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Micronutrient regulation in pregnant and lactating women from Rio de Janeiro

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SUMMARY. **Micronutrient regulation in pregnant and lactating women from Rio de Janeiro.** Studies with low-income pregnant and lactating women from the city of Rio de Janeiro, concerned mainly with the changes in micronutrient homeostasis during pregnancy and lactation in the absence of overt clinical deficiencies, are reported. These studies focused on folate, cobalamin, iron, zinc and vitamin A. Factors that may affect the maternal micronutrient state, such as dietary intakes, use of supplements and interrelationships of micronutrients have been considered, as well as the implications of these changes for maternal-fetal transfer and milk composition. Although these studies were not designed to evaluate the prevalence of sub-clinical micronutrient deficiencies in pregnant and lactating women, they indicate that high frequencies of sub-clinical deficiencies of folate, iron, zinc and vitamin A, especially in pregnant women, are expected to be found in Rio de Janeiro.

INTRODUCTION

Pregnancy and lactation impose a high demand of nutrients for growth of maternal, placental and fetal tissues, and for milk production. This demand should be met by maternal body stores and increased intake of essential nutrients. Physiological and hormonal changes in these periods affect nutrient metabolism and regulation of homeostasis in order to attain an efficient nutrient utilization by the maternal organism and transfer to fetus and to milk.Iron and folate are good examples of nutrients which highly increased requirements during pregnancy are difficult to achieve through the habitual diet, especially for low income women in developing countries who may be at risk for the development of both nutrient deficiencies. As a consequence, iron and folate deficiencies are the most prevalent nutritional disorders during pregnancy (1).

It is estimated that 90% of all anemias in the world are due to iron deficiency. Worldwide, 60% of pregnant women are anemic and in developed countries the prevalence of anemia oscillates between 9 and 14% (2). In Brazil, data on prevalence of nutritional anemias in pregnant women are limited to some regions and the few studies available are concerned with iron-deficiency anemia, showing high intra and inter-regional differences (3). Prevalences of iron-deficiency anemia in pregnant women vary from 14% in São Paulo to 35% in Pernambuco (4). Data on sub-clinical iron deficiency during pregnancy and lactation are scarcely available, but limited studies in Rio de Janeiro (5,6) have shown that 40% of pregnant and 21% of lactating women have depleted iron reserves.

Nutritional folate deficiency has a worldwide distribution, differing greatly in severity, with the sub-clinical, non-anemic forms being predominant in pregnant women. Megaloblastic anemia due to dietary folate deficiency affects up to 25% of non-supplemented pregnancies in certain parts of Asia, Africa and Latin America, and 2.5-5.0 % in developed countries. It is estimated that sub-clinical folate deficiency has even higher prevalences, affecting up to one-third of the pregnant women on a global scale (1). In Brazil, folate has received much less attention than iron and only very few studies have been carried out concerning folate status, in general, and in pregnancy and lactation in particular. In this country, megaloblastic anemia is not considered common during pregnancy, but limited studies in the urban regions of Rio de Janeiro (6) and Manaus (7) have shown that 20% and 22% of non-supplemented pregnant women, respectively, have sub-clinical folate deficiency.

The undesirable effects of overt, clinical maternal deficiency of these micronutrients on pregnancy development and outcome, and on infant status and milk composition are generally well-known. However, as mentioned previously, sub-clinical deficiencies are more prevalent, this being probably especially true in urban regions and metropolises. Moreover, their effects are poorly evaluated and understood.

The objective of this paper, in the context of the present workshop, is to review the main results obtained in studies carried out by our research group with low-income pregnant and lactating women, from the city of Rio de Janeiro. These studies have been concerned mainly with the changes in micronutrient homeostasis during pregnancy and lactation, in women without clinical micronutrient deficiencies. The implications of these changes for fetal nutritional status, maternal-fetal transfer and milk composition have been considered, as well as factors that may affect these changes, such as maternal sub-optimal states, dietary intakes and use of supplements. We have focused our interest in the following micronutrients: folate, cobalamin, iron, zinc, and more recently vitamin A.

CHANGES IN MICRONUTRIENT STATUS OF LOW-INCOME WOMEN DURING PREGNANCY AND LACTATION

Pregnancy is usually associated with iron and folate depletion, especially in women of low socio-economic level, multiparas, and with low dietary intakes (1), but information regarding Brazilian women is scarce. Because of the risks of the negative effects on the mother, conceptus and infants caused by iron and folate deficiencies, routine supplementation with both nutrients throughout pregnancy has been recommended (1,2). In the case of folate, supplementation of all women in the periconceptional period is recommended in some developed countries to decrease the risk of first occurrence and recurrence of neural tube defects of the fetus (8). Despite the known beneficial effects of iron and folate supplementation of deficient women during pregnancy, its indiscriminate use as a routine procedure for all pregnant women is controversial regarding its potential benefits and possible risks, such as those resulting from excessive nutrient intake and/or adverse nutrient-nutrient interactions (9).

