this infrastructure could be used for the delivery of micronutrient supplements to infants [Expanded Program on Immunization (EPI)[3] Global Advisory Group 1987]. Vitamin A, through its active metabolites, controls the transcriptional activation of many genes, is a potent modulator of many different pathways in the immune system and is known to influence responses to vaccination (Semba 1998). Oral high dose vitamin A given simultaneously with standard titer Schwarz measles vaccine may interfere with seroconversion to the measles virus in 6-mo-old infants who have high levels of maternal antibody to measles virus (Semba et al.1995), but not in 9-mo-old infants who have low levels or no maternal antibody to measles virus (Benn et al. 1997, Semba et al. 1997). Trivalent oral poliovirus vaccine (TOPV) is also a live attenuated vaccine, and there is concern that oral vitamin A could interfere with seroconversion to TOPV when given simultaneously in childhood immunization programs. We conducted a randomized, double-blind, placebo-controlled clinical trial to determine the effect of simultaneous vitamin A administration on antibody responses to TOPV in infants.

SUBJECTS AND METHODS

The study population consisted of infants in 27 villages in Bogor District, West Java, Indonesia, an area with a high prevalence of subclinical vitamin A deficiency (Semba 1995). After written, informed consent was obtained from the mother or father, infants were enrolled in the clinical trial between October 1992 and February 1993. Details of this clinical trial have been described elsewhere (Semba et al. 1997). Infants were randomly allocated by number table in blocks of 10 to receive 15 mg retinol equivalent (RE), 7.5 mg RE of vitamin A or placebo at 6, 10 and 14 wk of age. At age 6, 10 and 14 wk, infants received TOPV [Polioral, Biocine Sclavo, Siena, Italy, lot 73A12, 1.15 mL volume, tissue culture dose 50% infectivity ($TCID_{50}$) for type 1, 2 and 3: $10^{6.0}$, $10^{5.0}$, and $10^{5.8}$, respectively] and diphtheria-pertussis-tetanus vaccine (Biofarma, Bandung, Indonesia) administered by a pediatrician. Vitamin A and placebo solutions were dispensed from amber glass micronutrient dispensers (Swift dispensers, Englass, Leicester, U.K.) within 10–30 min after TOPV immunization. TOPV was not administered to the infants at birth. The cold chain was extensively monitored during shipping from the factory in Siena, Italy to the study laboratory in Bogor, Indonesia using temperature-sensitive monitors. All infants were seen and examined by a pediatrician at each visit. At the time of treatment allocation, both the pediatrician and study nurse were required to verify the identification number of the infant. Height, weight, mid upper-arm circumference and head circumference were measured by a well-trained anthropometrist when the infants were 6 and 14 wk of age.

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[3] Abbreviations used: EPI, Expanded Program on Immunization; RE, retinol equivalent; $TCID_{50}$ tissue culture dose, 50% infectivity; TOPV, trivalent oral poliovirus vaccine.

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TABLE 1

Characteristics of mothers and infants in Indonesia by allocation group

Characteristic[1,2]	Placebo (n = 156)	Vitamin A 7.5 mg RE (n = 156)	Vitamin A 15 mg RE (n = 155)
Maternal age, *y*	26.5 ± 0.5	26.2 ± 0.5	26.0 ± 0.5
Infant age, *d*	53.8 ± 0.8	53.1 ± 0.8	52.7 ± 0.9
Infant gender	82 M, 74 F	88 M, 68 F	86 M, 69 F
Weight, *kg*	4.75 ± 0.06	4.65 ± 0.05	4.74 ± 0.05
Height, *cm*	55.8 ± 0.2	55.8 ± 0.2	55.9 ± 0.2
Mid upper-arm circumference, *cm*	12.55 ± 0.08	12.35 ± 0.09	12.56 ± 0.09
Head circumference, *cm*	37.7 ± 0.1	37.5 ± 0.1	37.7 ± 0.2
Plasma retinol, baseline, *μmol/L*	0.61 ± 0.01	0.61 ± 0.01	0.61 ± 0.01
Plasma retinol, 14 wk, *μmol/L*	0.64 ± 0.01	0.73 ± 0.02[3]	0.76 ± 0.02[3]
Plasma retinol <0.35 μmol/L at baseline, %	10.97	11.04	10.97
Plasma retinol <0.35 μmol/L at 14 wk, %	7.19	1.40	1.38[4]

