Anemia-associated factors in infants born at term with normal weight

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Summary

Objective: To investigate the factors involved in the genesis of infant iron deficiency anemia. **Methods:** This is a cross-sectional study, which evaluated 104 children in their second year of life who were born at term with adequate weight in Viçosa, Minas Gerais, Brazil. An interview, a 24-hour recall to parents, and anthropometric assessment were used. Laboratory exams included blood count, ferritin, and serum retinol. This study was approved by the Ethics Committee of UFMG and UFV. Statistical analyses were conducted using the Epi Info and SPSS softwares. Poisson generalized linear regression model was used to determine the association of anemia with the study variables, with results expressed as prevalence ratio. **Results:** Vitamin A deficiency and anemia were identified in 9.6% e 26% of the children, respectively. Infant anemia was associated with the date of onset of prenatal care, maternal use of iron after childbirth, paternal working status, prior use of iron by the child, and duration of breastfeeding. Thus, in the second year of life, lactating children of women who began prenatal care late and did not use iron compounds after birth, with unemployed parents, who never received iron compounds, and who were predominantly breastfed for more than four months had significantly higher prevalence of anemia. **Conclusion:** The results have demonstrated the importance of nutrition during pregnancy and infancy in the prevention of anemia in children.

Keywords: Feeding; anemia; pregnancy; prevalence ratio; vitamin A.

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Introduction

Iron-deficiency anemia is considered a public health problem in Brazil and worldwide, affecting, mainly, children and pregnant women. Due to its magnitude and harmful effects, it is one of the main nutritional deficiencies[1].

In Brazil, the prevalence of anemia in children range from 20% to 70% depending on the region, age, and socioeconomic conditions, among others[2].

Causes of anemia in infancy are mainly secondary to the high iron requirements associated with deficient consumption of this mineral[3]. Other factors have been reported such as: low family income, inadequate duration of breastfeeding, low parent schooling[4], elevated number of people residing in the same household[5], low weight at term, and children of adolescent mothers[6], among others. Considering these factors, we emphasize gestational age and weight at term, since iron stores accumulated by the child during pregnancy depends on the weight and develops, mainly, in the last trimester. These stores, associated with exclusive breastfeeding during the first six months, guarantee the adequate nutritional status of the child[7].

Although strategies to control anemia have been widely reported, it still has an elevated prevalence rate in the Brazilian population, which justifies the search for other predisposing factors for this deficiency to guarantee better approach to anemia. Therefore, the objective of the present study was to investigate the factors involved in the genesis of infant iron-deficiency anemia in those infants born at term with normal weight in a group of children in Viçosa, Minas Gerais, Brazil.

Methods

This is a transversal study developed as part of a larger study entitled "Evaluation of different iron and vitamin A schedules in the treatment and prevention of iron deficiency anemia in children in the second year of life" conducted in Viçosa, Minas Gerais, Brazil.

Data on the Liveborn Declaration of the municipality of Viçosa was used to select participants. Children in the age group of interest were visited at their homes, where their parents were interviewed to determine the presence of inclusion criteria, which were: children living in the urban area of the county, without neonatal intercurrences or congenital anomalies, born of a single pregnancy, at term, with weight at birth ≥ 2,500 g, ages from 12 to 20 months, and born to women aged ≥ 20 years. Exclusion criteria were refusal to participate in the study, not at home at the time of interview, and those who did not perform all the steps of the study. Therefore, 104 children were evaluated. The final number was obtained considering the sample necessary for the later segment of the study, which was a clinical assay.

During the interview, child identification, socioeconomic status and living conditions, maternal variables, morbidities, vaccination history, and nutritional practices were collected. All interviews were conducted by the same investigator. Dietary, parasitological, laboratorial, and anthropometric assessment were also conducted.

Through a 24-hour record, dietary composition (calories, iron, vitamin A, and vitamin C) was evaluated with the Diet-Pro 4.0 software. Dietary ingestion was evaluated based on DRIs using the EAR, as a cutting point, or the AI, to evaluate the proportion of individuals with adequate ingestion.

The density of nutrients was calculated considering the mg of nutrient per 1,000 kcal. Due to the lack of recommendations for the analysis of inadequate dietary density, a calculation was performed to estimate the recommended value considering nutritional and caloric recommendations (EER calculated with the mean weight of the children evaluated).

