International Scholarly Research Network
ISRN Public Health
Volume 2012, Article ID 121697, 7 pages
doi:10.5402/2012/121697

Clinical Study

The Effects of Two Maternal Vitamin A Supplementation Regimens on Serum Retinol in Postpartum Mothers: A Randomised Controlled Trial in Brazil

Luciana Marques Andreto,[1,2,3] Ilma Kruze Grande de Arruda,[1,2] Ariani Impieri Souza,[2] José Natal Figueiroa,[2] and Alcides da Silva Diniz[1]

[1] *Diretoria de Pesquisas, Instituto de Medicina Integral Professor Fernando Figueira, Caixa Postal 1393, 50070-550 Recife, PE, Brazil*
[2] *Departamento de Nutrição, Centro de Ciências da Saúde, Universidade Federal de Pernambuco (UFPE), 50670-901 Recife, PE, Brazil*
[3] *Rua Joaquim Marques de Jesus, 86, apto 101, Piedade, 54420-240 Jaboatão dos Guararapes, PE, Brazil*

Correspondence should be addressed to Luciana Marques Andreto, lucianandreto@hotmail.com

Received 9 July 2012; Accepted 26 July 2012

Academic Editors: S. M. Ahmed and A. R. Mawson

Objective. To test whether the serum retinol level in mothers supplemented with 400,000 IU of vitamin A is higher than in those supplemented with 200,000 IU and to estimate duration of the protective effect of vitamin A supplementation in the serum retinol level. *Methods*. Double-blind, randomised controlled trial performed in two hospitals in the state of Pernambuco in northeast Brazil. Three hundred twelve mothers were recruited immediately postpartum. All women received a capsule containing 200,000 IU of vitamin A, and 10 days after delivery, they were randomly assigned to one of two treatment groups. One group received a second capsule containing vitamin A and the other group received a placebo. Each group was invited back after 2, 4, and 6 months for serum retinol analyses. *Results*. No difference was found between the two groups in serum maternal retinol concentration at 2 months (2.13 versus 2.03 μmol/L), 4 months (2.20 versus 2.24 μmol/L) or 6 months (2.29 versus 2.31 μmol/L). Because there was no further effect and because this population has a level of vitamin A deficiency considered mild, our results do not support a proposal to increase the dosing schedule for vitamin A in postpartum women as recommended by the IVACG.

1. Introduction

Estimates show that each year approximately 20 million pregnant women living in areas at risk for vitamin A deficiency (VAD) have low concentrations of retinol (<0.70 μmol/L) in blood. In the Americas and Brazil VAD prevalence estimates are around 8% and 5%, respectively [1].

The Brazilian Northeast is one of the poorest regions of the country with a 46% Human Poverty Index. A study in a big city of the Northeast region observed a 25% prevalence of inadequate serum retinol concentration among breastfeeding women [2].

The World Health Organisation (WHO) recommends mass supplementation with a megadose of vitamin A in the immediate postpartum period in countries where VAD is endemic [3]. Several randomised controlled clinical trials have shown that vitamin A supplementation in areas at risk for VAD can significantly reduce child mortality [4–6]. However, there is still no consensus on whether the vitamin A supplementation during pregnancy produces the same benefits in reducing maternal morbidity and mortality [7].

Although a strategy for supplementation during the immediate postpartum period has been implemented in several countries (including Brazil), there are still doubts regarding the most effective regimen. Investigations have warned that a dose of 200,000, IU seems to be insufficient to correct subclinical vitamin A levels in women and their children during the first six months of life [8, 9]. Thus, since 2002 the International Vitamin A Consultative Group-IVACG recommends supplementation with an additional megadose, with a fractional intake of a total of 400,000 IU of vitamin A during the safe period of infertility [9, 10].

This dose has been tested in some clinical trials developed in Africa and Asia, but there is no consensus as to the additional benefit of a dose higher than that recommended by the WHO [10–13].

The rationale for maternal supplementation with a megadose of vitamin A is that such a substantial supplementation, in addition to ensuring an adequate supply of vitamin A to the mother, helps to provide adequate levels of vitamin A to children via breast milk [14].

