

# Influence of breastfeeding type and maternal anemia on hemoglobin concentration in 6-month-old infants

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## Abstract

**Objective:** To verify the influence of breastfeeding type and of maternal anemia on hemoglobin concentration in 6-month-old infants.

**Methods:** This was a cross-sectional study nested in a community-based, randomized and controlled intervention study that aimed to prolong the duration of exclusive breastfeeding during the first 6 months of life. This study was conducted in four towns in the Brazilian state of Pernambuco and newborn infants were recruited from March to August of 2001. The hemoglobin concentrations of 330 mothers and infants were assayed and type of breastfeeding was assessed 6 months after delivery. Multivariate linear regression analysis was used to identify factors that independently contributed to the infants' hemoglobin concentration.

**Results:** The type of feeding had no influence on the hemoglobin concentration in the sample as a whole, however, there was a significant difference when the "exclusive + predominant breastfeeding" subset of infants was analyzed, with the children of anemic mothers exhibiting a reduction of 0.7g/dL in median hemoglobin. Mothers' hemoglobin level, type of flooring at home, type of delivery, and birthweight all significantly contributed to the variation in the infants' hemoglobin concentration.

**Conclusions:** In contrast with type of breastfeeding, maternal anemia did have an influence on the hemoglobin levels of 6-month-old infants, even when only children on "exclusive + predominant breastfeeding" were analyzed. These findings highlight the need to prevent maternal anemia before conception, during pregnancy and throughout lactation.

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## Introduction

Nutrition is the most important factor in child health promotion, growth and development; especially during the first 2 years of life, when the speed of neuropsychomotor growth and development is greatest.<sup>1</sup> The health and nutrition of mothers and their children are intimately related. The effects of nutrition begin even before conception, promoting intrauterine growth and development, physical growth and mental development, with repercussions for adulthood.<sup>2</sup>

Iron deficiency anemia is the most prevalent type of food deprivation in the world and it particularly affects expectant mothers and infants in developing countries.<sup>2</sup> Several factors can contribute to iron deficiency in infants. Of note during the first year of life are conditions linked to pregnancy (maternal anemia) and delivery (premature ligation of the umbilical cord) and their effects on the child's iron reserves, in addition to the increased need for iron from 6 to 12 months, due to the accelerated velocity of neuropsychomotor growth and

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development, which exhausts reserves during this period if they are not adequately replaced.<sup>3</sup>

Children born full term have good iron reserves in the liver and hematopoietic tissues as a result of breaking down fetal red blood cells soon after birth, which deposits iron in these locations, especially if umbilical cord ligature is delayed until it has stopped pulsing.<sup>4</sup> There is evidence that the transfer of iron to the fetus increases when the mother has reduced reserves. However, since the mother then becomes anemic, this transfer reduces, and the fetus becomes at risk of deficiency.<sup>5</sup> There is evidence that even children with normal birth weights, but anemic mothers, will have low iron reserves at birth and are more likely to develop anemia.<sup>6</sup>

There is a lack of data in the literature on the prevalence of anemia among children less than 6 months old, because of the assumption that at this age breastmilk suffices to provide the necessary iron, irrespective of maternal nutritional status. Only in extreme cases of deprivation would maternal deficiency affect the child.<sup>7</sup>

There is consensus that there is no substitute for exclusive breastfeeding during the first months of a child's life; however, the duration of breastfeeding continues to be the subject of debate. In a recent review specialists concluded that the available evidence is sufficient to recommend exclusive breastfeeding for the first 6 months of life.<sup>8</sup> Although the iron concentration of breastmilk is low, it has very good bioavailability ( $\pm$  50% absorption), and breastmilk alone is enough to keep iron levels ideally balanced in children born full term for at least the first 6 months of life.<sup>9</sup>

An intervention study aimed to promote breastfeeding conducted in the Zona da Mata Meridional in Pernambuco state achieved a significant increase in the percentage of children still on exclusive breastfeeding at 6 months, with an aggregated prevalence of 45% in the intervention group vs. 13% in the control group.<sup>10</sup> This cohort offered the possibility of investigating the influence of the type of breastfeeding and of maternal anemia on the hemoglobin levels of 6-month-old infants.