Parasitological test was performed to detect the presence of parasites and to identify them. On laboratorial assessment, parameters of iron nutritional status evaluated included hemoglobin, hematocrit, MCV, MCH, MCHC, erythrocytes, and ferritin, obtained through an electronic counter. Serum retinol levels were used to evaluate vitamin A nutritional status using high-performance liquid chromatography (HPLC).

Weight and height were used in anthropometric evaluation, calculating weight/age, height/age, weight/height, and BMI/age indices, expressed as Z-scores. To detect nutritional deviations, Z-score cutting points 2 and -2 were used. World Health Organization growth curves[8] were considered reference, and the WHO Anthro software was used to analyze the data.

The Epi Info 6.04 and SPSS version 15.0 software were used for statistical analysis. The first one was used for descriptive analysis and the second for the rest. Before the analyses, the data base underwent consistency verification, which means verify the presence of outliers and typing errors. Whenever inconsistencies were detected, the original files were consulted for clarification and corrections.

Correlation analyses were performed among laboratorial and dietary data using Pearson' or Spearman's correlation test, the first was used for symmetrical variables and the second for asymmetrical variables. To verify the symmetry of variables, the Shapiro-Wilk test was used.

The association between anemia and other variables was evaluated by the Poisson generalized linear regression model with robust variance and the results were expressed as prevalence ratios. This model was chosen due to the high prevalence of the outcome, which would determine significantly higher odds ratio than prevalence ratios if logistic regression were used.

Pearson's Chi-square test and tendency Chi-square test were used for univariate analysis. Multivariate analysis was used for variables with a level 0.20 association. Then, less significant variables were slowly removed, until a model in which all variables had significance level of 0.05 remained.

The study protocol was approved by the Ethics on Human Research Committees of Universidade Federal de Viçosa and Universidade Federal de Minas Gerais. Those responsible for the children were informed on the objectives and procedures of the study; those who agreed to participate signed an informed consent.

Results

One-hundred and four children with mean age of 15.9 ± 2.2 months were evaluated; 53.8% were females and 46.2%, males.

Table 1 shows some of the socio-demographic characteristics of the study population.

Underweight was not observed, but 4.8% of the children were overweight when we used the weight-to-height index. Low stature was observed in 6.7% of them.

Ten children (9.6%) had inadequate nutritional status for vitamin A, and 27 (26%) were anemic, and the mean retinol and hemoglobin levels were 30.9 ± 17.2 mg/L and 11.6 ± 1.3 mg/dL, respectively. Regarding intestinal parasitosis, 10.4% had infestations, and among them, 40% had *Ascaris lumbricoides* and the others *Giardia lamblia*.

The median consumption of vitamin C, calcium, vitamin A, iron, and iron density were: 47.8 mg; 753.9 mg; 518.3 µg; 5.2 mg; 4.4 mg/1,000 kcal, respectively. The percentage of infants with consumption lower than the EAR or AI was 8.7%; 33.7%; 9.6%; 17.3%; and 31.7% for vitamin C, calcium, vitamin A, iron, and iron density, respectively.

To correlate dietary and laboratorial parameters, correlation analysis was performed (Table 2).

Positive correlations were observed among some of the laboratorial and dietary variables, especially those that assess the amount of iron in the diet. Besides, dietary ferritin and vitamin A showed negative correlation and calcium showed correlation with hematocrit. However, none of them were strong correlations.

Among variables that did not show significant correlation with the frequency of anemia are: gender, serum retinol levels, vitamin A consumption, dietary density of vitamin A, vitamin C consumption, dietary density of vitamin C, dietary density of calcium, nutritional status, current intestinal parasitosis, prenatal care, use of iron compounds during gestation, onset of gestational supplementation, regular use of gestational supplementation, gestational use of vitamin complex, type of birth, post-gestational use of vitamin complex, parity, multigerational household, number of people in the household, presence of children younger than 5 years old in the household, maternal schooling, maternal marital status, maternal working status, presence of diarrhea, fever, sore throat, coughing, runny nose, bronchitis, infection, ear pain or allergies in