Therefore, the purpose of this study was to compare the effectiveness of supplementation with 400,000 IU versus 200,000 IU of vitamin A in the immediate postpartum period in regards to maternal serum retinol concentration within the first 6 months postpartum. As a secondary objective we sought to estimate the time of protective effect of vitamin A supplementation in the prevention of inadequate levels of vitamin A.

1.1. Description of Trial Design. A randomised, controlled, triple-blind, hospital-based clinical trial was conducted with women in the postpartum period divided into two supplementation groups to receive either 400,000 IU (200,000 + 200,000) or 200,000 IU (200,000 + placebo) of vitamin A.

1.2. Eligibility Criteria for Participants. Postpartum women aged between 13 and 42 years with a single foetus during pregnancy and at term. We excluded 20 mothers with HIV infections, diabetes mellitus, hypertensive disorders, and severe mental disorders.

1.3. Settings and Locations Where the Data Were Collected. The study was conducted from August 2007 to June 2009 in the public maternity hospitals *Maternidade Bandeira Filho* and *Instituto de Medicina Integral Prof. Fernando Figueira*-IMIP, both located in Recife, Brazil, and serving pregnant women that use the unified health system.

1.4. Sample Size. Sample size was calculated by the formula $N = (u + v)^2(dp_1{}^2 + dp_2{}^2)/(\mu_1 - \mu_2)^2$ [15] taking the assumptions of an α error = 5% (u), a β error = 20% (v), a standard deviation of the distribution of serum retinol concentrations in both groups (dp_1 and dp_2) equal to 0.5 μmol/L [16], and the intergroup difference between the retinol means $(\mu_1 - \mu_0)^2$ equal to 0.20 μmol/L; the minimum sample size for each group was 98 women. In order to allow for losses, a sample of 156 women was established for each group.

1.5. Selection of Participants. Were selected 330 women upon admission for delivery from August 2007 until August 2008. After clarification of the study objectives, 312 who agreed to participate signed a free and informed consent form.

1.6. Randomisation and Followup. During the immediate postpartum period, before hospital discharge, each woman received a capsule containing 200,000 IU vitamin A (Farmanguinhos/FIOCRUZ, Rio de Janeiro, RJ, Brazil) from a nurse, accordingly to the protocol of the Program of Vitamin A Supplementation developed by the Ministry of Health of Brazil. On the tenth day postpartum, randomisation was performed in the two individual treatment groups (group 1 and group 2) using a table of random numbers generated by EPI INFO version 6.04d (WHO/CDC, Atlanta, GE, USA), and the second capsule (vitamin A or placebo) was administered by the researcher during routine consultation at the clinic service.

All women were monitored to assess the presence of any side effects related to the toxicity of vitamin A during the three days following supplementation. No references to symptoms that could be related to these side effects were mentioned to women to avoid suggestibility about some of the symptoms.

During the consultations on the secon, forth and sixth postpartum months, blood samples were collected again for retinol and PCR analysis. Blood sample collection was carried out in fastening women and between 8 and 10 hours of the morning. The randomisation codes were kept confidential throughout the study and were only revealed after the completion of the data analysis. Flow chart selection, randomisation, and followup of participants are shown in Figure 1.

