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RESEARCH ARTICLE

Neonatal Anemia: Prevalence, Associated Factors, and Impact of Malaria in a Newborn Population at the University Hospital Center of Libreville

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Abstract

Introduction: There is very little data on neonatal anemia in Sub-Saharan Africa, particularly in Gabon. This study aimed to determine the frequency of anemia and its associated factors, including malaria, in a population of newborns.

Methods: Demographic, clinical, and paraclinical variables of the newborns, as well as maternal histories were prospectively collected in 2015. A thick drop and a complete blood count were performed.

Results: Out of the 100 newborns included, 77% were less than 8-days-old, 53% had a low birth weight, and 45% were born prematurely. Most mothers were unemployed (81%), primipara or paucipara (75%) and had fewer than 4 prenatal visits (60%). The prevalence of anemia was 52%, higher among children aged 8 to 15 days (88.9%) ($p < 0.01$). The same trend was observed when mothers had only one dose of IPT-SP or fewer than three prenatal visits. The prevalence of severe anemia was 11%, with moderate anemia predominating at birth (18.9%). Most children with malaria were anemic (61.1%).

Conclusion: The prevalence of neonatal anemia is high. Young age, irregular pregnancy follow-up, and malaria are associated with it.

Keywords

Anemia, Newborns, Factors, Malaria, Gabon

Introduction

Anemia is a major public health issue in developing countries, particularly in Sub-Saharan Africa, affecting all age groups, especially children. The WHO estimates that about 50% of children under four years living in developing countries are anemic [1]. Anemia in children is multifactorial, including nutritional deficiencies, hemoglobinopathies such as sickle cell disease, maternal and neonatal infections, etiologies whose impact on hemoglobin levels varies depending on their prevalence, children's age, and living conditions [2]. It also contributes to increasing pediatric morbidity and mortality [3]. Maternal pathologies during pregnancy and fetal or neonatal anemia are also considered risk factors for anemia during the first six months of life, but also risk factors for neonatal infectious morbidity [4,5]. The prevalence of neonatal anemia is estimated at 23-66% in Africa [6]. Moreover, in newborns, prematurity and low birth weights are significant causes of neonatal and infant mortality [7]. Prevention and proper management of these risk factors will reduce this morbidity and mortality, which requires knowledge of their actual weight within the exposed populations. In Gabon, the frequencies of malaria, prematurity, maternal age under 30 years, HIV infection, low birth

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weight, as well as neonatal infections, all considered risk factors for anemia and neonatal mortality, are high [8]. However, there is no data on the prevalence of anemia and associated maternal-fetal or neonatal factors in Gabon. This study aimed to determine the prevalence and factors associated with anemia and the impact of malaria on its occurrence, in a population of hospitalized newborns at the University Hospital Center of Libreville (CHUL).

Patients and Methods

Type and period of study

This was a prospective analytical study conducted in the Neonatology and Neonatal Intensive Care Unit of CHUL from June to November 2015.

Study population

Newborns hospitalized in the neonatal intensive care and neonatology department of CHUL for suspected neonatal infection (NNI) were included if they met the following criteria: being less than 28-days-old and having obtained parental consent.

Data collection

The following data were collected on a standardized data collection form: age, sex, origin of the newborn, pregnancy and delivery history, gestational age in weeks of amenorrhea (SA), birth weight in grams (g), postnatal age at admission, reason for consultation, and clinical signs found during hospitalization. For mothers, data included age, educational level, hemoglobin electrophoresis results, parity, number of prenatal visits (PNV), and intermittent preventive treatment with sulfadoxine-pyrimethamine (IPT-SP).

Paraclinical evaluation

The paraclinical assessment was guided by the clinical examination and diagnostic hypotheses made upon the newborn's admission. It systematically included a complete blood count and a search for haemoparasites. No additional tests were requested for this observational study; however, results available in the patient's file were recorded. This included, among others, C-reactive protein (CRP) levels, stool culture, or blood cultures. These tests helped to classify neonatal infections (NNI).