## Methods

The study was carried out in the urban areas of the towns of Palmares, Agua Preta, Catende, and Joaquim Nabuco, all located in the Zona da Mata Meridional in Pernambuco, Northeast Brazil, and with a population of approximately 135,000 inhabitants. The region's economy is based on sugar cane production, approximately 2/3 of the economically active population report an income of less than twice the minimum monthly wage and illiteracy among women is around 26%.

This was a cross-sectional study nested in a community-based, randomized and controlled intervention study that

aimed to promote and prolong the duration of exclusive breastfeeding by means of home visits from birth to 6 months.

From March to August of 2001, 350 mother-baby pairs were recruited at maternity units in the area and allocated at random into one of the two intervention study groups. All singleton children born full term were eligible, except if they had congenital anomalies, needed intensive care or if their mothers had severe morbidities or mental disease or intended to leave the study area within 6 months.<sup>10</sup>

Twenty children were lost to follow-up during the 6-month period, which is equivalent to 5.7% of the sample. The characteristics of these children did not differ from those who remained on the study, with 330 mother-child pairs completing the study. Considering that just 25% of this population of children were on exclusive/predominant breastfeeding at 6 months of age, a minimum sample size of 60 children+mothers was estimated (20 children of anemic mothers and 40 children of mothers without anemia) to be necessary in order to detect an expected difference in median hemoglobin of 0.8 g/dL, adopting a significance level of 5%, study power of 80% and an allocation ratio of 1:2, according to G\*Power version 3.1 software.

At 6 months of age, all children and mothers had their hemoglobin concentration in capillary blood assayed with HemoCue, during home visits made by two research assistants who had been appropriately trained in advance. Children were defined as anemic if their hemoglobin level was below 11 g/dL, with severe anemia being  $<$  7.0 g/dL; moderate from 7.0 to 8.9 g/dL, and mild from 9.0 to 10.9 g/dL. Mothers were defined as anemic if they had a hemoglobin level of  $<$  12 g/dL.<sup>11</sup>

Data were collected at the maternity unit using a pre-coded structured questionnaire including questions on family income, living conditions (type of flooring, water supply and WC), family size, literacy, and mother's age, parity and prenatal care. Six months after delivery, the mothers were interviewed during a home visit by four research assistants, using a structured questionnaire that covered breastfeeding, and feeding with water, tea, other liquids, other milks or foods during the 24 hours prior to the interview, in addition to information on anemia during pregnancy.

Types of breastfeeding were defined in accordance with the World Health Organization (WHO),<sup>12</sup> with exclusive breastfeeding defined as when children were fed nothing but breastmilk, whether directly from the breast or pumped in advance, with no other solid or liquid foods, with the exception of vitamins, minerals or medications in the form of drops or syrups, during the 24 hours prior to the interview; predominant breastfeeding as when water, teas, or juices were given in addition to breastmilk; complemented breastfeeding as when the child was fed breastmilk and complementary foods; mixed breastfeeding when the child

was given breastmilk and other milks; and, finally, other milk if the child had not been fed breastmilk.

Prenatal care was defined as adequate if it included six or more consultations starting within the first trimester; inadequate if there had been four or five consultations starting by the sixth month of pregnancy, or six or more consultations started after the fourth month; deficient if the number of consultations was three or less and deficient if the mother had had prenatal care, but was unable to say how many consultations she had attended or in which month she had first attended; and, finally, as no prenatal care if the mother had not attended any consultations. This classification was based on the minimum criteria adopted by the Brazilian Ministry of Health (MS)<sup>13</sup> adapted by the authors of this study.

Children were weighed within 24 hours of birth. At 6 months, weight and length were measured by two duly trained research assistants using the standardized equipment, techniques and procedures recommended by the WHO.<sup>14</sup> Nutritional status was classified using the WHO software Anthro 2006, version 2.0, on the basis of z scores for weight/age and length/age. The cutoff point chosen for malnutrition was  $< -2$  z scores.

The data collected were coded and recorded on dedicated forms, then double-input on Epi-Info, version 6.04, after weekly reviews to check for inconsistent coding. Statistical analyses were carried out using the Statistical Package for the Social Sciences (SPSS), version 12. The t test and analysis of variance (ANOVA) were used to compare the children's mean hemoglobin levels. Since the hemoglobin concentrations of children on exclusive/predominant breastfeeding were asymmetrical, medians were used to measure central tendency and the Wilcoxon test was used to compare it with maternal hemoglobin concentration. The level of statistical significance was set at  $p \leq 0.05$ .