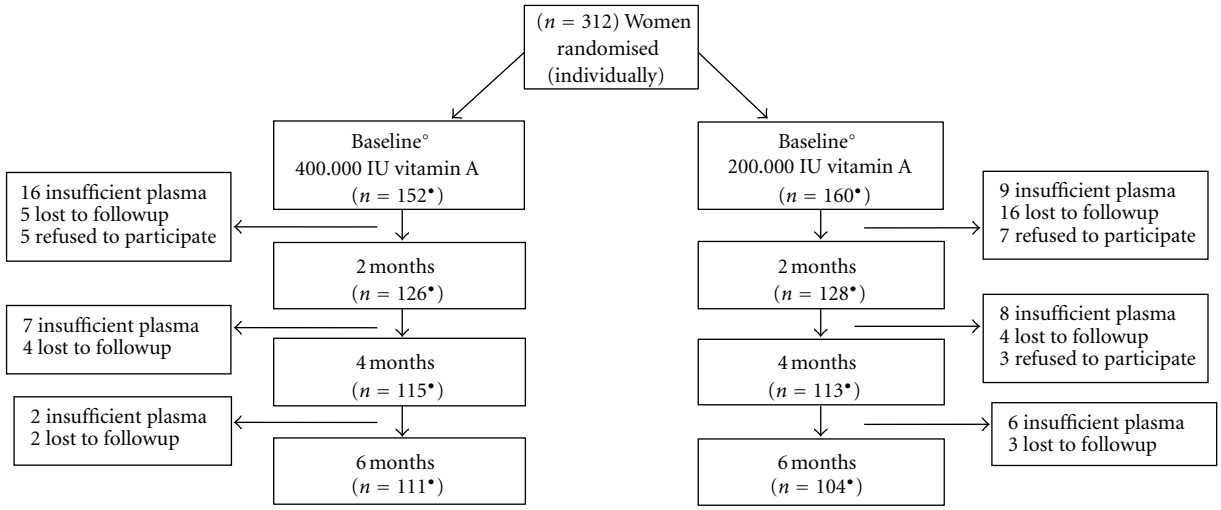
Breakdown of Groups: Group 1 received a capsule of 200,000 IU (retinyl palmitate) + 40 mg vitamin E orally immediately after delivery, and a second capsule of 200,000 IU (retinyl palmitate) + 40 mg vitamin E was administered 10 days postpartum.

Group 2 received a capsule of 200,000 IU (retinyl palmitate) + 40 mg vitamin E orally immediately after delivery, and a second "placebo" capsule was administered 10 days postpartum containing 40 mg of vitamin E dissolved in soybean oil.

The vitamin A capsules and the "placebo" capsules were prepared to be identical in size, shape, colour, and flavour by Relthy Laboratories (Indubatuba, SP-Brazil). The capsules contain vitamin E in order to prevent any toxic effects of vitamin A megadoses in addition to providing greater stability to the capsule.

2. Methods and Evaluation Techniques

All blood samples were obtained by a trained laboratory technician. For the analysis of serum retinol, 7 mL of blood was collected by puncturing the cubital vein with a tube protected from light; after complete coagulation, the samples were centrifuged and the serum was transferred to eppendorf tubes. The samples were then stored in a freezer at −20°C and were later transported to the Micronutrients Research Centre Paraíba Federal University (UFPB), kept cold at all times. Concentrations of serum retinol were analysed by high performance liquid chromatography (HPLC, model 305, Gilson, France) using the technique established by Furr et al. [17]. The intra- and inter-assay variations for low and high concentrations of retinol ranged from 2.4 to 6.7 for intra-assay and 4.3 to 8.5 for inter-assay replication. The detection limit was 1.25 (0.04). The cutoff established as an



°Haemoglobin; anthropometric evaluation; questionnaire with: dietary consumption; socioeconomic, demographic and obstetric variables.
•Serum retinol

Figure 1: Consort flowchart.

inadequate concentration of serum retinol was <1.05 μmol/L (<30 μg/dL) [11, 12, 18–22].

Haemoglobin concentrations were analysed with an electronic cell counter (Sysmex SF 3000 Automated Hematology Analyzer, GMI, Inc. Ramsey, MN, USA). The cut-off point adopted for the identification of anaemia was haemoglobin <12 g/dL [23].

Anthropometric measurements of weight and height were used to assess nutritional status and to calculate the body mass index (BMI). Socioeconomic, demographic, and obstetric variables were obtained by interview and confirmed with prenatal cards and clinical records. This questionnaire was administered no baseline to confirm the comparability between the groups.

Vitamin A consumption was evaluated using the Quantitative food frequency questionnaire previously validated [24], adapted to commonly used food in the region. This questionnaire was administered at the end of the trial and it takes into consideration the food consumption referring to the intake of different types of food in previously defined small, medium, or large portions, eaten daily, weekly, or monthly. The dietary consumption was analyzed using DietSys v. 4.01 (National Cancer Institute, Bethesda, MD, USA) that is based on Nutrient Database for Standard Reference [25]. The results of the vitamin A intake (retinol equivalent) were compared to the Dietary Reference Intakes (DRI) proposed by the Institute of Medicine [26].