Diagnosis of plasmodial infection

Thick blood smears were performed using the Lambarene method from peripheral blood samples [Planche 2001]. Briefly, 10 microliters of blood were placed and dehemoglobinized over a rectangular area of 18 mm long by 10 mm wide on a slide. After staining the slide in a 20% GIEMSA solution, the thick smear was examined under an immersion lens (×100) by microscopists. It was considered negative when no parasites were found after scanning 100 fields.

Diagnosis of anemia

The hemogram was performed using a Sysmex® hematology analyzer, which facilitated the diagnosis of anemia, leukocytosis, leukopenia, and thrombocytopenia. Hemoglobin (Hb) levels, white blood cell counts, and mean corpuscular volume (MCV) were classified according to normal values presented in Table 1.

Ethical considerations

Oral consent from the parents was obtained for the use of information contained in the patients' observation notebook. All data were anonymized. All exams performed were part of the routine evaluation of hospitalized newborns, so no additional biological sampling was prescribed.

Sample size calculation

The minimum number of patients to be included was estimated taking into account the frequency of plasmodial infection in newborns, in the absence of data on anemia and neonatal infections in this population [9]. A precision of 5% with a confidence level of 95%, an expected prevalence of neonatal malaria of 6.0%, and the following formula were used:

$$N = (Z^2 * p * (1\text{-}p)) / d^2$$

Where Z = 1.96; p = previous prevalence of congenital malaria = 6%; d = precision. Thus, a minimum of 87 newborns was required.

Statistical analysis

Data were collected in an Excel file with double entry. Statistical analysis was performed using Statview 5.0 software (SAS Institute Cary, USA). The Chi-square or Fisher's exact test was used to compare proportions. Quantitative data were compared using non-parametric tests and ANOVA. The significance threshold was set at 5% ($p < 0.05$) for all analyses.

Definitions

Pregnancy was considered well-followed from 4

Table 1: Normal hemogram values by age.

Age	Hemoglobin (g/dL)	White Blood Cells (elements/mm³)	Mean Corpuscular Volume (µ³)
Birth	16-20	12-25	99-110
Day 1-Day 7	16-20	9-14	95-110
Day 8-Day 15	13-18	9-14	95-110
Day 16-Day 28	11-15.1	8-13	93-105

Table 2: General characteristics of newborns and their mothers.

Characteristics	N	%
Male sex	53	53.0
Prematurity		
Extreme prematurity	4	4.0
Significant prematurity	23	23.0
Moderate prematurity	18	18.0
Birth weight		
Low birth weight	31	31.0
Very low birth weight	19	19.0
High birth weight	3	3.0
Origin		
Delivery room	52	52.0
Age at admission (days)		
Day 0	37	37.0
Day 1-Day 7	40	40.0
Day 8-Day 15	9	9.0
Day 16-Day 28	14	14.0
Maternal age at delivery (years)		
< 18	5	5.0
18-24	44	44.0
≥ 25	51	51.0
Parity		
Primiparous	26	26.0
Few births	49	49.0
Multiparous	25	25.0
Number of Prenatal Visits (PNV)		
None	10	10.0
≤ 3	50	50.0
≥ 4	40	40.0
Number of IPT-SP Doses		
None	26	26.0
1 dose	23	23.0
2 doses	28	28.0
3 doses	23	23.0

prenatal visits. Prematurity was defined for a gestational age (GA) less than 37 weeks of amenorrhea (SA) and divided into extreme prematurity (EP) for a GA less than 28 SA, great prematurity (GP) between 28 and 32 SA, and moderate prematurity (PM) between 33 and 36 SA. Birth weight was considered normal between 2500 and 4000 grams; low (LBW) between 1500 and 2499 g, very low (VLBW) when less than 1500 g, and high (HBW) for a value above 4000g.

Leukocytosis was defined for a white blood cell count > 25,000 elements/mm^3, thrombocytopenia for a platelet count < 100,000 elements/mm^3, and elevated CRP when it was above 6 mg/l.