Table 1 – Socio-demographic characteristics of a group of infants born at term with adequate weight. Viçosa, Minas Gerais, Brazil, 2006/2007

Varibles	n (%)
Multigerational household	
Yes	20 (19.2)
No	84 (80.8)
Number of people in the household	
≤ 5 people	91 (87.5)
> 5 people	13 (12.5)
Younger than 5 years old in the household	
0	72 (69.2)
≥ 1 child	32 (30.8)
Maternal schooling	
0 to 4 years	29 (27.9)
≥ 5 years	75 (72.1)
Paternal schooling	
0 to 4 years	29 (28.7)
≥ 5 years	72 (71.3)
Maternal marital status	
Married (formal or informally)	86 (82.7)
Not married (separated, single, widow)	18 (17.3)
Paternal working status	
Formal	54 (52.9)
Informal	41 (40.2)
Unemployed	7 (6.9)
Maternal working status	
Formal	8 (7.7)
Informal	17 (16.3)
Unemployed + "housewife"	79 (76)
Income*	
< R$ 445,00	47 (49.5)
≥ R$ 445,00	48 (50.5)
Per capita income*	
< R$116,60	43 (45.3)
≥ R$116,60	52 (54.7)
Beneficiary of any government program	
Yes	24 (23.1)
No	80 (76.9)
Garbage collection	
Ideal	104 (100)
Not ideal	0 (0)
Water supply	
Ideal	103 (99.1)
Not ideal	1 (0.9)

*Cutting point based on the median of the study population.

Table 2 – Correlation between laboratorial and dietary variables of a group of infants born at term with adequate weight. Viçosa, Minas Gerais, Brazil, 2006/2007

Dietary variables	Laboratorial variables							
	Hb	Ht	MCV	MCH	MCHC	Erythrocytes	Ferritin[+]	Retinol
Calories	0.198[2*]	0.263[1*]	0.100[2]	0.065[2]	0.077[1]	0.136[1]	0.123[2]	0.236[2*]
Iron	0.226[2*]	0.251[2*]	0.224[2*]	0.155[2]	0.032[2]	0.014[2]	0.098[2]	0.240[2*]
Iron density	0.180[2]	0.172[2]	0.242[2*]	0.18[2*]	0.056[2]	-0.100[2]	0.058[2]	0.165[2]
BFe	0.102[2]	0.099[2]	0.140[2]	0.085[2]	0.016[2]	-0.074[2]	0.056[2]	0.241[2*]
BFe density	0.073[2]	0.051[2]	0.141[2]	0.096[2]	0.024[2]	-0.138[2]	0.017[2]	0.197[2*]
HFe	0.055[2]	0.041[2]	0.010[2]	0.023[2]	-0.029[2]	0.014[2]	0.031[2]	0.152[2]
HFe density	0.028[2]	0.013[2]	-0.003[2]	0.009[2]	-0.043[2]	-0.004[2]	0.042[2]	0.133[2]
Vitamin A	-0.03[2]	-0.06[2]	0.055[2]	0.069[2]	0.04[2]	-0.07[2]	-0.23[2*]	-0.021[2]
Vitamin A density	-0.14[2]	-0.19[2]	-0.02[2]	0.012[2]	0.019[2]	-0.111[2]	0.151[2]	-0.147
Vitamin C	0.057[2]	0.186[2]	0.132[2]	0.027[2]	-0.115[2]	0.045[2]	0.071[2]	0.169[2]
Vitamin C density	-0.02[2]	0.102[2]	0.087[2]	-0.01[2]	-0.137[2]	0.009[2]	0.019[2]	0.108[2]
Calcium	0.165[2]	0.207[2*]	0.069[2]	0.035[2]	0.015[2]	0.139[2]	0.120[2]	0.180[2]
Calcium density	0.123[2]	0.142[2]	0.028[2]	0.013[2]	0.015[2]	0.104[2]	0.078[2]	0.101[2]

Hb, hemoglobin; Ht, hematocrit; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration; BFe, bioavailable iron; HFe, heme iron. *p-value < 0.05; [+]Test performed in 76 children, since the amount of the sample of some children was not enough; Variables that presented normalcy by the Shapiro-Wilk test: hematocrit, MCHC, erythrocytes, calories, protein, lipids. [1]Pearson's correlation; [2]Spearman's correlation.

the last 15 days, presence of parasites in prior exam, current use of iron compounds or vitamin complex, breastfeeding, duration of exclusive breastfeeding, type of milk consumed currently, and ingestion of milk close to meals.