The profile of breastfeeding was classified according to the categories recommended by the World Health Organization [27] as follows 1-Breastfeeding, feeding with breast milk, regardless of consumption of any supplement, dairy, or not. 2-Exclusive breastfeeding, feeding exclusively on breast milk without any other liquid or solid, but can receive vitamins, minerals, or medicines.

2.1. Statistical Methods. Data were entered in duplicate and verified with the "Validate" module of the statistical package Epi-info version 6.04d (WHO/CDC, Atlanta, GE, USA) for consistent verification and validation. Statistical analysis was performed using the Statistical Package for Social Sciences-SPSS for Windows, version 12.0 (SPSS Inc., Chicago, IL, USA) and Stata 10.1 (Statacorp, College Station, Texas, USA). All variables were normally distributed after the application of the Kolmogorov-Smirnov test and are presented as mean and standard deviation.

To compare means between groups, Student's t-test for unpaired data was used; for medians Mann-Whitney U test was used and to compare between proportions, the chi-square test and Fisher's exact test were used. The variation of retinol as a function of time and dosage was adjusted by linear regression models using the technique GEE (generalized estimation equations), which adjusts the parameter estimates and their standard errors, taking into account the possible correlation of repeated measurements. To avoid the occurrence of bias in estimating the temporal variation of retinol after administration of supplementation was introduced in the regression models an indicator variable of baseline measurements.

2.2. Ethical Aspects. The study was approved by the IMIP Ethics Committee (under number 720/2006) on January 27, 2006, and the trial was registered at www.clinicaltrials.gov under # NCT00742937. Anaemic women received treatments with ferrous sulphate at the doses recommended by the Brazilian Ministry of Health and were monitored by the health service. There were no formal rules for interruption of the clinical trial. If the woman wanted to leave the research, there would be no prejudice to her or her child's care at the health service.

3. Results

Of the 312 women recruited for the study, 215 completed the protocol. The distribution and reasons for dropout are shown in Figure 1. The comparison between the supplementation groups showed no significant differences regarding sociodemographic, obstetric, and nutritional characteristics at baseline. The study included women with a mean age of 24.6 years, 8.8 years of schooling, and US$ 92.82 per capita income (Table 1).

As observed in Table 2, the pattern of breastfeeding was similar in both supplementation groups during the monitoring period.

Adjustment of the linear regression model with a GEE (generalized estimation equations) method indicated that during the postpartum followup, the mean serum retinol concentrations were similar between the two supplementation groups ($P = 0.253$). There was no significant interaction between supplementation group and followup period ($P = 0.834$) (Table 3).

Combining the serum retinol concentration results of the two supplementation groups, we found that between baseline and two months postpartum, serum retinol concentrations increased (on average) 0.39 μmol/L (CI 95%: 0.27–0.51). Comparing the second and sixth months of followup, there was a significant increase in mean serum retinol concentration, estimated to be 0.045 μmol/L/month ($P = 0.001$). These results report both the adjusted and the unadjusted values (Table 4).

The results were similar when serum retinol concentrations were categorised, because the percentage of women with a low vitamin A concentration was homogeneous between the two supplementation groups (Table 5).

4. Discussion

The absence of a significant additional increase in serum retinol concentration in response to a 400,000 IU dose of vitamin A compared to a 200,000 IU dose is consistent with results reported by other studies [12, 13]. A potential explanation for this lack of a striking effect would be that the serum concentration of retinol is homeostatically controlled, and the serum levels will only be modified when hepatic storage is very high [28]. Therefore, a dose of 200,000 IU of vitamin A is sufficient to ensure an adequate supply of this vitamin, and a potential increase in serum level tends to be counteracted by homeostatic regulation. Accordingly, the serum retinol concentration may not be the most appropriate indicator for assessing the effect of this alternative dosing schedule, although it is the indicator recommended by the WHO and the IVACG to evaluate the organic status of vitamin A. Thus, a more appropriate evaluation of vitamin A would be the use of dose-response tests.