A probable or confirmed bacterial infection was indicated by at least two of the following variables: white blood cell count > 25,000 elements/mm^3 and/or CRP ≥ 20 mg/L, meconium-stained amniotic fluid, premature rupture of membranes > 12h, positive blood and stool cultures if performed; a plasmodial infection for the presence of any parasite regardless of the parasitic density; and a non-bacterial, non-plasmodial infection in the absence of all the previous criteria.

Results

In total, 100 newborns were included; the sex ratio was 1.12. The majority were between 0 and seven-days-old (Table 2). The proportion of those with a low birth weight was 50%. The appearance of the amniotic fluid of 17 newborns was not reported, more than a third (37.0%) of the other 83 were born with abnormally colored amniotic fluid. Prematurity affected 45% of the patients. The median age of the mothers was 23 [22-25] years (Table 2). Nearly half of the mothers were paucipara, the majority had fewer than three (3) prenatal visits. Among the 34 mothers who had pathology during pregnancy, only 12 had infectious antecedents.

Hematological constants

MCV was known for 98 patients, it was low in 16 (16.3%) of them. The median MCV (104.1 [98.7-114.1] μ^3) and median hemoglobin level (15.4 [11.4-17.1] g/dL) were within normal limits. More than half of the newborns (52.0%) were anemic. The median CRP was elevated (8.9 [3-48] mg/l). Nearly a quarter of the newborns had leukocytosis (24.0%), and the median leukocyte count was 12000 [8945-19790] mg/L. Moreover, almost all the blood cultures performed were positive.

Prevalence of anemia

The prevalence of anemia was higher in newborns aged 8 to 15 days (88.9%) with a median hemoglobin level of 11.4 [9.9-12.2] g/dL in this age group. It was 54% at birth, 40% during the first week of life, and 57.1% between J16 and J28 ($p < 0.01$). Overall, the frequency of anemia was comparable among term newborns (51.1%) and preterm infants (55.8%) ($p = 0.60$). However, there were more moderately anemic preterm infants (66.7%) than non-anemic. No relationship was found between birth weight and the presence of anemia (Table 3).

Anemia in newborns

Anemia was not related to the mother's age but predominated in newborns of multiparous mothers (56%), in those whose mothers received only one dose of IPT (60.9%), and those whose mothers had fewer than three prenatal visits (Table 2).

Severity of anemia and associated factors

Newborns more frequently had mild (22%) or moderate (19%) anemia. The frequency of severe anemia was 11%. Moderate to severe anemia was less

Table 3: Frequency of anemia based on perinatal parameters.

Parameters	Anemic		Non-anemic		P-value
	N	%	N	%	
Prematurity					0.3
Term born	29	55.8	26	54.2	
Extremely premature	1	1.9	3	6.2	
Very premature	10	19.2	13	27.1	
Moderately premature	12	23.1	6	12.5	
Birth Weight					0.3
Very low birth weight	7	13.5	12	25	
Low birth weight	16	30.8	15	31.2	
Normal birth weight	28	53.8	19	39.6	
High birth weight	1	1.9	2	4.2	
Maternal Data					0.65
Age < 18 years	3	5.8	5	10.4	
Age 18-24 years	37	71.1	34	70.8	
Age > 25 years	12	23.1	8	16.7	
Parity					0.82
Primiparous	14	26.9	12	25	
Few births	24	46.2	25	52.1	
Multiparous	14	26.9	11	22.9	
Type of Pregnancy					0.88
Single	45	86.5	42	87.5	
Multiple	7	13.5	6	12.5	
Hemoglobin Electrophoresis					0.18
AA	52	100	45	93.7	
AS	-	-	2	4.2	
AC	-	-	1	2.1	
Number of IPT-SP Doses					0.5
None	12	23.1	14	29.2	
1 dose	14	26.9	9	18.7	
2 doses	13	25	15	31.2	
3 doses	13	25	10	20.8	
Number of PNV					0.2
None	3	5.8	7	14.6	
≤ 3 PNV	24	46.2	16	33.3	
≥ 4 PNV	25	54.8	25	52.1	

frequent during the first week of life (Table 4). Severe and moderate anemia was not found in newborns with VLBW. Children born to mothers who had not received any prenatal care frequently had moderate anemia. Nearly half of those born to mothers who had received three doses of IPT had severe anemia (n = 10/11, 43.59%).

