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# Lipid-Based Nutrient Supplements Providing Approximately the Recommended Daily Intake of Vitamin A Do Not Increase Breast Milk Retinol Concentrations among Ghanaian Women[1–3]

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## Abstract

**Background:** Vitamin A deficiency remains a global public health problem. Daily supplementation with a lipid-based nutrient supplement (LNS) has potential for increasing milk vitamin A concentrations.

**Objective:** The objective of this study was to determine whether daily supplementation with approximately the recommended daily intake of vitamin A in an LNS or a multiple-micronutrient supplement (MMN) during pregnancy and the first 6 mo postpartum has an effect on breast milk retinol concentration at 6 mo postpartum.

**Methods:** Women ≤20 wk pregnant ($n$ = 1320) were randomly assigned to receive either the MMN providing 18 micronutrients, including 800 μg retinol equivalents of vitamin A, or the LNS with the same nutrients as the MMN group, plus 4 minerals and macronutrients, until 6 mo postpartum; a control group received iron and folic acid during pregnancy and a placebo (calcium tablet) during the first 6 mo postpartum. Breast milk samples collected at 6 mo postpartum were analyzed for retinol and fat concentrations by HPLC and creamatocrit, respectively, in a subsample of 756 women.

**Results:** The breast milk retinol concentration was (mean ± SD) 56.3 ± 2.1 nmol/g fat, with no significant differences between groups [iron and folic acid ($n$ = 243): 59.1 ± 2.8; MMN ($n$ = 260): 55.4 ± 2.5; LNS ($n$ = 253): 54.7 ± 2.5 nmol/g fat; $P$ = 0.45], regardless of whether the woman had or had not received a high-dose vitamin A supplement (200,000 IU) soon after childbirth. Around 17% of participants had low milk retinol (≤28 nmol/g fat). We estimated that 41% of infants were potentially receiving vitamin A at amounts above the Tolerable Upper Intake Level (600 μg retinol activity equivalents/d), with no group differences in percentages with low or high milk retinol concentration.

**Conclusion:** Daily consumption of approximately the recommended intake of vitamin A did not increase breast milk retinol concentrations in this sample of Ghanaian women. This trial was registered at clinicaltrials.gov as NCT00970866. *J Nutr* 2016;146:335–42.

**Keywords:** micronutrient supplement, vitamin A, deficiency, pregnancy, lactation, breast milk, children

## Introduction

Vitamin A deficiency, defined by serum retinol concentration <0.70 μmol/L (1), remains a major public health problem in many parts of the world and affects 190 million preschool children and 19 million pregnant women (2). In Ghana, about 76% of preschool children (2.42 million children) had serum retinol concentrations <0.70 μmol/L in 2005, indicating a severe public health problem (2), and 19% of Ghanaian pregnant women (127,000 pregnant women) had a serum retinol concentration <0.70 μmol/L, indicating a moderate public health problem (2). The consequences of vitamin A deficiency include stillbirth and malformation (3, 4) and increased risk of morbidity and mortality due to infectious diseases, particularly in children (5–8).

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[3] Supplemental Table 1 is available from the "Online Supporting Material" link in the online posting of the article and from the same link in the online table of contents at jn.nutrition.org.

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Measures to control vitamin A deficiency among vulnerable populations include dietary diversification, food fortification with preformed vitamin A, and biofortification with provitamin A carotenoids (2). In addition, periodic supplementation with high-dose vitamin A is recommended by the WHO to reduce mortality in children and has been the most widely used intervention, reaching millions of children in many developing countries, including Ghana. The usual recommendation is for infants 6–11 mo of age to be supplemented once a year with 100,000 IU vitamin A and for children 12–59 mo of age to be supplemented with 200,000 IU vitamin A twice a year (9), usually through national health days and the Expanded Program of Immunizations in many countries. Based on current evidence linking vitamin A supplementation of postpartum women to a lack of effect on morbidity and mortality among infants and young children, the WHO does not recommend vitamin A supplementation of postpartum women for prevention of morbidity and mortality in women and children (10).

Studies that documented the impact of high-dose vitamin A supplements (200,000–400,000 IU) given to women soon after giving birth have reported increased breast milk retinol concentration lasting from 4 wk to 6 mo postpartum, with no impact detectable at 6–9 mo postpartum or beyond (11–13). In Ghana, up to 60% of women have reported receiving high-dose postpartum vitamin A supplements (14). Other studies in which lower doses of vitamin A (2033–2700 IU/d) were given regularly to women reported increases in milk vitamin A concentrations lasting from 10 wk to 11 mo postpartum (15–17).

Animal studies have shown that vitamin A is delivered to the mammary gland as retinol by retinol-binding protein and as retinyl esters by chylomicrons (18, 19); chylomicrons provide at least one-third to one-half of vitamin A to mammary gland tissue during lactation (18). Because vitamin A in chylomicrons comes directly from dietary sources, milk retinol varies with dietary vitamin A intake even when plasma retinol concentrations are relatively unaffected in rats (20). Thus, supplementing breastfeeding women may be a good strategy for increasing vitamin A status of breastfed infants.

Providing a daily adequate supply of vitamin A via breast milk is likely to sustain infant vitamin A status better than an intermittent supply of vitamin A provided as high-dose supplements. Kinetic models suggest that liver stores of vitamin A increase temporarily in response to high-dose supplements, and those stores become depleted between 1 and 6 mo after dosing, whereas liver stores are maintained in children who receive vitamin A in breast milk daily, assuming the breast milk contains adequate amounts of vitamin A (21). The mean daily dietary intake of vitamin A among unsupplemented women in developing countries [660 retinol equivalents (RE)[7] of vitamin A/d] is less than half the intake among women in developed countries (1540 RE/d) (22) and less than the North American [1200 retinol activity equivalents (RAE)/d for ages 14–18 y and 1300 RAE/d for ages 19–50 y] (23) and FAO/WHO (850 μg RE/d) (24) recommended intakes for lactating women.

The International Lipid-based Nutrient Supplements (iLiNS) Project is investigating the use of lipid-based nutrient supplements (LNSs) for women and children to improve nutritional status and child growth. LNSs are made using vegetable oil, powdered milk, groundnut paste, sugar, and multiple micronutrients and are intended for home fortification of maternal and child diets (25). LNSs are highly acceptable among Ghanaian pregnant and lactating women (26). The aims of this study were to *1*) assess the effect of an LNS providing daily doses of vitamin A to Ghanaian women on their breast milk retinol concentration at 6 mo postpartum and *2*) assess whether there is any interaction between intake of daily doses of vitamin A and high-dose postpartum vitamin A supplementation with respect to breast milk retinol concentration.