Changes in folate and iron status during pregnancy and lactation (2-3 months post partum) were evaluated in a study with a cohort of non-anemic pregnant women attending a public prenatal clinic in Rio de Janeiro (6). We also assessed whether the indiscriminate supplementation with iron (80-160 mg/d) and with combined folate (2-4 mg/d) and iron supplementation (80-160 mg/d), as routinely prescribed during pregnancy, brings a substantial benefit in terms of pregnancy outcome and of nutrient status to women who begin pregnancy with adequate iron and folate reserves, and do not become anemic during pregnancy. Maternal weight gain, absence of serious complications in pregnancy, delivery and post partum, and the infants' birth weight and length showed that pregnancy proceeded normally and adequately, independently of the use of supplements. Hemoglobin and hematocrit remained adequate throughout these periods and did not further improve with the use of supplements.

Mean dietary intakes of iron and folate during pregnancy were, respectively, 50% and 83% of the recommended intakes for pregnant women (10). Despite this, mean plasma and erythrocyte folate levels were adequate and did not change during pregnancy and lactation in unsupplemented women (6). It would be interesting to ascertain whether their folate catabolism and urinary excretion are lower than the reported high values for well-nourished pregnant women (11). Folate indices were further improved in both periods with the use of folate supplements, but were not affected by exclusive iron supplementation. Plasma cobalamin levels were adequate throughout these periods and correlated with plasma folate (6).

Iron stores of most of the women were adequate at the beginning of pregnancy and were able to prevent iron-deficiency anemia during gestation. Although mean plasma ferritin levels were adequate throughout the study, they decreased during pregnancy, regardless the use of iron supplements (6), as has also been shown by studies with Scandinavian women (12), with the frequency of depleted iron reserves (plasma ferritin < 12 mg/l) increasing to 40% in the third trimester. However, iron supplementation was effective in maintaining adequate circulating plasma iron during pregnancy and was beneficial for the recovery of iron stores in the lactation period (6).

Since plasma iron represents the iron in transit from liver stores and intestine to the highly demanding hematopoietic, placental and fetal tissues during pregnancy, the extra iron provided by the supplements during pregnancy seems to be preferentially directed to these sites rather than to the stores. The fact that oral iron supplementation of non-anemic women during pregnancy was not effective in avoiding a decrease in plasma ferritin (6), whereas supplementation during 3 months of lactation, a period of lower iron demands, was effective in increasing iron stores (13), suggests that iron mobilization from stores during pregnancy may be regulated by a specific mechanism that favors intense mobilization independently of intestinal iron absorption.

INTER-RELATIONSHIP OF MICRONUTRIENTS DURING PREGNANCY AND LACTATION

Evidences provided by several studies in humans and animals suggest that iron deficiency can affect folate utilization and that a complex relationship between both nutrients occurs during reproduction (14).

A relation between iron stores and folate status in non-anemic women from Rio de Janeiro was found in several studies (6,15,16). There was a correlation between plasma ferritin and erythrocyte folate in the second and third trimesters of pregnancy, but not in the first trimester or during lactation. The women with iron depleted reserves (serum ferritin < 12µg/l) in the third trimester, either supplemented or not with folate, presented lower erythrocyte folate levels than the non-depleted women in this period, in spite of a similar dietary folate intake. Also, erythrocyte folate levels in the women with depleted iron reserves tended to be less responsive to folate supplementation and, in the case of no folate supplementation, showed a decrease in comparison with the beginning of pregnancy (6).

In other groups of volunteers without depleted iron reserves, the women with ferritin levels in the lower quartile at parturition (15) and 1-2 days post partum (16) had erythrocyte folate levels lower than those with ferritin in the higher quartile, possibly reflecting the situation during pregnancy. Again, this was not evident at 3 months post partum (16).

These results suggest that a relation between erythrocyte folate and iron stores occurs in periods of high simultaneous demand for both nutrients and increased erythropoiesis, such as pregnancy.

MATERNAL-FETAL TRANSFER OF MICRONUTRIENTS

Maternal-fetal transfer of folate, cobalamin, iron and zinc is very efficient and competes favorably with maternal tissues, assuring an adequate fetal status even in maternal deficiency. Umbilical cord and newborn serum levels of these nutrients are usually higher than in maternal serum. The high rates of placental transfer from mother to fetus and preferential uptake of the first three nutrients by the placenta are mainly explained by the higher expression of folate, transcobalamin and transferrin receptors in the microvilli (maternal surface) of the placental syncitiothrophoblast (17-19). For zinc, the mechanism for placental transfer is not well known, and involves more than one carrier system in the microvilli and intracellular zinc ligands, such as metallothionein (20).