A second possible explanation for the lack of an additional effect on serum retinol concentration in women who receive a higher dose of vitamin A (400,000 IU) could be the fact that, in our study, the mean serum retinol concentration at baseline was considered to be at an acceptable level (1.61 μmol/L), suggesting that vitamin A deficiency in this population is mild; that is, 21.2% of women had inadequate serum retinol concentrations (1.05 μmol/L), of which about 70% of the retinol concentrations were considered to be marginal (0.70–1.05 μmol/L). Thus, it is plausible to expect a small impact on vitamin A levels considering the fact that a therapeutic response would be conditioned to the organic status of this micronutrient, that is, a smaller response for a smaller deficiency. However, other studies with similar designs are consistent with our data, wherein no significant effects were observed on retinol levels in maternal blood even among women with relatively low levels of serum retinol [10, 12, 13].

The increased concentrations of serum retinol in both supplementation groups (200,000 IU and 400,000 IU) over the six-month followup constituted a typical behaviour for serum retinol concentrations after supplementation; the literature shows that the use of vitamin A supplementation elevates postpartum serum retinol concentration over a course of three months and then gradually decreases [14, 29]. Stolftuz et al. attributes the decline in maternal serum retinol concentration after three months of supplementation to the transfer of large amounts of vitamin A from maternal storage to the child via breastfeeding [14].

In our study, one possible explanation for the nondecline and even the increase observed in serum retinol over the six months could lie in the pattern of breastfeeding seen in our research. Our results showed that the prevalence of exclusive breastfeeding decreased during the six months postpartum, with 38% of participants remaining in EB in the fourth month and only 19% in the sixth month. Thus, due to the early cessation of breastfeeding, these mothers were less depleted because of the lower transfer of maternal vitamin A (about 300 μg/day) storage to the infant via breastfeeding.

A supplementation strategy with megadoses (400,000 IU and 200,000 IU) during the immediate postpartum period was effective for reducing vitamin A deficiency considering that there was a decline in the prevalence of VAD from 21.1% to 2.35% over six months postpartum. On the other hand, the absence of any clinical manifestations that could be attributed to vitamin A toxicity [30] when administered at high dosages such as 400,000 UI shows the innocuity of vitamin A levels that have been administered in preventive action programs.

The above finding adds to the fact that the dietary intake of vitamin A was above the estimated average requirement (EAR), which tends to substantially increase hepatic reserves and consequently increase concentrations of circulating retinol.

A possible explanation for the increase of serum retinol levels at the end of the six months would be the fact that these women not only had a higher intake of vitamin A but also were less depleted due to breastfeeding interruption (exclusive breastfeeding rate at 4 months was only 40%). And it is also probable that these women received care in the health service and therefore have greater participation in the pharmacological supplementation program Ministry of Health. Due from this fact, they could be more protected

Table 1: Baseline characteristics from Brazilian lactating mothers enrolled in a vitamin A supplementation trial; Recife, Brazil—2007–2009.

Characteristics	400,000 UI vitamin A			200,000 UI vitamin A			*P* value*
	n	Mean	SD	*n*	Mean	SD	
Age (y)	152	24.6	5.8	160	24.3	6.3	0.208
Pregnancy number	152	2.1	1.5	160	2.0	1.4	0.883
Pregnancy interval (months)	150	31.5	39.9	158	25.2	34.6	0.678
Antenatal consultations (number)	151	6.3	2.7	156	6.1	2.2	0.287
Years of education	152	8.8	2.8	158	8.8	2.5	0.786
Income (US$)	147	230.6	142.5	156	222.9	128.8	0.641
Weight (kg)	123	71.5	12.4	131	71.1	12.5	0.391
Height (m)	128	1.6	0.1	138	1.6	0.1	0.235
BMI (kg/m^2)	113	28.5	4.2	121	28.3	4.6	0.809
Retinol (μmol/L)	114	1.62	0.74	115	1.60	0.69	0.440
Haemoglobin (g/dL)	133	11.6	1.7	144	11.3	1.5	0.086
Vitamin A consumption† [Med; IQ]	69	1090.4	817.7–1648.5	72	1164.7	841.0–1748.7	0.760**

Med: median. CI: confidence interval. IQ: interquartile interval.
†Dietary intake of vitamin A in equivalent of retinol.
*Student's *t*-test for unpaired data.
**Mann-Whitney *U* test.