## Methods

*Study design.* This study formed part of the iLiNS-DYAD study in Ghana, a randomized, partially double-blind, controlled trial targeting women during pregnancy (≤20 wk of gestation to delivery) and the first 6 mo postpartum and their infants thereafter. A detailed description of the iLiNS-DYAD study is provided elsewhere (27). The 3 intervention groups were *1*) daily supply of an LNS providing 800 μg RE of vitamin A from enrollment to 6 mo postpartum (LNS group), *2*) daily supply of a multiple micronutrient supplement (MMN) providing 800 μg RE of vitamin A from enrollment to 6 mo postpartum (MMN group), and *3*) control group given a daily supply of iron and folic acid (IFA) only during pregnancy and a calcium placebo tablet during the first 6 mo postpartum (IFA group).

The MMN and LNS were modeled on the International Multiple Micronutrient Preparation (28) and that used in Guinea Bissau (29). The LNS was produced by Nutriset S.A.S. as individual 20-g sachets, and the IFA, MMN, and placebo were provided by DSM South Africa as capsules in blister packs. In Ghana, 2 independent individuals color-coded each type of capsule as 3 different supplements to maintain blinding.

*Study area, enrollment and consent process, and baseline data collection.* The study was conducted between 2009 and 2013 in the Manya Krobo and Yilo Krobo districts of the Eastern Region of Ghana. The major occupations in the study area are farming, trading, fishing, and vocational jobs.

Potential participants were pregnant women attending antenatal clinics at 2 hospitals and a polyclinic from December 2009 to December 2011 and a health post from September 2010 to December 2011. Women were invited for screening if they were aged ≥18 y, were ≤20 wk of gestation (as determined at the antenatal clinics mostly based on fundal height), and had antenatal cards with a complete history and examination. Women were excluded if their antenatal cards indicated HIV infection, asthma, tuberculosis, epilepsy, or any malignancy requiring frequent medical attention. Additional exclusion criteria were not residing in the study area, intention to move within the next 2 y, not willing to receive field workers or take study supplements, known allergy to peanut or milk, concurrent participation in another clinical trial, or gestational age >20 wk at the time of enrollment.

Eligible women were contacted at home for their consent, after which they immediately completed baseline laboratory (hemoglobin and malaria parasitemia) and anthropometric assessments, unless their gestational age at screening was based on fundal height and was >18 wk; in that case, women completed baseline measurements only if an ultrasound-confirmed gestational age was ≤20 wk. Women who were anemic or tested positive for malaria were referred to the study hospitals and clinic for treatment and counseling. Women whose gestational age at screening was <18 wk based on fundal height underwent an ultrasound scan immediately after enrollment. At 2–3 d after enrollment, home visitors completed 7-d qualitative food group recalls covering consumption of various types of foods, including red palm oil and typical dishes prepared with red palm oil. The food group recall was repeated at 36 wk of gestation and at 6 mo postpartum. These recalls were intended to describe consumption at the group level. Ethical approvals for the study were obtained from the Institutional Review Boards of the University of California, Davis and the Noguchi Memorial Institute for Medical Research, University of Ghana and the Ghana Health Service. Written consent was obtained from each participant after the nature and purpose of the study had been fully explained to them.

This trial was registered at clinicaltrials.gov as NCT00970866.

[7] Abbreviations used: IFA, iron and folic acid supplement; iLiNS, International Lipid-based Nutrient Supplements Study; LNS, lipid-based nutrient supplement; MMN, multiple micronutrient supplement; RAE, retinol activity equivalents; RE, retinol equivalents; UL, Tolerable Upper Intake Level.

*Randomization and masking.* Women were enrolled by random assignment to 1 of the 3 intervention groups immediately after baseline laboratory assessment. Randomization was based on a computer-generated scheme (SAS, version 9.3; SAS Institute) in blocks of 9. Numbered sheets with group allocation were placed in opaque envelopes and stacked in increasing order, from 1 to 1320. For each enrollment, the 9 topmost envelopes in the stack (except toward the end of enrollment when few envelopes were left) were shuffled, and the woman was asked to pick 1 to reveal her group allocation. Only the field supervisor in Ghana and the study statistician at the University of California, Davis had access to the password-protected file with information on which women in the full sample received which particular capsules or LNS. Dummy codes were assigned to the intervention groups during data collation and analysis, so the study statistician remained blinded to the women's group assignments until analyses were completed.

*Supplement delivery and intake.* Each woman received a 2-wk supply of the supplement to take home and was instructed to take 1 supplement every day. Women receiving capsules were instructed to take 1 capsule with water after a meal, and women receiving the LNS were instructed to take the entire contents of 1 sachet mixed with any food. All women were encouraged to consume diverse diets, including meat, fish, eggs, fruit, and vegetables. Home visit teams visited the women biweekly to deliver supplements, document adherence, and collect morbidity data until 6 mo postpartum. At 6 mo postpartum, the women made a second laboratory visit for breast milk collection.

*Breast milk sample collection, storage, and transportation.* Casual "spot" breast milk samples were collected at 6 mo postpartum, after feeding the infant for 1 min on that breast, by manual expression into plastic tubes that were wrapped in aluminum foil to protect from light. Approximately 10 mL breast milk was collected from each participant. The breast milk was homogenized by inverting the container at least 10 times. About 2 mL of each milk sample was immediately aliquoted into 5 polypropylene tubes labeled with the woman's unique identification number, and one set of tubes was designated for milk vitamin A and fat analysis. The milk samples were temporarily stored at −20°C in the laboratory and transferred on ice to a −80°C freezer for storage. Frozen breast milk samples were transported in dry ice to the University of California, Davis, where they were stored at −80°C until analyzed for vitamin A and fat.

*Milk vitamin A analysis.* Breast milk vitamin A was measured under dim light using an HPLC system (Class VP; Shimadzu Corporation), as previously described (30). Retinal (O-ethyl) oxime was synthesized (31) and added to each sample vial as an internal standard. Retinol was measured using a 3-μm, C-18 column (Adsorbosphere HS; Grace Industries) at a flow rate of 1 mL/min with a mobile phase of 85% (85% acetonitrile: 15% methanol) and 15% isopropanol. Retinol was detected at a wavelength of 325 nm using a photo-diode array detector (Class VP; Shimadzu Corporation).

Three control plasma samples (UTAK Laboratories) were extracted, saponified, and analyzed according to the same procedures with each batch of milk samples. The UTAK control plasma was calibrated for retinol by using control serum (fat-soluble vitamins) from the National Institute of Standards and Technology (SRM 968c) that is certified for the retinol concentration. The mean within-day and between-day CV for the retinol concentration of the UTAK control plasma was 2.9% and 9.3%, respectively.