In the case of vitamin A, the placenta ensures an adequate supply to the fetus despite a wide variation in the maternal vitamin A circulating levels, except when there is a maternal clinical deficiency of vitamin A, and probably also when an excess intake occurs (21). However, the umbilical cord serum concentrations of retinol and carotenoids are usually lower than in the maternal serum (22,23). The mechanism and regulation of transplacental flux of vitamin A are still poorly understood. Retinol uptake in placental microvillous membrane is possibly regulated by a specific receptor for the serum retinol-binding protein, which would control the accumulation of retinol in the placenta and its transfer to the fetus (24).

A main concern regarding the maternal-fetal transfer of micronutrients to the fetus is the extent to which the process is regulated by the nutritional status of the mother. The relation of iron, folate and cobalamin states of the newborn with maternal circulating levels and reserves, which varied within an adequate range, was investigated in a study with 24 pregnant women at parturition (15). Cord levels of serum and erythrocyte folate, serum cobalamin, iron, transferrin saturation and ferritin, as well as hemoglobin and hematocrit, were higher than in maternal blood, as expected. Cord serum folate and cobalamin were correlated with maternal levels, whereas iron status of the newborn did not depend on the maternal circulating iron or iron reserves. Folate and cobalamin states of the newborn were not related to maternal iron status. In a study with pregnant women at parturition (25), grouped according to their serum zinc levels (low < 7.6 ; intermediate 7.6-10.7, high zinc > 10.7 µmol/l), cord serum zinc levels were higher than the maternal ones, adequate in all groups, and slightly higher in the high zinc group.

There was a correlation between maternal and cord serum zinc. Erythrocyte zinc and metallothionein in blood cord were similar for all groups, and the low zinc levels in maternal serum did not affect zinc and metallothionein levels in placenta.

In vitro models using microvillous membrane vesicles from human placental syncitiothrophoblast have been used in our laboratory to further explore the transport of folate, cobalamin and zinc at the membrane level in various conditions (26-30). These experiments showed that neither folate or iron deficiency affected biochemical parameters of folate transport (27). Folate receptor affinities for folic acid (Kd= 0.5-1.8 nmol/l) in placentas from deficient mothers were similar to that of the controls and would be appropriate for an efficient folate uptake, even in the presence of a maternal folate deficiency, when concentration of serum folate is lower than 6.8 nmol/l. In the case of placentas from women with low serum zinc levels, also no difference was observed in zinc transport from those with normal levels (28). Conditions, such as gestational period, severe maternal hypertension and fetal neural tube defects, in which structural and morphological characteristics of the placenta are different from the normal placenta, do not impair folate transport (29). However, in premature placentas, the transport of cobalamin and zinc was more efficient. Cobalamin transport was twice higher (30) and there was an increase in concentration of zinc-binding sites, with no effect on affinity (28).

A sub-clinical vitamin A deficiency of the mothers may have no effect on fetal circulating retinol, but may affect the placental handling of retinol and carotenoids to ensure an adequate retinol supply to the fetus. The placental transfer of retinol and β-carotene was investigated in pregnant women with adequate (serum retinol > 20 µg/dl) and sub-adequate (serum retinol ≤ 20 µg/dl) vitamin A status (22). Although there was no difference in retinol and β-carotene levels in placenta and cord serum between these groups, and no correlation between maternal serum retinol and placental or cord retinol, a significant correlation was found between maternal serum β-carotene and placental retinol in mothers with sub-adequate vitamin A status, particularly in those with retinol levels lower than 15µg/dl, whom also presented a significant correlation between maternal β-carotene and cord serum retinol. These results suggest that β-carotene may be a precursor of retinol in placenta, which would then be available to the fetus, and that it could be dependent on the retinol status of the mother. Also, the decrease of β-carotene in relation to α-carotene and licopene in the placenta, when compared to maternal serum (31), is suggestive of β-carotene catabolism in placenta.

MATERNAL MICRONUTRIENT STATUS AND MILK COMPOSITION

Mean levels and range of folate, cobalamin, iron and zinc in milk from low-income women at different periods of lactation (5) were similar to those found for well-nourished mothers in developed countries (32,33).

There was no relationship between maternal indices of iron, zinc, folate and cobalamin status and the milk concentrations of these nutrients, in spite of the risk of poor status observed in some of the mothers. Milk components related to a better availability of these nutrients to the infants, such as cobalamin-binding protein, folate-binding protein and lactoferrin, or whey bound zinc and iron, were not affected by maternal status. Partial weaning (pasteurized cow's milk, cooked vegetables and bananas substituting at least two daily feedings from the breast) did not affect milk composition, except for total iron which was lower in milk from mothers who were exclusively breast feeding. Folate and iron supplementation during pregnancy did not affect milk composition (5).