Table 2: Duration of breastfeeding in Brazilian lactating mothers enrolled in a vitamin A supplementation trial; Recife, Brazil—2007–2009.

Followup	400,000 IU vitamin A		200,000 IU vitamin A		*P* value*
	n	%	*N*	%	
2 months					
Exclusive breastfeeding	77	65.3	67	56.8	0.313
Breastfeeding	34	28.8	46	39.0	0.367
4 months					
Exclusive breastfeeding	46	41.1	39	35.1	0.610
Breastfeeding	52	46.4	45	40.5	0.542
6 months					
Exclusive breastfeeding	20	17.7	23	20.7	0.813
Breastfeeding	72	63.7	56	50.5	0.114

* Pearson's chi-square test.

Table 3: Serum retinol concentration in Brazilian mothers enrolled in a postpartum supplementation trial. Recife, Brazil—2007–2009.

Group	Time postpartum			
	Baseline mean ± SD (*n*) (μmol/L)	2 months mean ± SD (*n*) (μmol/L)	4 months mean ± SD (*n*) (μmol/L)	6 months mean ± SD (*n*) (μmol/L)
400,000 IU vitamin A	1.62 ± 0.74 (114)	2.04 ± 0.67 (119)	2.10 ± 0.71 (103)	2.14 ± 0.57 (95)
200,000 IU vitamin A	1.60 ± 0.69 (115)	1.96 ± 0.63 (117)	2.22 ± 0.55 (110)	2.22 ± 0.55 (105)

SD: Standard deviation. Linear regression model with GEE methods, interaction between treatment groups ($P = 0.253$).

Table 4: Estimated values* of serum retinol means in the combined group during the monitoring period. Recife, Brazil—2007–2009.

	Time postpartum			
	Baseline mean ($CI_{95\%}$) (μmol/L)	2 months mean ($CI_{95\%}$) (μmol/L)	4 months mean ($CI_{95\%}$) (μmol/L)	6 months mean ($CI_{95\%}$) (μmol/L)
Combined group	1.61 (1.52–1.70)	2.01 (1.93–2.09)	2.11 (2.05–2.16)	2.19 (2.13–2.27)
Combined group*	1.61 (1.52–1.70)	2.00 (1.92–2.08)	2.12 (2.03–2.20)	2.18 (2.11–2.26)

CI: Confidence interval. *Obtained by adjustment of linear regression model with GEE methods.

Table 5: Serum retinol concentration in Brazilian mothers enrolled in a postpartum supplementation trial at baseline and during followup at 2, 4, and 6 months after treatment with 200,000 UI or 400,000 IU vitamin A; Recife, Brazil—2007–2009.

Time postpartum	400,000 UI vitamin A Serum retinol < 1.05 μmol/L			200,000 UI vitamin A Serum retinol < 1.05 μmol/L			P value
	N	n	%	N	n	%	
Baseline	123	25	20.3	123	27	22.0	0.755*
2 months	126	9	7.1	128	10	7.8	0.839**
4 months	115	5	4.3	113	10	8.8	0.170**
6 months	111	2	1.8	104	03	2.9	0.599**

* X^2 Pearson test. ** Fisher's exact test.

from this nutritional disorder in comparison to women's population, in general.

Our results cannot be extrapolated to populations where VAD has a moderate or severe level because this clinical trial was conducted in an environment particularly favourable for this investigation where vitamin A deficiency was considered mild. Because there was no further effect and because the level of vitamin A deficiency in the studied population was considered mild, our results support the idea of maintaining the prevention strategy recommended by the WHO.

Conflict of Interests

None of the authors of this paper has any conflict of interests to disclose related to employment, consultancies, honoraria, stock, expert testimony, patents, royalties, or any other relationships related to this project.