Breast milk retinol concentration was initially calculated per volume (μmol/L). However, because casual breast milk samples were collected from participants, breast milk retinol concentration was also calculated per gram of fat (nmol/g) by dividing the retinol concentration per volume (μmol/L) by the milk fat concentration (g/L) and multiplying the result by 1000. This was done because the fat content of milk varies within a feeding episode and is low in fore milk and high in hind milk. Retinol is found in milk fat, and the fat content of casual spot milk samples is variable, depending on when the milk is collected within a feeding episode. Thus, milk retinol is expressed per gram of fat to account for variability in the fat content (32, 33).

According to criteria established by the WHO, milk retinol concentrations ≤1.05 μmol/L or ≤28 nmol/g fat indicate low breast milk retinol concentration (32). It is estimated that a breast milk retinol concentration of 1.05 μmol/L (28 nmol/g fat) will meet the metabolic needs of infants 0–6 mo of age who are exclusively or almost exclusively breastfed, with little or no effect on their liver vitamin A stores (32, 34), whereas a breast milk retinol concentration of 1.75 μmol/L (46.7 nmol/g fat) will both meet the metabolic needs of infants and build their liver vitamin A stores (33).

Because it is possible that milk retinol concentrations may be substantially higher than 1.75 μmol/L, the potential exists for breast milk to provide excessive vitamin A to exclusively breastfed infants at 6 mo of age. The WHO global recommendation on breastfeeding states that infants should be exclusively breastfed for the first 6 mo of life to achieve optimal growth, development, and health (35). It is estimated that exclusively breastfed infants in developing countries consume a mean ± SD of 753 ± 137 mL breast milk/d (36). At this level of intake, a milk retinol concentration of ≥2.79 μmol/L will result in infant vitamin A intakes that exceed the Tolerable Upper Intake Level (UL) of 600 μg RAE/d at 6 mo of age (23). The UL is defined as the highest level of daily nutrient intake that is likely to pose no risks of adverse health effects for almost all individuals in the general population; for infants, the UL is one-tenth of the lowest observable adverse effect level, the lowest intake at which bulging of the fontanel and increased intracranial pressure have been reported, which is 6000 μg RAE/d for infants at 0–6 mo of age (23). A milk retinol concentration of 2.79 μmol/L translates into 74.4 nmol retinol/g fat, using a mean daily milk fat concentration of 37.5 g/L. This is based on the value used to convert the cutoff value of 1.05 μmol/L to μmol/g fat (1.05/37.5 = 0.028 μmol/g fat) (32). Similarly, a milk retinol concentration of 91.2 nmol/g fat or 62.9 nmol/g fat would result in infant vitamin A intakes that exceed the UL, when milk intakes are 616 mL/d or 890 mL/d, respectively (mean ± SD of milk intake: 753 ± 137 mL/d). The proportions of mothers with milk vitamin A concentrations that could potentially result in infant vitamin A intakes that exceed the UL, based on milk intakes of 616 mL/d, 753 mL/d, and 890 mL/d, were compared among the intervention groups.

*Milk fat determination.* The fat content of the milk samples was measured using the Medela Creamatocrit Plus centrifuge (Separation Technology, Inc.), using the creamatocrit method (37). The mean milk fat concentration was calculated using the 3 measurements for each breast milk sample. The Creamtocrit Plus centrifuge was calibrated before each use by using a test strip with preset creamatocrit values supplied by the manufacturer. The overall mean CV for within-individual milk fat determinations was 9.3%.

*Sample size.* The initial sample size for the outcome of breast milk retinol concentration was based on an ability to detect differences between groups equivalent to an effect size (difference between groups divided by the pooled SD) of 0.5, based on results from a randomized controlled trial that provided vitamin A supplements to lactating women in Vietnam (6 d per week for 10 wk) (15), with 80% power and a level of significance (α) of 0.05. The required sample size per group was 79, for a total of 237; allowing for a 20% attrition rate, the initial target sample size was 285.

To assess the interaction effect of low-dose vitamin A and high-dose postpartum vitamin A on breast milk retinol, we increased the sample size to allow detection of differences between groups after stratification by high-dose postpartum vitamin A consumption, with an effect size of ≥0.50 with 80% power and α of 0.10. The calculated sample size was 125 participants per group, making a total of 750 participants (6 groups). We decided to use samples from the first ~750 women who provided usable milk samples, whose samples were available to the laboratory during the period of breast milk vitamin A analysis.

For the main iLiNS Ghana trial (27), the original sample size was 288 per group, for a total sample size of 864. Due to a temporary mislabeling of study capsules between 14 April 2010 and 30 September 2010, 92 women in the MMN group and 82 women in the IFA group received both the MMN and IFA for some period during pregnancy. The Data Safety and Monitoring Board and funder agreed to enroll more women to ensure sufficient statistical power, even if women with the mixed exposure were excluded. The sample size was recalculated to 440 women per group, assuming 20% attrition, for a total sample size of 1320. Thus, the analyses herein are based on women representing ~57% of the entire cohort enrolled.

*Statistical analysis.* Statistical analysis was done using SAS version 9.4 (SAS Institute). Continuous outcome variables were checked for normality (Shapiro-Wilk >0.97) and transformed when appropriate before statistical testing.

Analysis was by intention to treat. Data for all women whose milk samples were analyzed for vitamin A and fat concentrations were included. As a sensitivity analysis, breast milk vitamin A concentrations were compared between women who received mixed supplements and those who received the correct supplements. We compared maternal characteristics between women whose milk samples were analyzed and women whose milk samples were not analyzed to determine whether women whose milk samples were analyzed were representative of the whole cohort, by using *t* tests and $\chi^2$ tests. ANOVA and $\chi^2$ tests were used to compare maternal characteristics among the different intervention groups. Values in the text are means ± SD/SE and proportions (%).

The main effects of the intervention on mean milk vitamin A concentration at 6 mo postpartum were assessed by using ANOVA. ANCOVA was used to adjust for variables significantly related to milk vitamin A and fat concentrations to increase the power to detect statistically significant differences. Significant covariates ($P < 0.10$) were identified for inclusion in a final multivariate model. The potential covariates, which were also examined as potential effect modifiers, were maternal age, education, BMI (in kg/m$^2$), gestational age, and hemoglobin concentration at baseline; high-dose postpartum vitamin A supplementation; primiparity; household assets; housing and food insecurity indexes; season of sample collection; and milk fat concentration. The assets index is a composite index for baseline household ownership of the following set of assets: radio, television, refrigerator, cell phone, and stove. The housing index is a composite housing quality index based on the following baseline housing characteristics: drinking water supply, sanitation facilities, wall material, flooring material, roofing material, and lighting source. Household food insecurity access score is a continuous measure of the degree of food insecurity in the household, based on a set of questions that encompass 3 domains of food insecurity: *1*) anxiety and uncertainty about the household food supply, *2*) insufficient quality, and *3*) insufficient food intake and its physical consequences (38).