Anemia showed a significant association level 0.20 with the following variables: iron density, calcium consumption, onset of prenatal care, number of pre-natal appointments, presence of gestational anemia, post-partum use of iron compounds, paternal schooling, paternal working status, low income, low per capita income, benefits from any governmental program, disease in the last 15 days, presence of anemia in prior exam, prior use of iron compounds, current breastfeeding, duration of predominant breastfeeding, and age of introduction of supplementary feeding (Table 3).

Univariate analysis showed that children who had diets with low iron density and high calcium levels had higher incidence of anemia, as well as children who were maintained predominantly on breastfeeding longer than 4 months.

In this sample, mean duration of breastfeeding was 210 days (minimum of zero and maximum of 600 days). The duration of exclusive and predominant breastfeeding was 60 days (0-240 days) and 102 days (0-240 days), respectively. Regarding the age of introduction of supplementary feeding, we observed a median of 155 days, ranging from 30 to 270 days.

As for gestational and post-partum data, it was observed that women who began prenatal care after three months of gestation and had less than six appointments, as well as those who did not take post-partum iron compounds, had children with higher frequency of anemia.

All variables associated with the outcome at a level of significance lower than 0.20 were included in the multivariate analysis. After adjustment of variables, the post-partum or prior use of iron compounds, onset of prenatal care, duration of predominant breastfeeding, and paternal working status remained associated with anemia (Table 4). Thus, in the second year of life, infants born to women who began prenatal care late in their gestation and did not use iron compounds after birth, parental unemployment, who never received iron compounds, and who were predominantly breastfed for more than four months had higher frequency of anemia.

Discussion

Although transversal studies are not the most adequate to determine risk factors, since all the data is observed at the same time, it has been frequently used to diagnose health problems. The 24-hour record, despite being a fast, low cost, and easy to apply method, it does not reflect habitual ingestion, depends on the memory of the interviewee, and requires a well trained interviewer. However, this method provides reliable estimates of mean consumption of

Table 3 – Distribution of the study variables according to frequency, gross prevalence ratio (PR), according to Poisson generalized Linear Regression Model and respective 95% confidence interval (95% CI) at the level of 0.20 in a group of infants born at term with adequate weight. Viçosa, Minas Gerais, Brazil, 2006/2007

Variables	Frequency (%)	Prevalence ratio (PR)	95% CI-PR	p-value
Onset of prenatal care				
≥ 3 months	37.3	3.082	1.350-7.036	0.008*
< 3 months	12.2	1		
Number of prenatal appointments				
< 6	38.3	2.585	1.239-5.393	0.011*
≥ 6	14.8	1		
Presence of gestational anemia				
Yes	35.9	2.010	0.997-4.052	0.051
No	17.9	1		
Postpartum use of iron compounds				
No	33.3	2.533	1.045-6.140	0.040*
Yes	13.2	1		
Paternal schooling				
0 to 4 years	37.9	1.821	0.953-3.480	0.070
≥ 5 years	20.8	1		
Paternal working status				
Formal	18.5	1		
Informal	34.1	1.844	0.913-3.723	0.088
Unemployed	42.9	2.314	0.833-6.432	0.108
Income+				
< R$ 445,00	34.0	1.165	0.978-1.388	0.086
≥ R$ 445,00	18.8	1		
Per capita income+				
< R$ 116,60	34.9	1.169	0.979-1.398	0.085
≥ R$ 116,60	19.2	1		
Beneficiary of any government program				
Yes	37.5	1.667	0.864-3.216	0.128
No	22.5	1		
Sick in the last 15 days				
Yes	30.3	1.645	0.767-3.527	0.201
No	18.4	1		
Anemia on a prior exam				
Yes	45.2	6.022	1.896-19.120	0.002*
No	7.5	1		
Prior use of iron compounds				
No	40.0	1.859	0.982-3.520	0.057
Yes	21.5	1		
Current breastfeeding				
Yes	37.5	1.750	0.929-3.297	0.083
No	21.4	1		
Iron density				
< 3.58 mg/1,000 Kcal	37.5	1.998	1.062-3.758	0.032
≥ 3.58 mg/1,000 Kcal	18.8	1		
Dietary calcium				
≥ 500 mg	40.0	3.962	2.841-5.523	0.000*
< 500 mg	18.8	1		
Duration of predominant breastfeeding				
> 4 months	40.0	2.327	1.205-4.495	0.012*
≤ 4 months	17.2	1		
Age supplemental feeding was introduced				
> 4 months	31.7	1.769	0.825-3.791	0.143
≤ 4 months	17.9	1		