Variables with statistical significance of less than 0.20 in the bivariate analyses were analyzed using hierarchical multivariate linear regression. Model 1 contained the socioeconomic variables (type of flooring, WC and water supply), model 2 included variables related to the children (birth weight and type of delivery) and model 3 analyzed maternal hemoglobin concentration.

The study was approved by the Research Ethics Committee at the Instituto Materno Infantil de Pernambuco (IMIP) (Protocol number 079). Consent was obtained from the mother, or the person responsible for the child, who were invited to participate after the study objectives and procedures had been explained.

## Results

The frequency of anemia among the 330 infants assessed at 6 months of age (hemoglobin  $< 11$  g/dL) was

65.2% [95% confidence interval (95%CI) 59.9-70.2], with 26 children (7.9%; 95%CI 5.3-11.2) having hemoglobin below 9.0 g/dL and just one with hemoglobin  $< 7.0$  g/dL. Mean hemoglobin was 10.5 g/dL (standard deviation = 1.2 g/dL) and the frequency of maternal anemia (hemoglobin  $< 12$  g/dL) was 31.5% (95%CI 26.8-36.9).

The majority of families (82%) had monthly family income per capita of less than or equal to half the minimum monthly wage (the minimum wage was R\$ 200.00). Around 23% of the mothers were illiterate or could only read with difficulty and 32% were adolescents. Just 5% of the mothers had not attended prenatal consultations, but only 37% had received prenatal care considered adequate.

Table 1 shows that mean hemoglobin was lower among children who lived in homes with low-quality flooring and among children born by caesarean.

Table 2 illustrates that more than half of the children had stopped breastfeeding by 6 months and that just 25% were being breastfed exclusively/predominantly. There was no significant association between type of feeding and children's hemoglobin concentration.

Table 3 demonstrates that mean hemoglobin among children of anemic mothers was lower than for children whose mothers were not anemic ( $p = 0.04$ ). In the group of children on exclusive/predominant breastfeeding, median hemoglobin was 0.7 g/dL lower among the children of anemic mothers when compared with the children of mothers without anemia ( $p = 0.03$ ).

The effect of maternal anemia on the hemoglobin concentration of children at 6 months of age remained significant after adjustment of the multivariate linear regression models. Socioeconomic conditions, represented by type of flooring at home, birth weight and type of delivery, also made a significant contribution to the children's hemoglobin (Table 4).

## Discussion

In this study, two-thirds of the children exhibited anemia at 6 months of age, corroborating prevalence data for the study area and the state of Pernambuco as a whole.<sup>15,16</sup> Mean hemoglobin among mothers 6 months after delivery was significantly lower among those who said they had suffered from anemia during pregnancy (data not shown). This datum, in combination with other socioeconomic variables, suggests that anemia had been present since pregnancy. Mean hemoglobin was also significantly lower among children whose mothers were anemic. These results are in agreement with accumulated evidence, since it has been observed that children born to anemic mothers have reduced iron reserves, even when born at full term and with normal weight.<sup>5,6,17</sup> A study conducted in Pernambuco with a sample representative of the state