Mean ± SD adherence to study supplement (percentage of days the study supplement was reportedly consumed during the first 6 mo postpartum) was assessed and compared among the intervention groups by using ANOVA. The presence of an interaction between group assignment and high-dose postpartum vitamin A supplementation, parity, and other potential effect modifiers was assessed by using ANOVA; when there was a significant interaction ($P < 0.10$), ANCOVA was used to assess the differences between groups in stratified analyses.

Logistic regression was used to examine the effect of the intervention on low milk retinol concentration. The proportions of infants potentially

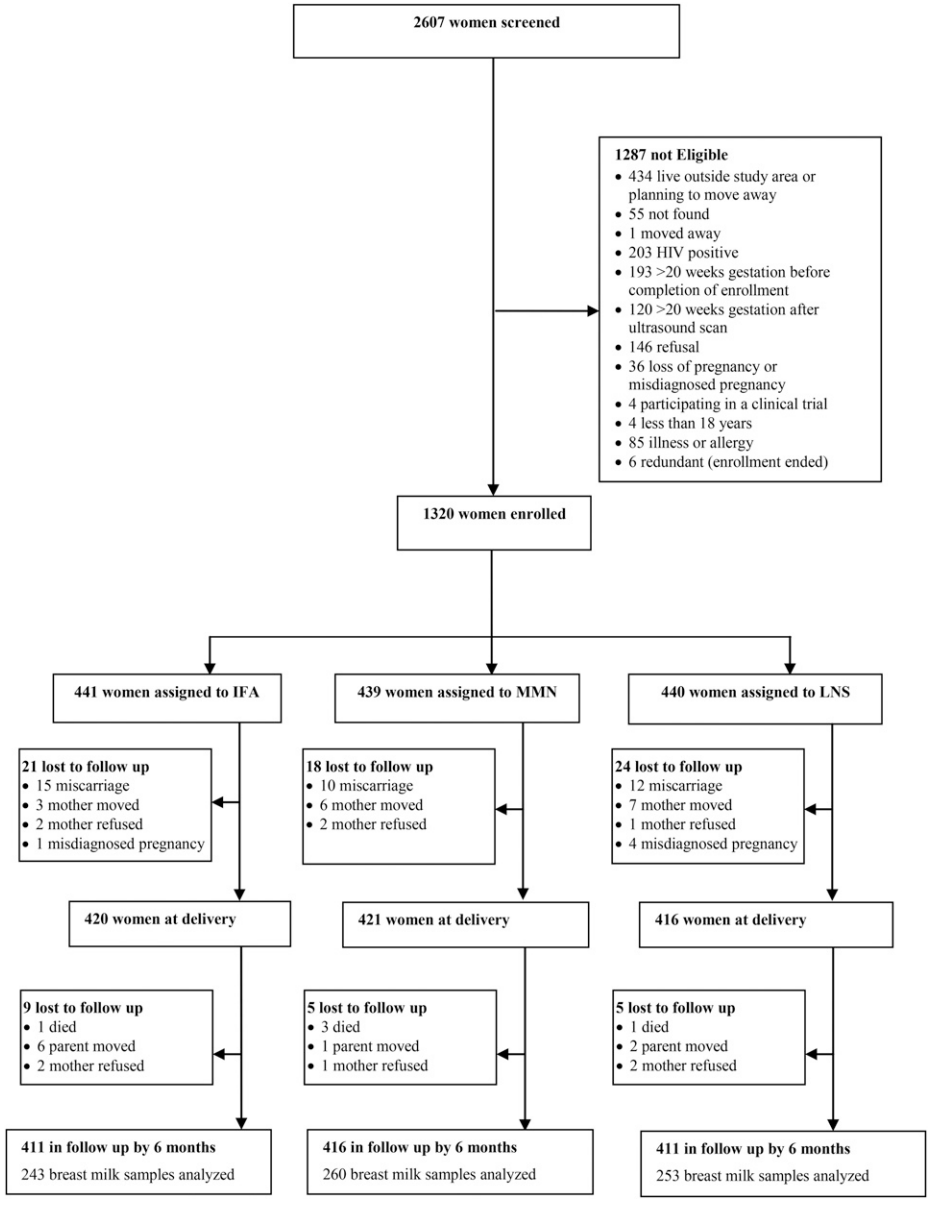


**FIGURE 1** Profile of Ghanaian women enrolled in and completing the study. IFA, iron and folic acid capsules; LNS, lipid-based nutrient supplement for pregnant and lactating women; MMN, multiple-micronutrient supplement.

**TABLE 1** Background characteristics of Ghanaian women who provided breast milk samples[1]

| Background characteristic | IFA ($n$ = 243) | MMN ($n$ = 260) | LNS ($n$ = 253) |
|---|---|---|---|
| At enrollment | | | |
| Age, y | 26.6 ± 5.5 | 26.8 ± 5.1 | 27.1 ± 5.5 |
| Formal education, y | 7.7 ± 3.6 | 7.4 ± 3.7 | 7.6 ± 3.8 |
| Gestational age, wk | 16.2 ± 3.4 | 16.2 ± 3.2 | 16.3 ± 3.3 |
| BMI, kg/m$^2$ | 24.7 ± 4.7 | 24.8 ± 4.4 | 25.2 ± 5.1 |
| Hemoglobin, g/dL | 11.0 ± 1.3 | 11.2 ± 1.2 | 11.1 ± 1.0 |
| Assets index[2] | −0.03 ± 1.13 | 0.02 ± 1.00 | −0.19 ± 1.04 |
| Housing index[3] | 0.06 ± 0.97 | −0.04 ± 1.07 | 0.04 ± 1.00 |
| Household food insecurity access score[4] | 2.8 ± 4.3 | 2.9 ± 4.6 | 2.8 ± 4.3 |
| Primiparous women, % | 31.3 | 31.5 | 30.8 |
| At delivery | | | |
| High-dose vitamin A postpartum, % | 58.9 | 57.1 | 58.6 |
| At 6 mo postpartum | | | |
| Adherence to study supplements,[5] % | 79.4 ± 20.4 | 77.6 ± 21.2 | 72.6 ± 25.7 |
| Days in last 7 d ate food made with red palm oil, $n$ | 3.3 ± 2.0 | 3.4 ± 2.0 | 3.3 ± 2.0 |
| Ate food made with red palm oil yesterday, % | 68.6 | 67.3 | 65.3 |
| Sample collection in dry season,[6] % | 60.3 | 62.3 | 60.9 |