Although the means of folate, cobalamin and iron indices in the infants with ages varying from 31-280 d were within the normal limits, regardless the type of feeding, a substantial proportion of exclusively breast-fed and partially weaned infants presented low levels of serum folate (33 and 67%, respectively) and hemoglobin (33 and 43%, respectively) (34).

In another study (13), iron supplementation (40 mg/d) of lactating women for three months after delivery did not affect iron and zinc levels, but increased total iron ligands and lactoferrin/protein ratio in milk, which could be a response of the mammary gland to the higher plasma transferrin saturation found in these mothers in comparison to the non-supplemented ones.

SUB-ADEQUATE MICRONUTRIENT STATUS IN PREGNANT AND LACTATING WOMEN

Although the afore-mentioned studies were not designed to evaluate the prevalence of sub-clinical micronutrient deficiencies and are not representative of low-income pregnant and lactating women from Rio de Janeiro, they have drawn attention to micronutrient inadequacies in these groups. They provide some evidence regarding high frequencies of sub-clinical deficiencies of folate, iron, zinc and vitamin A and the striking beneficial effect of folate supplementation during pregnancy.

In one study with non-anemic women (6), 22% started pregnancy with low erythrocyte folate levels (<360 nmol/l; 35) and this proportion was also found in the third trimester of pregnancy and during lactation (2-3 months) only in those who did not use folate supplements. Regarding iron status, eight percent of the women started pregnancy with depleted iron stores (plasma ferritin levels <12 µg/l; 36), whereas in the third trimester this frequency had increased to about 40%, irrespective of iron supplementation. However, in the post partum, 40% of the women who did not take iron supplements during pregnancy had low iron reserves as compared to 18% in the group using iron supplements. None of the women presented low levels of plasma cobalamin (<110 pmol/l; 37).

Other studies showed that 25% of the pregnant women at parturition (25), and 11% after 3 months of lactation (5) had low serum zinc levels (< 7.6 and 9.9 µmol/l, respectively; 38). Low serum retinol levels (<0.7 µmol/l, 36) were found in 35% of pregnant women at parturition (22).

Dietary intakes of folate, iron, zinc and calcium were assessed in pregnant and lactating women (6,39) by food frequency questionnaires. Mean dietary intakes of these nutrients were 83, 50, 73 and 53%, respectively, of the recommended intakes for pregnant women (10). In this period, 80, 100, 82 and 95% of the women had lower dietary intakes, respectively, than the recommendations.

During lactation, mean dietary intakes of these nutrients were similar to intakes during pregnancy, but adequacy of dietary intakes of folate and iron improved, since recommendations for this period are lower, with mean intakes of 120 and 100%, respectively, of the recommended intakes. As for zinc and calcium, mean dietary intakes were 64 and 57% of recommendations in this period, since recommended intakes for zinc are higher and for calcium are the same in comparison of recommendations for pregnant women. In the lactation period, 35, 52, 93 and 90% of the women, respectively, had folate, iron, zinc and calcium intakes lower than the recommendations.

In both periods, mean cobalamin dietary intakes were higher and none of the women had cobalamin intakes lower than the recommendations.

CONCLUSIONS

Representative data on prevalence of sub-clinical and clinical deficiencies of micronutrients are generally lacking in different population groups from Rio de Janeiro, particularly in pregnant and lactating women. Although the studies reported in this paper were not designed to assess prevalences of micronutrient deficiencies in pregnant and lactating women from Rio de Janeiro, they indicate that high frequencies of sub-clinical deficiencies of folate, iron, zinc and vitamin A, especially in pregnant women, are to be expected.

Sub-adequate micronutrient status may not have an important impact on pregnancy development and outcome, or on milk composition, but may be involved in problems that have not been specifically addressed, such as chronic and degenerative diseases.

For instance, in the case of folate deficiency, besides its manifestation as the classical clinical deficiency, megaloblastic anemia, subclinical deficiency has been associated with increased risk for cardiovascular diseases, due to hyperhomocysteinemia, and various types of cancer. The possible impact of transient and chronic sub-clinical folate deficiencies during pregnancy and lactation on folate homeostasis and on women's health, may be related to a future onset of these diseases. Serum homocysteine is a sensitive indicator of intracellular folate and it is increased when depletion of this vitamin occur. Evaluation of homocysteine levels in pregnant and lactating women is lacking, as well as studies concerned with the extent to which a marginal folate deficiency in these periods contributes to hyperhomocysteinemia. Validated biochemical and functional tests to evaluate micronutrient status specifically in pregnancy and lactation are needed, since at present this evaluation relies on poor indicators of status for some nutrients such as zinc, and disputable cut off values for most nutrients.

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