[1] No significant difference was noted among groups for all characteristics shown, except for plasma retinol at 14 wk. Plasma retinol was measured in 139, 143, and 145 subjects of placebo, 7.5 mg retinol equivalent (RE), and 15 mg RE vitamin A groups at 14 wk, respectively.
[2] For continuous variables, mean ± SEM.
[3] Paired *t* test, difference in plasma retinol between baseline and 14 wk, $P < 0.0001$.
[4] Vitamin A–supplemented groups different from placebo by Mantel-Haenszel χ-square, $P < 0.006$.

Blood was obtained by venipuncture at 6 and 14 wk, and 9 mo of age, and plasma was separated and frozen at −70°C. Plasma vitamin A levels were measured by HPLC at 6 and 14 wk (Semba et al. 1997). Antibody titers to poliovirus serotypes were measured by a microvirus neutralization assay (Kapsenberg et al. 1981) and were standardized against poliovirus reference sera (World Health Organization, Geneva, Switerland). Seroconversion to poliovirus was defined as a positive antibody titer at 9 mo of age (≥2), minus the calculated expected titer of passively acquired maternal antibody to poliovirus, assuming a half-life of immunoglobulin G to be 4 wk. The virus neutralizing antibody level considered to be consistent with protection against poliovirus at 9 mo of age was ≥8 (WHO Expert Committee on Biological Standardization 1988). Categorical analyses were conducted using χ-square and exact tests. Student's and Bonferroni's *t* tests were used for appropriate comparisons of continuous data. A paired *t* test was used to compare plasma vitamin A levels at baseline and follow-up. The study protocol was approved by the ethical review committees at the Johns Hopkins School of Medicine, the Ministry of Health, Government of Indonesia, and the WHO Secretariat Committee on Research Involving Human Subjects.

RESULTS

There were 467 infants enrolled in the study at age 6 wk. Follow-up rates of the infants to 10 and 14 wk and 9 mo were 93.6, 91.6 and 84.5%, respectively. Characteristics of infants in the three treatment groups are shown in **Table 1**. There were no significant differences in weight, height or other anthropometric measurements at baseline. Antibody titers to poliovirus were measured in 353 of 394 infants who were seen both at enrollment and at 9 mo of age. Plasma samples were unavailable for 41 infants because of insufficient sample volume. Seroconversion rates to the three poliovirus types by treatment group are shown in **Table 2**. The rates of seroconversion were 97–100%, with no significant differences among treatment groups in seroconversion to any of the three poliovirus types. The proportion of infants with titers ≥8, a level considered to confer protection against poliovirus, is shown in Table 2. The proportion of infants with protective titers against the three respective polioviruses ranged from 93.1 to 99.1%, with no significant differences in protection among treatment groups. Log_e antibody titers to the three poliovirus types did not differ by treatment group (**Table 3**). There were no significant differences in geometric mean titers to any of the poliovirus types among treatment groups. There were no cases of polio among the infants in the study during follow-up.

DISCUSSION

This study shows that vitamin A supplementation given concurrently with TOPV immunization at the 6-, 10- and 14-wk visits of the Expanded Program on Immunization does

TABLE 2

Seroconversion rates and protection against poliovirus types 1, 2, 3 in Indonesian infants

Poliovirus	Placebo	Vitamin A (25,000 IU)	Vitamin A (50,000 IU)	*P*-value
Seroconversion[1]				
Type 1	91/92 (98.9%)	109/110 (99.1%)	100/102 (98.0%)	0.77
Type 2	113/113 (100%)	124/124 (100%)	116/116 (100%)	1.00
Type 3	111/112 (99.1%)	122/122 (100%)	113/113 (100%)	0.99
Protection[2]				
Type 1	87/92 (94.5%)	104/110 (94.5%)	95/102 (93.1%)	0.88
Type 2	112/113 (99.1%)	122/124 (98.4%)	114/116 (98.3%)	0.84
Type 3	108/113 (95.6%)	121/123 (98.3%)	111/115 (96.5%)	0.45