Acknowledgments

The authors thank the Ministry of Science and Technology and Ministry of Health from Brazil for the financial support and thank the CIMICROM (micronutrients research center) for the retinol analyses. Relthy Laboratories prepared and supplied the vitamin A placebo capsules. The authors also thank women who participated in the survey for their willingness to be involved in the study.

References

[1] K. J. West, "Extent of vitamin A deficiency among preschool children and women of reproductive age," *Journal of Nutrition*, vol. 32, pp. 2857S–2866S, 2002.

[2] R. E. Lopes, K. S. Ramos, C. C. Bressani, K. Arruda, and A. I. Souza, "Prevalência de anemia e hipovitaminose A em puérperas do Centro de Atenção à Mulher do Instituto Materno Infantil Prof. Fernando Figueira, IMIP: um estudo piloto," *Revista Brasileira De Sau'De Materno Infantil*, vol. 6, supplement 1, pp. S63–S68, 2006.

[3] World Health Organization, *Safe Vitamin A Dosage during Pregnancy and Lactation. Recommendations and Report of a Consultation*, WHO/ NUT, Geneva, Switzerland, 1998.

[4] G. H. Beaton, R. Martorell, K. A. L'Abbe et al., *Effectiveness of Vitamin A Supplementation in the Control of Young Child Morbidity and Mortality in Developing Countries. Final Report To CIDA*, University of Toronto, Toronto, Canada, 1993.

[5] W. W. Fawzi, T. C. Chalmers, M. G. Herrera, and F. Mosteller, "Vitamin A supplementation and child mortality: a meta-analysis," *Journal of the American Medical Association*, vol. 269, no. 7, pp. 898–903, 1993.

[6] P. P. Glasziou and D. E. M. Mackerras, "Vitamin A supplementation in infectious diseases: a meta-analysis," *British Medical Journal*, vol. 306, no. 6874, pp. 366–370, 1993.

[7] N. van den Broek, R. Kulier, A. M. Gülmezoglu, and J. Villar, "Suplementación con vitamina A durante el embarazo (Revisión Cochrane traducida)," *Cochrane Plus*. In press.http://www.update-software.com.

[8] A. L. Rice, R. J. Stoltzfus, A. De Francisco, J. Chakraborty, C. L. Kjolhede, and M. A. Wahed, "Maternal vitamin A or β-carotene supplementation in lactating Bangladeshi women benefits mothers and infants but does not prevent subclinical deficiency," *Journal of Nutrition*, vol. 129, no. 2, pp. 356–365, 1999.

[9] A. Sommer and F. R. Davidson, "Assessment and control of vitamin A deficiency: the annecy accords," *Journal of Nutrition*, vol. 132, no. 9, pp. 2845S–2850S, 2002.

[10] D. A. Ross, "Recommendations for vitamin A supplementation," *Journal of Nutrition*, vol. 131, pp. 2902S–2906S, 2002.

[11] L. C. Malaba, P. J. Iliff, K. J. Nathoo et al., "Effect of postpartum maternal or neonatal vitamin A supplementation on infant mortality among infants born to HIV-negative mothers in Zimbabwe," *American Journal of Clinical Nutrition*, vol. 81, no. 2, pp. 454–460, 2005.

[12] S. K. Tchum, S. A. Tanumihardjo, S. Newton et al., "Evaluation of vitamin A supplementation regimens in Ghanaian postpartum mothers with the use of the modified-relative-dose-response test," *American Journal of Clinical Nutrition*, vol. 84, no. 6, pp. 1344–1349, 2006.

[13] M. K. Darboe, D. I. Thurnham, G. Morgan et al., "Effectiveness of an early supplementation scheme of high-dose vitamin A versus standard WHO protocol in Gambian mothers and infants: a randomised controlled trial," *The Lancet*, vol. 369, no. 9579, pp. 2088–2096, 2007.

[14] R. A. Ayah, D. L. Mwaniki, P. Magnussen et al., "The effects of maternal and infant vitamin A supplementation on vitamin A status: a randomised trial in Kenya," *British Journal of Nutrition*, vol. 98, no. 2, pp. 422–430, 2007.