[1] Values are means ± SDs or proportions (%). Analyses are based on ANOVA (SAS, PROC GLM) or $\chi^2$ tests (SAS PROC FREQ). IFA, iron and folic acid capsules; LNS, lipid-based nutrient supplement for pregnant and lactating women; MMN, multiple-micronutrient supplement.
[2] Assets index: composite index for baseline household ownership of a set of assets (radio, television, refrigerator, cell phone, and stove).
[3] Housing index: composite housing quality index based on baseline housing characteristics (drinking water supply, sanitation facilities, wall material, flooring material, roofing material, and lighting source).
[4] Household food insecurity access (HFIA) score: continuous measure of the degree of food insecurity in the household, based on a set of questions that encompass 3 domains of food insecurity: *1*) anxiety and uncertainty about the household food supply, *2*) insufficient quality, and *3*) insufficient food intake and its physical consequences (38). HFIA scores were on a scale from 0 to 27, with 0 being no food insecurity and 27 the most food insecurity.
[5] Percentage of days supplement was reportedly consumed during the first 6 mo postpartum.
[6] Milk samples collected between November and April.

receiving breast milk vitamin A at concentrations adequate to build their liver vitamin A stores, as well as the proportions of infants potentially receiving breast milk vitamin A above the UL or twice the UL, were compared among intervention groups by using Fisher's exact test. All $P$ values <0.05 were considered statistically significant.

## Results

**Figure 1** shows the flow of participants through the study. Women who were eligible at screening were contacted at home for their informed consent. Some women with baseline assessments were not enrolled in the study because they were >20 wk of gestation as confirmed by ultrasound scan. Women were asked to visit the laboratories at the study hospitals at 6 mo postpartum for their final laboratory assessments; some women did not complete the final laboratory visit ($n$ = 145). Milk samples were obtained from 1053 women who visited the study laboratories at 6 mo postpartum. Some of the milk samples collected were not analyzed because *1*) they were not available at the time of laboratory analysis ($n$ = 244), or *2*) the fat had separated from the milk and it was not possible to solubilize the milk ($n$ = 53).

The results presented are for participants whose breast milk samples were analyzed for vitamin A and fat concentrations. These women did not differ from those whose milk samples were not analyzed in terms of maternal age, education, gestational age, BMI, baseline hemoglobin, housing index, or in consumption of red palm oil (**Supplemental Table 1**), but they had a significantly lower assets index and higher household food insecurity access scores, and a lower proportion were primiparous.

The characteristics of the women in these analyses are presented by intervention group (**Table 1**). Age, years of formal education, and BMI were 26.8 ± 5.4 y, 7.5 ± 3.7 y, and 24.9 ± 4.7, respectively. Around 31% of the women were primiparous. A high proportion (67%) consumed foods made with red palm oil the day before the survey, and on average, women had consumed these foods 3.3 ± 2.0 d in the last 7 d. Frequency of consumption of red palm oil was very similar at 36 wk of gestation and at 6 mo postpartum (results not shown). The mean number of days of intake of other vitamin A–rich foods (colored roots and tubers, colored fruits, dark green leafy vegetables, organ meats, eggs) in the last 7 d was low (≤1.5 d for each of these food categories).

**TABLE 2** Breast milk retinol and fat concentrations of lactating Ghanaian women, by intervention group[1]

| Outcome | IFA ($n$ = 243) | MMN ($n$ = 260) | LNS ($n$ = 253) | $P$ value |
|---|---|---|---|---|
| Milk retinol, μmol/L | 2.5 ± 0.1 (0.5–6.2) | 2.5 ± 0.1 (0.6–9.4) | 2.5 ± 0.1 (0.5–9.3) | 0.88 |
| Milk fat, g/L | 41.9 ± 1.8 (12–186) | 45.2 ± 1.9 (11–153) | 44.7 ± 1.9 (11–236) | 0.42 |
| Milk retinol, nmol/g fat | 59.1 ± 2.8 (13–366) | 55.4 ± 2.5 (7–620) | 54.7 ± 2.5 (12–323) | 0.45 |

[1] Values are means ± SDs (ranges). Analyses are based on ANOVA (SAS, PROC GLM). IFA, iron and folic acid capsules; LNS, lipid-based nutrient supplement for pregnant and lactating women; MMN, multiple-micronutrient supplement.

**TABLE 3** Comparison of treatment effects on milk retinol per gram of fat in multiparous compared with primiparous lactating Ghanaian women[1]

| | Milk retinol, nmol/g fat | | | |
|---|---|---|---|---|
| Parity | IFA ($n$ = 243) | MMN ($n$ = 260) | LNS ($n$ = 253) | $P$ value |
| Multiparous | 56.4 ± 1.9 | 56.9 ± 1.9 | 55.4 ± 1.8 | 0.83 |
| Primiparous | 58.5 ± 2.9 | 56.1 ± 2.7 | 56.5 ± 2.8 | 0.82 |

[1] Values are means ± SEs. Adjusted analyses are based on ANCOVA (SAS, PROC GLM), controlling for milk fat concentration and season of sample collection. IFA, iron and folic acid capsules; LNS, lipid-based nutrient supplement for pregnant and lactating women; MMN, multiple-micronutrient supplement.

Adherence to the study supplement was 76.6% ± 22.7%; mean adherence was lower in the LNS group compared with the IFA and MMN groups ($P$ = 0.002 and 0.03, respectively) but did not differ between the IFA and MMN groups ($P$ = 0.63). Breast milk retinol, fat, and retinol per gram of fat concentrations were 2.5 ± 1.6 μmol/L, 44.0 ± 2.0 g/L, and 56.3 ± 2.1 nmol/g, respectively. There were no significant differences in any of these outcomes among intervention groups (**Table 2**), and results did not change after controlling for significant covariates (women's age and season of sample collection; data not shown) or in the sensitivity analyses taking into account the women who received both the MMN and IFA capsules during pregnancy.

Approximately 58% of women reported receiving high-dose vitamin A supplements in the immediate postpartum period. There was no significant interaction between intake of high-dose postpartum vitamin A supplements and intervention group with respect to breast milk vitamin A concentrations at 6 mo postpartum ($P$-interaction = 0.41). There was a significant interaction between parity and intervention group for milk retinol per gram of fat concentration ($P$-interaction = 0.09); however, in stratified analysis by parity status, no significant intervention group differences were seen in milk retinol concentration among multiparous or primiparous women, before or after adjusting for significant covariates (**Table 3**). There were no other interactions between BMI or any of the other potential effect modifiers and intervention group (data not shown).

Approximately 17% of participants had low milk retinol per gram of fat concentration (≤28 nmol/g), with no significant differences between groups (IFA: 16.1%, MMN: 18.5%, LNS: 16.6%; $P$ = 0.98). Approximately 56% of participants had adequate vitamin A (>46.7 nmol/g) to build infant liver vitamin A stores (33), with no group differences (**Table 4**).

Assuming an intake of 753 ± 137 mL breast milk/d (36), around 41% (23–59%) of exclusively breastfed infants would have had vitamin A intakes above the UL of 600 μg RAE/d for infants 0–6 mo of age (23), and approximately 3% would have had intakes above twice the UL, with no differences between groups (Table 4).