**Table 1** - Mean hemoglobin at 6 months of life, broken down by maternal socioeconomic and reproductive variables

| Variables                               | n (%) (n = 330) | mean Hb | SD  | p       |
|---|-----------------|---------|-----|---------|
| Family income per capita (MMW)*         |                 |         |     |         |
| ≤ 0.25                                  | 143 (51.8)      | 10.5    | 1.0 | 0.53    |
| 0.25-0.50                               | 84 (30.4)       | 10.5    | 1.2 |         |
| > 0.50                                  | 49 (17.8)       | 10.7    | 1.3 |         |
| Literacy of mother†                     |                 |         |     |         |
| Illiterate                              | 33 (10.0)       | 10.5    | 1.3 | 0.65    |
| Reads with difficulty                   | 13 (13.0)       | 10.3    | 1.1 |         |
| Reads easily                            | 254 (77.0)      | 10.5    | 1.1 |         |
| Flooring                                |                 |         |     |         |
| Ceramics                                | 53 (16.0)       | 11.0    | 1.1 | < 0.001 |
| Concrete/earth                          | 277 (84.0)      | 10.4    | 1.2 |         |
| Water supply                            |                 |         |     |         |
| Running water inside                    | 303 (91.8)      | 10.5    | 1.2 | 0.06    |
| No running water                        | 27 (8.2)        | 10.1    | 0.9 |         |
| WC                                      |                 |         |     |         |
| Yes                                     | 304 (92.1)      | 10.5    | 1.1 | 0.09    |
| No                                      | 26 (7.9)        | 10.1    | 1.3 |         |
| Size of family (people living together) |                 |         |     |         |
| ≤ 4                                     | 118 (35.8)      | 10.5    | 1.1 | 0.32    |
| 5-6                                     | 114 (34.5)      | 10.6    | 1.1 |         |
| > 6                                     | 98 (29.7)       | 10.4    | 1.3 |         |
| Age of mother (years)                   |                 |         |     |         |
| < 20                                    | 106 (32.1)      | 10.5    | 1.3 | 0.98    |
| 20-29                                   | 186 (56.4)      | 10.5    | 1.1 |         |
| ≥ 30                                    | 38 (11.5)       | 10.5    | 0.9 |         |
| Parity                                  |                 |         |     |         |
| 1                                       | 127 (38.5)      | 10.5    | 1.2 | 0.29    |
| 2                                       | 81 (24.5)       | 10.3    | 1.0 |         |
| ≥ 3                                     | 122 (37.0)      | 10.6    | 1.2 |         |
| Reported having anemia when pregnant †  |                 |         |     |         |
| Yes                                     | 179 (59.7)      | 10.5    | 1.2 | 0.64    |
| No                                      | 121 (40.3)      | 10.5    | 1.0 |         |
| Quality of prenatal care                |                 |         |     |         |
| Adequate                                | 121 (36.7)      | 10.5    | 1.1 | 0.90    |
| Inadequate                              | 117 (35.4)      | 10.4    | 1.3 |         |
| Deficient                               | 75 (22.7)       | 10.6    | 1.0 |         |
| None                                    | 17 (5.2)        | 10.4    | 1.4 |         |
| Type of delivery                        |                 |         |     |         |
| Normal                                  | 232 (70.3)      | 10.6    | 1.1 | 0.01    |
| Caesarean                               | 98 (29.7)       | 10.3    | 1.2 |         |

Hb = hemoglobin; MMW = multiples of minimum wage (R\$ 200.00 per month); SD = standard deviation.

\* Information missing on 54 cases.

† Information missing on 30 cases.

found a significant association between the prevalence of anemia among mothers and among their children less than 36 months old.<sup>18</sup>

Despite the increase in the prevalence of exclusive breastfeeding at 6 months in the area studied,<sup>10</sup> the number of children in this category was small and so they were analyzed together with the group of children on predominant breastfeeding. Even so, there were no significant associations between type of feeding and mean hemoglobin. However, when just the group of children on

exclusive/predominant breastfeeding were analyzed, it was observed that median hemoglobin was significantly lower, in the order of 0.7 g/dL, for those whose mothers were anemic. Since type of feeding was measured at a single time point, children who stopped exclusive breastfeeding very early on were assigned to the same category as those who stopped soon before reaching 6 months of age, which could have contributed to underestimating the influence of exclusive/predominant breastfeeding on mean hemoglobin concentration in these children.

A study conducted in Indonesia with breastfeeding children aged 3 to 5 months found an elevated prevalence of anemia and observed that maternal hemoglobin was an important associated factor.<sup>17</sup> Maternal anemia therefore emerges as a fundamental factor in infant anemia, since its onset is generally before the start of breastfeeding. Evidence demonstrates that the iron content of breastmilk is independent of the mother's iron status.<sup>19</sup>

Data in the literature on the association between breastfeeding and anemia are conflicting. Some authors have not detected any association,<sup>20</sup> as observed in this study, while others identify breastfeeding, especially exclusive, as an important factor of protection in infants less than 6 months old.<sup>21,22</sup>

Another study of infants under 6 months found a risk of anemia around two and three times greater among children