Severity of anemia and type of NNI

Patients with an isolated or associated plasmodial infection were more frequently anemic (n = 11/18; 61.1%) than those without malaria (n = 39/83; 47.0%) (p = 0.045). They also more frequently had moderate to severe anemia (Table 5).

Discussion

Anemia is a major public health problem in developing countries, especially among children; approximately 43% of African children under the age of four are anemic [10]. In Gabon, two hospital-based studies over a ten-year period highlighted anemia prevalences ranging from 75% to 95% [11,12]. Among children under three months, anemia was found in 85% of cases. Its multifactorial origin has been demonstrated by several authors. Nutritional deficiencies, hemoglobinopathies, infections, including malaria, HIV infection, and intestinal parasites-all endemic in Gabon-are major etiologies [13-15]. Bacterial infections, prematurity and low birth

Table 4: Relationship between severity of anemia and neonatal and maternal factors.

Parameters	No anemia		Mild anemia		Moderate anemia		Severe anemia		*p*
	N	%	N	%	N	%	N	%	
Age of Newborns									< 0.01
At birth	17	45.9	8	21.6	7	18.9	5	13.5	
D1-D7	26	65.0	9	22.5	4	10.0	1	2.5	
D8-D15	1	11.1	2	22.2	4	44.4	2	22.2	
D16-D28	4	28.6	3	21.4	4	28.6	3	21.4	
Birth Weight									0.4
Very Low Birth Weight	4	80.0	1	20.0	0	0.00	0	0.0	
Low Birth Weight	23	51.2	6	13.3	10	22.2	6	13.3	
Normal Birth Weight	19	40.4	14	29.8	9	19.2	5	10.6	
High Birth Weight	2	66.7	1	33.3	0	0.0	0	0.0	
Number of Prenatal Visits (PNV)									0.3
None	7	70.0	1	10.0	2	20.0	0	0.0	
≤ 3 PNV	15	37.5	8	20.0	11	27.5	6	15.0	
≥ 4 PNV	26	52.0	13	26.0	6	12.0	5	10.0	
Number of IPT-SP Doses									0.4
None	14	53.9	5	19.2	5	19.2	2	7.6	
1 Dose	11	47.8	6	26.1	2	8.7	4	17.4	
2 Doses	14	50.0	7	25.0	5	17.9	2	7.1	
3 Doses	9	39.1	4	17.4	7	30.5	3	13.0	

Table 5: Relationship between severity of anemia and type of neonatal infection.

	No anemia		Mild anemia		Moderate anemia		Severe anemia		*p*
	N	%	N	%	N	%	N	%	
Bacterial Infection	30	50.0	10	16.7	13	21.7	7	11.6	0.3
NBNPI	11	57.9	6	31.6	1	5.2	1	5.3	
Plasmodial Infection	2	25.0	3	37.5	3	37.5	0	0.0	
Bacterial-Plasmodial Co-infection	5	38.5	3	23.1	2	15.3	3	23.1	

weight also contribute [7]. The relationship between all these risk factors for morbidity and neonatal mortality was sought using data from 100 children hospitalized at CHUL.

Prevalence of anemia

Half of the newborns hospitalized during the study period were anemic. This high frequency is comparable (52%) to those obtained at Yopougon Hospital (59%), in Mali (56%), and Morocco (53%) [16-18]. Lower prevalences were reported in Benin (46.3%) and Kenya (41.3%). However, these were obtained from children aged 0 to 6 months or 1 year [19,20]. In Libreville in 2000 and 2009, anemia was found in more than 50% of 0 to 5-month-old children consulting and/or hospitalized for febrile episodes at CHUL [12]. Neonatal anemia is therefore very common in Gabon.