## Discussion

We found no significant effect of daily low-dose vitamin A provided in an LNS or MMN during pregnancy and the first 6 mo postpartum on breast milk retinol concentration at 6 mo postpartum. Less than one-fifth of the participants had low milk retinol per gram of fat. No significant interaction was observed between high-dose postpartum vitamin A supplementation and intervention group with respect to breast milk retinol concentration. Approximately 41% of exclusively breastfed infants in this study may have been ingesting >600 μg RAE/d from breast milk, at 6 mo of age, which exceeds the UL for vitamin A.

The lack of effect of daily maternal vitamin A supplementation on breast milk retinol concentration is not consistent with most previous studies. Daily or weekly supplementation of lactating women with vitamin A–rich plant or animal foods, β-carotene–fortified food, or β-carotene or vitamin A supplements resulted in increased breast milk retinol concentrations among Vietnamese (15), Indonesian (39, 40), and Bangladeshi (41) women. Daily supplementation with red palm oil maintained adequate milk retinol concentrations among Tanzanian lactating women compared with a control group (42). Regular intake of vitamin A–fortified sugar in Guatemala (43) and monosodium glutamate in Indonesia (44) among lactating women also resulted in increases in breast milk vitamin A concentrations. Studies in animals indicate that supplementation with vitamin A during pregnancy and lactation results in increased milk vitamin A concentrations (45, 46). Supplementation during pregnancy, particularly in the third trimester, can have a transient effect on milk retinol (47), but kinetic models suggest that supplementation during lactation results in a sustained increase in milk vitamin A concentration if the mother regularly ingests dietary or supplemental vitamin A (21). It has been shown in rats that supplementing dams during lactation resulted in a sustained increase in milk retinol and that pups born to dams that were supplemented during lactation had greater vitamin A stores than pups born to dams that were supplemented during pregnancy only (48).

It is possible that women enrolled in the current study had adequate intake of vitamin A from their usual diets and that this explains the lack of differences between groups. The frequent consumption of red palm oil, a rich source of β-carotene, supports

**TABLE 4** Estimated potential for maternal breast milk vitamin A to build liver stores of vitamin A in infants at 6 mo postpartum or exceed the UL[1]

| Breast milk vitamin A concentration | IFA ($n$ = 243) | MMN ($n$ = 260) | LNS ($n$ = 253) | $P$ value |
|---|---|---|---|---|
| Above level to build infant liver stores,[2] $n$ (%) | 144 (59.3) | 137 (52.7) | 139 (54.9) | 0.32 |
| Above the UL,[3] $n$ (%) | 96 (39.5) | 108 (41.5) | 104 (41.1) | 0.89 |
| Range[4] | 21.0–58.4 | 24.6–60.0 | 22.9–57.3 | |
| Above twice the UL,[3] $n$ (%) | 6 (2.5) | 8 (3.1) | 9 (3.6) | 0.81 |

[1] Breast milk vitamin A concentrations were measured in nmol/g fat. Analyses are based on Fisher's exact text (SAS, PROC FREQ). UL is defined as the highest level of daily nutrient intake that is likely to pose no risks of adverse health effects for almost all individuals in the general population (23); the UL for vitamin A is set at 600 μg RAE/d for infants 0–6 mo of age. IFA, iron and folic acid capsules; LNS, lipid-based nutrient supplement for pregnant and lactating women; MMN, multiple-micronutrient supplement; UL, Tolerable Upper Intake Level.
[2] Based on a cutoff of 46.7 nmol/g fat (1.75 μmol/L) breast milk vitamin A concentration (32).
[3] Based on a mean ± SD breast milk intake of 753 ± 137 mL/d for infants aged 6–8 mo, with breast milk retinol concentration >74.4 nmol/g fat (2.79 μmol/L) (36).
[4] Range of values for 1 SD below and above the mean breast milk intake for infants aged 6–8 mo (616 or 890 mL/d), with breast milk retinol concentration >91.2 or >62.9 nmol/g fat, respectively (36).

this possibility. The relatively low prevalence of low milk retinol per gram of fat concentration (17.1%) in this population is similar to that reported in an earlier trial in Ghana (13.2%) (12) and to the low prevalence of low serum retinol concentrations (15%) reported for pregnant Ghanaian women in a separate trial (49). These data support the hypothesis that vitamin A status may have been adequate for these women. Breast milk vitamin A concentrations >46.7 nmol/g fat are considered desirable to meet infant nutritional requirements and build vitamin A stores (32), whereas breast milk vitamin A concentrations of 28 nmol/g fat are considered adequate to meet infant requirements (32, 33) but not to build stores. On the basis of these values, we estimated that ~56% of the mothers had breast milk retinol concentrations that were sufficient to build the liver stores of their infants, and ~83% of the mothers had breast milk retinol concentrations that were sufficient to meet the daily vitamin A requirements of their breastfed infants.

We estimated that 41% of the mothers in this study had milk vitamin A concentrations that could potentially result in daily vitamin A intakes in their infants that exceed the UL (600 μg RAE/d), if the infants were exclusively breastfed and were consuming at least 753 mL milk/d (36). In our study, milk samples were collected from women when infants were 6 mo of age, and ~45% of infants were exclusively breastfed (with or without any prelacteal feeds) from 1 to 5 mo of age. The UL is one-tenth of the lowest observable adverse effect level (6000 μg RAE/d) for infants at 0–6 mo of age. Thus, it is unlikely that intakes of vitamin A from breast milk above the UL posed any risk of adverse clinical effects to infants in this study. Approximately 3% of mothers had a breast milk vitamin A concentration that would deliver twice the UL. Provision of daily doses of vitamin A in an LNS or MMN did not affect the proportion of women with breast milk vitamin A concentrations that could potentially result in infant intakes above the UL or twice the UL.

More than half (58%) of the women in this study reported receiving high-dose vitamin A supplements within 6 wk postpartum, but this was not associated with breast milk vitamin A concentration at 6 mo postpartum, and there was no significant interaction between intervention group and high-dose postpartum vitamin A supplementation. This is consistent with previous results from a multicenter trial that indicated that administration of 200,000 IU (60 mg) of vitamin A to women at 21–42 d postpartum increased breast milk vitamin A concentration and decreased the proportion of women with low milk retinol (≤28 nmol/g) at 2 mo postpartum, but the effect was not sustained at 6 and 9 mo postpartum (12). Other studies have also shown that high-dose postpartum vitamin A supplementation (200,000–400,000 IU) results in short-term increases in breast milk retinol concentration that do not last beyond 4 mo postpartum (11, 50, 51). This indicates that single mega-doses of vitamin A are insufficient to sustain adequate milk vitamin A concentrations throughout lactation. Smaller, daily doses of vitamin A or vitamin A–enriched diets (or both) that result in sustained chylomicron delivery of vitamin A to mammary tissue may be more effective in sustaining adequate milk retinol concentrations, as shown in lactating rats (18) and lactating sows (52).