**Table 2 -** Mean hemoglobin at 6 months of life, broken down by children's biological and nutritional characteristics

| Variables                        | n (%) (n = 330) | mean Hb | SD  | p    |
|----------------------------------|-----------------|---------|-----|------|
| Sex                              |                 |         |     |      |
| Male                             | 184 (55.8)      | 10.5    | 1.2 |      |
| Female                           | 146 (44.2)      | 10.5    | 1.1 | 0.98 |
| Birth weight (g)                 |                 |         |     |      |
| < 2,500 g                        | 13 (3.9)        | 10.1    | 1.3 |      |
| 2,500-2,999 g                    | 94 (28.5)       | 10.4    | 1.0 |      |
| ≥ 3,000 g                        | 223 (67.6)      | 10.6    | 1.2 | 0.18 |
| Breastfeeding at 6 months        |                 |         |     |      |
| Exclusive/predominant            | 82 (24.8)       | 10.6    | 1.1 |      |
| Mixed and/or complemented        | 75 (22.8)       | 10.4    | 1.3 |      |
| Other milk                       | 173 (52.4)      | 10.5    | 1.1 | 0.31 |
| Weight/age at 6 months (z score) |                 |         |     |      |
| < -2                             | 7 (2.1)         | 10.2    | 0.7 |      |
| ≥ -2                             | 323 (97.9)      | 10.5    | 1.2 | 0.49 |
| Length/age at 6 months (z score) |                 |         |     |      |
| < -2                             | 19 (5.8)        | 10.4    | 1.3 |      |
| ≥ -2                             | 311 (94.2)      | 10.5    | 1.2 | 0.61 |

Hb = hemoglobin; SD = standard deviation.

**Table 3 -** Hemoglobin concentration at 6 months, broken down by maternal anemia, for entire sample and for infants on exclusive/predominant breastfeeding

|  | Hemoglobin (g/dL) |                  |                   |
|--|-------------------|------------------|-------------------|
|  | n (%)             | Concentration    | p                 |
| Entire sample (n = 330), mean ± SD*  |                   |                  |                   |
| Maternal hemoglobin ≥ 12 g/dL  | 222 (68.5)        | 10.6±1.1         |                   |
| Maternal hemoglobin < 12 g/dL  | 102 (31.5)        | 10.3±1.3         |                   |
| Difference   |                   | -0.3             | 0.04              |
| Children on exclusive/predominant breastfeeding (n = 82), median (25-75%IQ) <sup>†</sup> |                   |                  |                   |
| Maternal hemoglobin ≥ 12 g/dL  | 61 (75.3)         | 10.8 (10.0-11.3) |                   |
| Maternal hemoglobin < 12 g/dL  | 20 (24.7)         | 10.1 (9.8-10.6)  |                   |
| Difference   |                   | -0.7             | 0.03 <sup>‡</sup> |

25-75%IQ = 25-75% interquartile range, SD = standard deviation.

\* Information missing on 6 cases.

<sup>†</sup> Information missing on 1 case.

<sup>‡</sup> Wilcoxon test.

**Table 4** - Multivariate linear regression of factors associated with hemoglobin concentration in 6-month-old infants

| Variables                                      | Hemoglobin at 6 months of age |       |                    |                |       |
|--|-------------------------------|-------|--------------------|----------------|-------|
|  | $\beta^*$ unadjusted          | p     | $\beta^*$ adjusted | 95%CI          | p     |
| Level 1  |                               |       |                    |                |       |
| Type of flooring (concrete/earth) <sup>a</sup> | -0.57                         | 0.001 | -0.58              | -0.92 to -0.24 | 0.001 |
| Level 2  |                               |       |                    |                |       |
| Birth weight (kg)                              | 0.35                          | 0.02  | 0.32               | 0.03 to 0.60   | 0.03  |
| Type of delivery (caesarean) <sup>a</sup>      | -0.34                         | 0.02  | -0.33              | -0.60 to -0.06 | 0.02  |
| Level 3  |                               |       |                    |                |       |
| Maternal hemoglobin (< 12 g/dL) <sup>a</sup>   | -0.28                         | 0.04  | -0.27              | -0.53 to 0.00  | 0.05  |

95%CI = 95% confidence interval.

\* Regression coefficient.

<sup>a</sup> Reference categories = ceramic (type of flooring), vaginal delivery (type of delivery), maternal hemoglobin  $\geq$  12 g/dL.

Level 1 = adjusted for type of water supply and WC.

Level 2 = adjusted for the variables on this level and by the level 1 variable.