[1] Detectable neutralizing antibody titer at 9 mo of age, adjusting for half-life of maternal antibody titer at enrollment.
[2] Antibody titer ≥8 at follow-up visit at 9 mo of age (World Health Organization Expert Committee on Biological Standardization 1988).

TABLE 3

Log_e antibody titers against poliovirus types 1, 2, 3 in 9-mo-old Indonesian infants[1,2]

Poliovirus	Placebo	Vitamin A 7.5 mg RE	Vitamin A 15 mg RE
Type 1	5.26 ± 0.16 [92]	5.63 ± 0.16[110]	5.32 ± 0.17[102]
Type 2	5.95 ± 0.12[113]	6.02 ± 0.12[124]	5.96 ± 0.11[116]
Type 3	5.36 ± 0.14[113]	5.44 ± 0.12[123]	5.21 ± 0.12[115]

[1] No significant differences noted between allocation groups.
[2] *n* in each group in brackets.
RE, retinol equivalent.

not interfere with seroconversion to any of the three polio types. In addition, there were no significant differences among the three treatment groups in the proportion of infants who had antibody titers considered protective against polio and in the mean antibody titers against the three polio types. In a different but related study design, vitamin A supplementation to mothers but not their infants did not interfere with seroconversion to oral poliovirus vaccine (Bhaskaram and Balakrishna 1998). Vitamin A supplementation given at the time of measles immunization has been shown to interfere with antibody responses to measles in infants with high levels of maternal antibody, but not in infants with little to no maternal antibody to measles (Benn et al. 1997, Semba et al. 1995 and 1997). The mode of delivery of vaccine (oral vs. subcutaneous) and type of virus may account for some of the differences between possible effects of vitamin A on immune responses to vaccination.

Although TOPV is generally considered effective, this study shows that it is possible to have higher rates of seroconversion and protection than those generally reported from developing countries using TOPV (Patriarca et al. 1991). Factors that might affect the efficacy of TOPV include the vaccine potency, vaccine stability, age at first dose, the interval between doses and dosage volume. The TOPV used in this study met the requirements for potency developed by the Expert Committee on Biologic Standardization (WHO Expert Committee 1988), and the cold chain was strictly monitored from the factory until the moment it was administered in the clinic. The age at first dose and interval between doses followed the guidelines of the EPI. This study suggests that strict adherence to WHO guidelines for potency and administration of TOPV can result in high rates of seroconversion and protection from polio in a developing country such as Indonesia.

Infancy is a high risk period for the development of micronutrient deficiencies in many developing world populations, and the integration of vitamin A supplementation with the EPI, in addition to periodic high dose vitamin A supplementation for preschool children, is proceeding in some countries in which vitamin A deficiency is a public health problem. Integration of vitamin A supplementation with the EPI improves vitamin A status of infants but seems to have no effect on infant morbidity and mortality (WHO/CHD Immunisation-Linked Vitamin A Supplementation Study Group 1998). The infrastructure of the EPI also offers the opportunity to supplement infants with oral iodized oil. In Indonesia, infants receiving oral iodized oil capsules given through the EPI at 6 wk of age had lower mortality rates, presumably because of an effect of oral iodized oil on thyroid status and immunity (Cobra et al. 1997). In the same trial, oral iodized oil did not interfere with seroconversion to TOPV when given through the EPI (Taffs et al. 1999).

This study suggests that oral vitamin A supplementation can be integrated with the EPI without an adverse effect on antibody responses to oral poliovirus vaccine and that high seroconversion rates can be achieved with careful monitoring of the cold chain. Integration of vitamin A with the EPI should be considered in countries in which clinical and subclinical vitamin A deficiency is endemic.

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