The strengths of this study are the large number of women who participated and the rigorous follow-up of participants. The limitations include lack of blinding of participants who received the LNS, lack of data on baseline maternal vitamin A status, and the fact that the subsample of women who were included in this analysis had significantly lower household assets and greater food insecurity than the full cohort, which limits the generalizability of the results. We did not assess breast milk intake in our study population. Vitamin A intakes from breast milk were estimated based on the reported mean breast milk intake for infants 6–8 mo of age in developing countries and the measured vitamin A concentration of breast milk.

In conclusion, provision of daily doses of vitamin A in the LNS or MMN to this group of pregnant and breastfeeding Ghanaian women did not increase milk vitamin A concentration. Although the baseline vitamin A status of the women is not known, it is likely that most women had adequate dietary vitamin A intake. Despite the likelihood that the women were mostly vitamin A replete and that adherence to the supplement was high, daily maternal vitamin A supplementation did not increase breast milk vitamin A concentration or affect the proportion of infants with estimated vitamin A intakes from breast milk that exceeded the UL of 600 μg RAE/d.

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## References

1. Sommer A, Davidson FR. Assessment and control of vitamin A deficiency: the Annecy Accords. J Nutr 2002;132:2845S–50S.
2. WHO. Global prevalence of vitamin A deficiency in populations at risk 1995–2005. WHO Global Database on Vitamin A Deficiency. Geneva (Switzerland): WHO; 2009.
3. Clagett-Dame M, DeLuca HF. The role of vitamin A in mammalian reproduction and embryonic development. Annu Rev Nutr 2002;22:347–81.
4. Christian P, West KP Jr., Khatry SK, Kimbrough-Pradhan E, LeClerq SC, Katz J, Shrestha SR, Dali SM, Sommer A. Night blindness during pregnancy and subsequent mortality among women in Nepal: effects of vitamin A and β-carotene supplementation. Am J Epidemiol 2000;152:542–7.
5. Semba RD. The role of vitamin A and related retinoids in immune function. Nutr Rev 1998;56:S38–48.
6. Sklan D. Vitamin A in human nutrition. Prog Food Nutr Sci 1987;11:39–55.
7. van den Berg H. Vitamin A intake and status. Eur J Clin Nutr 1996;50:S7–12.
8. West KP Jr., Howard GR, Sommer A. Vitamin A and infection: public health implications. Annu Rev Nutr 1989;9:63–86.
9. WHO. Vitamin A supplementation in infants and children 6–59 months of age [Internet]. Geneva (Switzerland): WHO. 2011. [cited 2014 Apr 20]. Available from: http://apps.who.int/iris/bitstream/10665/44664/1/9789241501767_eng.pdf.
10. WHO. Vitamin A supplementation in postpartum women [Internet]. Geneva (Switzerland): WHO. 2011. [cited 2014 Apr 20]. Available from: http://www.who.int/nutrition/publications/micronutrients/guidelines/vas_postpartum/en/.