Few African studies provide data on the severity of anemia in the neonatal period [6,21]. Mild to moderate anemia predominated (41%), however, severe anemia was found in 11% of cases, more frequently than between 2000 and 2009 in the same hospital (2 to 5.5%) [9,12]. Despite the small population size, this prevalence should be confirmed by studies involving a larger population. Tao in China reported frequencies of mild anemia and moderate to severe anemia of 49% and 50% respectively in a population of 104 newborns [22].

Anemia and age of newborn

Almost all children aged 8 to 15 days were anemic, and it was in this age group that the frequency of moderate to severe anemia was highest (66.6%). Data in the literature on the relationship between the age of the newborn and the frequency and severity of anemia are scarce. These results were obtained by considering the normal values of the hemogram according to the age of the newborn, which is not often the case in other studies. Amorissani in Ivory Coast reported a frequency of moderate to severe anemia of 20% in a population of premature newborns during their first week of life, prevalence lower than that found in newborns at birth (32.4%) [16]. This figure thus indicates that a third of

children with a neonatal infection should benefit from a blood transfusion. The cost of blood bags, transfusion risks that can exist in emergency situations, and the frequent lack of reagents as well as high mortality in the absence of transfusion, should lead health authorities to introduce and make erythropoietin administration accessible in neonatology services.

Factors associated with anemia

The relationship between maternal anemia and a low hemoglobin level in the newborn at birth could not be found. It should be noted that all mothers were under iron and folic acid supplementation. Likewise, IPT-SP intake and the number of SP doses were not associated with the presence of anemia in children. In Benin and Zimbabwe, neither the intake of mefloquine nor vitamin A supplementation reduced the risk of anemia in the newborn [2,19]. The quality and regularity of prenatal follow-up were not associated with the presence of anemia in the newborn. IPT-SP intake, which is associated with the number of PNV performed, would reduce the prevalence of maternal malaria and the occurrence of malaria-related anemia.

The analysis of parity showed that the frequency of anemic newborns from primiparous and multiparous mothers was comparable, with a slight predominance of anemia among multiparas (56%). Multiparity can induce anemia by reducing maternal iron reserves with each pregnancy due to blood loss during childbirth. A study published in Brazil found the frequency of anemia to be 72.4% among multiparas and 27% among primiparas [21]. Another study in Cameroon reports a predominance of anemia in children born from multiparas (57.1% vs. 40% among those from pauciparas) [23]. In Benin, primiparity was found as a risk factor for anemia in the first months of life [19].

Anemia and neonatal infections

Sepsis and lower respiratory infections mark the severity of neonatal infections that kill 2.7 million children each year, among which at least 50% die in the first week of life [24]. Bacterial infections were suspected and/or confirmed in nearly three-quarters of newborns, yet their relationship with anemia was not demonstrated.

Malaria thus remains the primary infectious etiology of neonatal anemia as reported by many authors [6,15,20,23,25]. Its impact on hemoglobin levels exists even in cases of bacterial co-infection. The median hemoglobin level was reduced by 2 g/dL in cases of co-infection. Data accumulated in the study by Church in London suggested that children with malaria are at risk of bacterial infection [26]. The prevalence of anemia was higher in children with a positive thick smear as observed in Burkina Faso (61.1% vs. 54%) [27]. In Libreville, the risk of being anemic in case of malaria in children aged 1 to 5 months was 1.55, that of developing severe malaria-related anemia was 8.55. This risk of anemia gradually decreased with age up to 47 months [9].

Conclusion

These results obtained in a population of newborns with several risk factors for morbidity and mortality has highlighted the importance of anemia and malaria in the first days of life. Subsequent longitudinal studies with a larger sample of mother-child pairs and followed from the first trimester of pregnancy to the end of the first year of life will deepen the knowledge acquired and develop strategies for managing and controlling risk factors for neonatal anemia.

Contribution of Authors

All authors contributed to the writing of this manuscript, read and approved the final version.

Conflict of Interest

The authors declare no conflict of interest.

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