[15] B. Kirkwood and J. Sterne, *Essential Medical Statistics*, Blackwell Science, Oxford, UK, 2nd edition, 2003.

[16] H. Flores, M. N. A. Azevedo, F. A. C. S. Campos et al., "Serum vitamin A distribution curve for children aged 2–6 y known to have adequate vitamin A status: a reference population," *American Journal of Clinical Nutrition*, vol. 54, no. 4, pp. 707–711, 1991.

[17] H. C. Furr, A. J. Clifford, and A. D. Jones, "Analysis of apocarotenoids and retinoids by capillary gas chromatography-mass spectrometry," *Methods in Enzymology*, vol. 213, pp. 281–290, 1992.

[18] E. Accioly and S. Souza-Queiróz, "Deficiencia de vitamina A en embarazadas asistidas en una maternidad pública en Rio de Janeiro, Brasil," *Rev Chilena Nutrición*, vol. 27, pp. 352–357, 2001.

[19] E. Casanueva, R. Valdés-Ramos, F. Pfeffer, A. Ricalde-Moreno, E. García-Villegas, and C. Meza, "Serum retinol in urban Mexican women during the perinatal period," *Salud Publica de Mexico*, vol. 41, supplement 4, pp. 317–321, 1999.

[20] A. Rice, "Postpartum vitamin A supplementation: evaluating the evidence for action," A2Z Project Technical Brief, 2007, http://www.a2zproject.org/.

[21] P. H. Rondó, B. S. Villar, and A. M. Tomkins, "Vitamin A status of pregnant women assessed by a biochemical indicator and a simplified food frequency questionnaire," *Archivos Latinoamericanos de Nutricion*, vol. 49, no. 4, pp. 322–325, 1999.

[22] R. J. Stoltzfus, M. Hakimi, K. W. Miller et al., "High dose vitamin A supplementation of breast-feeding Indonesian mothers: effects on the vitamin A status of mother and infant," *Journal of Nutrition*, vol. 123, supplement 4, pp. 666–675, 1993.

[23] World Health Organization, *Iron Deficiency Anaemia: Assessment, Prevention, and Control: a Guide for Programme Managers*, World Health Organization, Geneva, Switzerland, 2001.

[24] Z. J. Salas, F. Faz-Cepeda, and L. N. B. Castañón, "Consumo de folatos de mujeres en edad fértil de Apocada, N.L. México," *Revista Salud Publica y Nutrición*, vol. 4, no. 4, 2003.

[25] U.S. Department of Agriculture, Agricultural Research Service, "USDA Nutrient Data Laboratory," 2001, Composition of Foods Raw, Processed, Prepared. Nutrient Database for Standard Reference, Release 14.

[26] Institute of Medicine. National, *Dietary Reference Intakes for Thiamin, Riboflavin, Niacin, Vitamin B6, Folate, Vitamin B12, Pantothenic Acid, Biotin, and Choline*, National Academy Press, Washington, DC, USA, 2002.

[27] Organización Mundial de la Salud, *Indicadores Para Evaluar Las Prácticas De Alimentación Del Lactante Y Del Niño Pequeño: Conclusiones De La Reunión De Consenso Llevada a cabo Del 6 Al 8 De Noviembre De 2007*, OMS, Washington, DC, USA, 2009.

[28] S. A. Tanumihardjo, "Assessing vitamin A status: past, present and future," *Journal of Nutrition*, vol. 134, no. 1, pp. 290S–293S, 2004.

[29] S. K. Roy, A. Islam, A. Molla, S. M. Akramuzzaman, F. Jahan, and G. Fuchs, "Impact of a single megadose of vitamin A at delivery on breastmilk of mothers and morbidity of their infants," *European Journal of Clinical Nutrition*, vol. 51, no. 5, pp. 302–307, 1997.

[30] L. H. Allen and M. Haskell, "Estimating the potential for vitamin A toxicity in women and young children," *Journal of Nutrition*, vol. 132, no. 9, pp. 2907S–2919S, 2002.