11. Ayah RA, Mwaniki DL, Magnussen P, Tedstone AE, Marshall T, Alusala D, Luoba A, Kaestel P, Michaelsen KF, Friis H. The effects of maternal and infant vitamin A supplementation on vitamin A status: a randomised trial in Kenya. Br J Nutr 2007;98:422–30.
12. Bahl R, Bhandari N, Wahed MA, Kumar GT, Bhan MK. Vitamin A supplementation of women postpartum and of their infants at immunization alters breast milk retinol and infant vitamin A status. J Nutr 2002;132:3243–8.
13. Basu S, Sengupta B, Paladhi PR. Single megadose vitamin A supplementation of Indian mothers and morbidity in breastfed young infants. Postgrad Med J 2003;79:397–402.
14. Ghana Statistical Service (GSS), Ghana Health Service (GHS), and ICF Macro. Ghana Demographic and Health Survey 2008 [Internet]. Accra (Ghana): GSS, GHS, and ICF Macro. 2008. [cited 2012 Aug 30]. Available from: http://dhsprogram.com/pubs/pdf/FR221/FR221[13Aug2012].pdf.
15. Khan NC, West CE, de Pee S, Bosch D, Phuong HD, Hulshof PJ, Khoi HH, Verhoef H, Hautvast JG. The contribution of plant foods to the vitamin A supply of lactating women in Vietnam: a randomized controlled trial. Am J Clin Nutr 2007;85:1112–20.
16. Villard L, Bates C. Effect of vitamin A supplementation on plasma and breast milk vitamin A levels in poorly nourished Gambian women. Hum Nutr Clin Nutr 1987;41:47–58.
17. Muhilal, Azis I, Saidin S, Jahari A, Karyadi D. Vitamin A–fortified monosodium glutamate and vitamin A status: a controlled field trial. Am J Clin Nutr 1988;48:1265–70.
18. Green MH, Green JB, Akohoue SA, Kelley SK. Vitamin A intake affects the contribution of chylomicrons vs. retinol-binding protein to milk vitamin A in lactating rats. J Nutr 2001;131:1279–82.
19. Ross AC, Gardner EM. The function of vitamin A in cellular growth and differentiation, and its roles during pregnancy and lactation. In: Allen L, King J, Lonnerdal B, editors. Nutrient regulation during pregnancy, lactation, and infant growth. New York: Plenum Press; 1994. p. 187–200.
20. Ross AC, Pasatiempo AM, Green MH. Chylomicron margination, lipolysis, and vitamin A uptake in the lactating rat mammary gland: implications for milk retinoid content. Exp Biol Med (Maywood) 2004;229:46–55.
21. Allen LH, Haskell M. Estimating the potential for vitamin A toxicity in women and young children. J Nutr 2002;132:2907S–19S.
22. Newman V. Vitamin A and breast-feeding: a comparison of data from developed and developing countries. Food Nutr Bull 1994;15:161–76.
23. National Research Council. Dietary reference intakes for vitamin A, vitamin K, arsenic, boron, chromium, copper, iodine, iron, manganese, molybdenum, nickel, silicon, vanadium, and zinc. Washington (DC): National Academies Press; 2001.
24. FAO/WHO. Human vitamin and mineral requirements: report of a joint FAO/WHO expert consultation. Rome: WHO/FAO; 2001.
25. Arimond M, Zeilani M, Jungjohann S, Brown KH, Ashorn P, Allen LH, Dewey KG. Considerations in developing lipid-based nutrient supplements for prevention of undernutrition: experience from the International Lipid-Based Nutrient Supplements (iLiNS) Project. Matern Child Nutr 2013 May 6 (Epub ahead of print; DOI: 10.1111/mcn.12049).
26. Adu-Afarwuah S, Lartey A, Mamane Zeilani M, Dewey KG. Acceptability of lipid-based nutrient supplements (LNS) among Ghanaian infants and pregnant or lactating women. Matern Child Nutr 2011;7:344–56.
27. Adu-Afarwuah S, Lartey A, Okronipa H, Ashorn P, Zeilani M, Peerson JM, Arimond M, Vosti S, Dewey KG. Lipid-based nutrient supplement increases the birth size of infants of primiparous women in Ghana. Am J Clin Nutr 2015;101:835–46.
28. UNICEF/WHO/UNU. Composition of a multi-micronutrient supplement to be used in pilot programmes among pregnant women in developing countries: report of a United Nations Children's Fund (UNICEF), World Health Organization (WHO) and United Nations University workshop [Internet]. New York: UNICEF. 1999. [cited 2015 Jan 18]. Available from: http://apps.who.int/iris/bitstream/10665/75358/1/UNICEF-WHO-multi-micronutrients.pdf?ua=1.
29. Kaestel P, Michaelsen KF, Aaby P, Friis H. Effects of prenatal multimicronutrient supplements on birth weight and perinatal mortality: a randomised, controlled trial in Guinea-Bissau. Eur J Clin Nutr 2005;59:1081–9.
30. Tanumihardjo SA, Penniston KL. Simplified methodology to determine breast milk retinol concentrations. J Lipid Res 2002;43:350–5.
31. Handelman GJ, Shen B, Krinsky NI. High resolution analysis of carotenoids in human plasma by high-performance liquid chromatography. Methods Enzymol 1992;213:336–46.
32. WHO. Indicators for assessing vitamin A deficiency and their application in monitoring and evaluating intervention programmes (WHO/NUT/96.10). Geneva (Switzerland): WHO; 1996.
33. Underwood BA. Maternal vitamin A status and its importance in infancy and early childhood. Am J Clin Nutr 1994;59:517S–22S.
34. Rice AL, Stoltzfus RJ, de Francisco A, Kjolhede CL. Evaluation of serum retinol, the modified-relative-dose-response ratio, and breast-milk vitamin A as indicators of response to postpartum maternal vitamin A supplementation. Am J Clin Nutr 2000;71:799–806.
35. WHO. Report of the expert consultation on the optimal duration of exclusive breastfeeding. Geneva (Switzerland): WHO; 2001.
36. WHO/UNICEF/ORSTOM/UC Davis. Complementary feeding of young children in developing countries: a review of current scientific knowledge. WHO/NUT/98.1. Geneva (Switzerland): WHO; 1998.
37. Lucas A, Gibbs JAH, Lyster RLJ, Baum JD. Creamatocrit: simple clinical technique for estimating fat concentration and energy value of human milk. BMJ 1978;1:1018–20.
38. Coates J, Swindale A, Paula Bilinsky P. Household Food Insecurity Access Scale (HFIAS) for measurement of food access: indicator guide. Washington (DC): Academy for Educational Development, Food and Nutrition Technical Assistance Project; 2007.
39. de Pee S, West CE. Muhilal, Karyadi D, Hautvast JG. Lack of improvement in vitamin A status with increased consumption of dark green leafy vegetables. Lancet 1995;346:75–81.
40. Muslimatun S, Schmidt MK, West CE, Schultink W, Hautvast JG, Karyadi D. Weekly vitamin A and iron supplementation during pregnancy increases vitamin A concentration of breast milk but not iron status in Indonesian lactating women. J Nutr 2001;131:2664–9.
41. Rice AL, Stoltzfus RJ, de Francisco A, Chakraborty J, Kjolhede CL, Wahed MA. Maternal vitamin A or beta-carotene supplementation in lactating Bangladeshi women benefits mothers and infants but does not prevent subclinical deficiency. J Nutr 1999;129:356–65.
42. Lietz G, Henry CJ, Mulokozi G, Mugyabuso JK, Ballart A, Ndossi GD, Lorri W, Tomkins A. Comparison of the effects of supplemental red palm oil and sunflower oil on maternal vitamin A status. Am J Clin Nutr 2001;74:501–9.
43. Arroyave G, Beghin I, Flores M, Soto de Guido CS, Ticas JM. Effects of consuming sugar fortified with retinol on pregnant and breastfeeding women whose normal diet is low in vitamin A: study of the mother and child. Arch Latinoam Nutr 1974;24:485–512.
44. Muhilal, Azis I, Saidin S, Jahari AB, Karyadi D. Vitamin A–fortified monosodium glutamate and vitamin A status: a controlled field trial. Am J Clin Nutr 1988;48:1265–70.
45. Davila ME, Norris L, Cleary MP, Ross AC. Vitamin A during lactation: relationship of maternal diet to milk vitamin A content and to the vitamin A status of lactating rats and their pups. J Nutr 1985;115:1033–41.
46. Green MH, Snyder RW, Akohoue SA, Green JB. Increased mammary tissue vitamin A levels associated with increased vitamin A intake during lactation are maintained after lactation in female rats. J Nutr 2001;131:1544–7.
47. Ortega RM, Andress P, Martinez RM, Sobalar AM. Vitamin A status during the third trimester of pregnancy in Spanish women: influence on concentrations of vitamin A in breast milk. Am J Clin Nutr 1997;66:564–8.
48. Akohoue SA, Green JB, Green MH. Dietary vitamin A has both chronic and acute effects on vitamin A indices in lactating rats and their offspring. J Nutr 2006;136:128–32.
49. Kirkwood BR, Hurt L, Amenga-Etego S, Tawiah C, Zandoh C, Danso S, Hurt C, Edmond K, Hill Z, Ten Asbroek G, Fenty J, Owusu-Agyei S, Campbell O, Arthur P; ObaapaVitA Trial Team. Effect of vitamin A supplementation in women of reproductive age on maternal survival in Ghana (ObaapaVitA): a cluster-randomised, placebo-controlled trial. Lancet 2010;375:1640–9.
50. Oliveira-Menegozzo JM, Bergamaschi DP, Middleton P, East CE. Vitamin A supplementation for postpartum women. Cochrane Database Syst Rev 2010;123:CD005944.
51. Stoltzfus RJ, Hakimi M, Miller KW, Rasmussen KM, Dawiesah S, Habicht JP, Dibley MJ. High dose vitamin A supplementation of breastfeeding Indonesian mothers: effects on the vitamin A status of mother and infant. J Nutr 1993;123:666–75.
52. Penniston KL, Valentine AR, Tanumihardjo SA. A theoretical increase in infants' hepatic vitamin A is realized using a supplemented lactating sow model. J Nutr 2003;133:1139–42.