*p < 0,05; +Cutting point based on median.

Table 4 – Distribution of the study variables according to prevalence ratios (PR) adjusted as per Poisson generalized Linear Regression Model and respective 95% confidence intervals (95% CI) in a group of infants born at term with adequate weight. Viçosa, Minas Gerais, Brazil, 2006/2007

Variables	Prevalence ratio (PR)	95% CI-PR	p-value
Onset of prenatal care			
≥ 3 months	3.541	1.717-7.302	0.0006
< 3 months	1		
Postpartum use of iron compounds			
No	3.072	1.30-7.26	0.011
Yes	1		
Paternal working status			
Formal	1		
Informal	1.640	0.852-3.159	0.139
Unemployed	4.002	1.416-11.310	0.009
Prior use of iron compounds			
No	1.929	1.062-3.505	0.031
Yes	1		
Duration of predominant breastfeeding			
> 4 months	2.603	1.503-4.508	0.0006
≤ 4 months	1		

Poisson generalized linear model with logarithmic connecting function. Value of p in the Chi-square test and deviance were, respectively, 0.581 and 0.509 resulting from good quality adjustment.

populations, even when applied a single time, being widely used[9]. Memory is another possible bias, which occurs when using recollected variables by the individual through interview. There is also the possibility of biases related to characterization and selection of the study population.

In the present study, some positive correlations were observed between parameters of nutritional status and dietary iron. This fact demonstrates not only that the amount of iron consumed is important, but also its proportion as a function of the caloric density of the diet because diets with high calorie content can reach the gastric capacity of infants, hindering adequate ingestion of this mineral.

Other studies also observed the relationship between diet and laboratorial parameters, with mean iron densities significantly lower in anemic children compared to non-anemic children[10]; association of anemia and iron density[11], iron bioavailability[12] and bioavailable iron[13]; and association of severe anemia and inadequate iron intake[14], and even the association of bioavailable iron and hemoglobin[15] and anemia[16]. Therefore, children with lower consumption had greater risk of this deficiency. Inadequate feeding seems to constitute a risk factor for anemia.

The pursuit of infant diet adaptation starts with breastfeeding and introduction of adequate complementary feeding to guarantee that iron requirements are met. In the studied population, the duration of breastfeeding was not adequate, with total time and exclusive medians below those recommended by the World Health Organization[17]. Estimated median age of children exclusively breastfed in the National survey of Demographics and Health with a representative sample of the Brazilian population was 2.2 months[18]. A study with infants in public daycare centers in São Paulo demonstrated that exclusive breastfeeding for less than two months is a risk factor for anemia[4].

A study in Canada showed that an educational approach based on foodstuff improves infant iron consumption[19], which should be encouraged. In Brazil, according to recommendations of the Health Ministry, complementary feeding should be adequately oriented, since it has an important role on iron supplies during infancy[20].

Although breastfeeding is important in preventing infancy anemia[21], maintenance of exclusive breastfeeding for longer than six months increases the risk of anemia in children[22]. In the present study, duration of predominant breastfeeding over four months was a risk factor for anemia. It is known that introduction of foodstuff reduces iron bioavailability in breast milk, and this might be an explanation. Maintenance of breastfeeding occurs in detriment of supplementary feeding, which is very important to guarantee iron intake, being another hypothesis observed in clinical practice.

Dietary calcium, a known inhibitor of iron absorption, did not show negative correlation with laboratorial iron parameters. On the contrary, it showed a positive correlation with hematocrit. This correlation was not expected, since calcium has a negative association with iron. It most likely represents a confounding factor, which was eliminated in the multivariate analysis.