Level 3 = adjusted for the variables on levels 1 and 2.

on breastfeeding and artificial feeding, respectively, when compared with those who were exclusively fed breastmilk, irrespective of socioeconomic conditions or birth weight.<sup>21</sup> Another study, conducted with children on exclusive breastfeeding in the same age group, found that they had higher hemoglobin levels when compared with children on different feeding regimes.<sup>22</sup> Another study identified breastfeeding as a risk factor for anemia in infancy; although it defined children as on breastfeeding if they were given formula less than four times a day, which could have impacted on the results.<sup>23</sup>

Recently, considerable advances have taken place in scientific knowledge of the basic mechanisms related to the benefits of breastfeeding, and its clinical management.<sup>24</sup> The impact on infant mortality of exclusive breastfeeding with 90% coverage has also been documented.<sup>25,26</sup> Countless advantages of breastfeeding and human milk for child nutrition, have been described for children, their mothers and families and for society.<sup>24</sup> Despite this, several different studies of childhood anemia have not correctly analyzed the role of breastmilk as an important factor in its control and prevention.<sup>27</sup>

With relation to other determinants of anemia in infants, our study found that, in addition to maternal anemia, there was a significant association with the type of flooring at home, which is an indicator of a family's socioeconomic conditions, and also with type of delivery and birth weight.

Mean hemoglobin concentration was significantly lower among children who were born by caesarean. This could be related to the position in which the baby is removed during a caesarean (higher than the mother) and also to the fact that the ligation of the cord occurs earlier in a surgical

delivery. Delaying clamping the cord could benefit the baby by a placental transfusion of 35 mL of blood per kg of weight and 30 to 50 mg extra iron reserves.<sup>7</sup> A randomized study demonstrated that 3 months after delivery, mean serum ferritin and hemoglobin were significantly greater in a delayed clamping group in children born to anemic mothers.<sup>28</sup> In another study of delayed clamping of the umbilical cord, significantly higher levels of ferritin were observed at 3 months of age when compared with those whose cords were clamped immediately.<sup>29</sup>

Birth weight had a significant and independent association with the concentration of hemoglobin, which is similar to data observed by other researchers.<sup>16,22,30</sup> The direct relationship between mean hemoglobin and birth weight indicates that the iron reserves of children born full term with low birth weight put them at risk of developing anemia during infancy.<sup>9</sup>

There was no relationship between the hemoglobin levels of the child and the mother's educational level or with the quality of prenatal care. Although the majority of mothers had attended prenatal consultations, only one third met the minimum quality expected. In Brazil, prenatal coverage has been widened, but the available data demonstrate that the quality of care has been compromised.<sup>13</sup> The high frequency of low quality care may explain why there was no association between quality of prenatal care and mean hemoglobin levels in the infants.

Finally, we conclude that this study raises points that are relevant to the health of children, demonstrating that the frequency of anemia at 6 months is extremely elevated in the area studied, agreeing with recent data in the literature.<sup>15,16</sup> Despite the significant increase in the

prevalence of exclusive breastfeeding resulting from the recent intervention,<sup>10</sup> it remains well below the 90% that is desirable to have an impact on children's health. The frequency of anemia among the mothers is also elevated, with evidence that it is long-term, which may have had repercussions for the iron status of their children at birth. The quality of prenatal care was deficient, which probably did not have a positive influence on the health of these women and their children.

Since the etiology of anemia is multicausal and complex, involving socioeconomic, geographical, biological and cultural factors, an effective and sustainable strategy to prevent it must be based on nutritional education of the population and a fairer distribution of resources. The iron status of children at birth is a fundamental factor in the development of anemia among infants and is influenced by their mothers' iron status, going back to the pre-conception period. It is therefore necessary to guarantee good nutritional status and adequate iron reserves for all women, from adolescence onwards, so that they are prepared for the demands of pregnancy, maintaining a good supply during pregnancy and lactation.

The prevention of iron deficiency anemia in infants should be based on an integrated approach, including delayed clamping of the umbilical cord and support for breastfeeding. This, according to WHO recommendations, should be exclusive up until 6 months and then maintained at least up to 2 years. We suggest that studies of anemia in children, especially in infants, should evaluate the iron status of mothers as being an important risk factor, in addition to the type of feeding given during the first 6 months of life.

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