Dietary vitamin A showed positive correlation with ferritin. A possible explanation for this correlation would be the ability of vitamin A to facilitate the use of ferritin stores[23]. Serum retinol showed positive correlation with iron, bioavailable iron, and density of bioavailable iron, corroborating the findings of other studies that showed correlation between vitamin A and iron.

Correlations with ferritin were not observed. The use of ferritin as a marker of infant nutritional status has been discussed, since it changes rapidly in the first months of life. Besides, children with infections can show changes in ferritin, which impairs the isolate use of this parameter[24].

The frequency of anemia and vitamin A deficiency characterizes those deficiencies as moderate public health problems[1]. However, it should be emphasized that the children in this study went through a screening process in which several known risk factors were excluded, making us believe that the frequency is even higher.

Midzi et al.[25] observed greater prevalence of anemia in children with intestinal parasites. This finding was not observed in our study possibly because the intestinal parasites found do not normally cause blood loss.

Although significant correlations between dietary and laboratorial parameters were observed in the present study, those were weak correlations, suggesting that other determinants of hemoglobin levels should be investigated. In an attempt to explain infant anemia, associations between deficiency and some variables were made in this study. Regarding maternal variables, the onset of prenatal care and use of iron compounds after birth were associated with anemia.

It has been suggested that anemia during pregnancy has a negative impact on infant nutritional status[26,27]. A study by Meinzen-Derr et al.[22] observed that maternal anemia was associated with infant anemia, increasing the risk of infant anemia by three-fold. The relationship between maternal and infant nutritional status is due to gestational iron deficiency, especially in the last trimester, promoting an increase in the number of premature birth and low weight. The last one has an impact on infant iron stores, constituting a risk factor for anemia in infancy[1]. However, the study population was composed of children who were born with adequate weight and gestational age.

Iron supplementation during gestation has an impact on the reduction in the prevalence of infant anemia[28]. Women who receive iron supplements during pregnancy in Nigeria had children with greater height on birth and greater concentrations of ferritin three months after birth[29]. However, in a study performed in inner São Paulo state, an influence of the gravidas' iron levels on their children's iron levels was not observed[30].

Schneider et al.[31] observed that participation in a care program for women and children had a positive impact on anemia in low income children in the United States, demonstrating the importance of women care. In Brazil, it is recommended that prenatal care should be initiated on the first trimester of pregnancy to guarantee, at the end of pregnancy, the birth of a healthy child, as well as maternal and neonatal well-being[32]. It is also recommended that iron supplementation should begin on the 20th week of pregnancy and be prolonged for three months after birth[33].

Regarding socio-demographic variables, only paternal working status was associated with anemia; children of unemployed fathers showed greater frequency of anemia than children of employed fathers. Note that better socio-demographic conditions could mean access to better feeding and, consequently, lower prevalence of anemia[7].

Prior use of iron compounds was another variable associated with anemia in the population investigated; children who had never taken this supplement had higher frequency of anemia. This result was also observed in another study performed in Viçosa in 6- to 12-month old infants[34]. Iron supplements in the first two years of life are recommended by the Brazilian Society of Pediatrics, since iron requirements in this age group are elevated and, even with adequate intake, the requirements are difficult to meet. The Brazilian Health Ministry[33] recommends supplementation of iron in children from six to 18 months to prevent iron-deficiency anemia in infancy.

This study indicated some factors that contribute for the development of anemia in infancy; however, it is important to emphasize that there is no way to determine all variables that could possibly interfere with the development of anemia in this age group. It is possible that other parameters not evaluated in the present study, also contribute for the development of this deficiency. For this reason, those should be investigated in further studies.

Conclusion

This study allows the conclusion that some infants, children of women who began prenatal care late in pregnancy and who did not use iron compounds after birth, with unemployed fathers, who never received iron compounds, and were maintained predominantly on breastfeeding for more than four months have higher incidence of anemia. These results demonstrate the importance of maternal nutrition, follow-up of pregnant women, and iron supplements during pregnancy as preventive measures for infant anemia. Besides, the importance of adequate nutrition in infancy with exclusive breastfeeding until six months of age and correct introduction of complementary feeding using good iron sources to meet the requirements of this mineral should be emphasized.

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