International Journal of Life Sciences, Biotechnology and Pharma Research Vol. 13, No. 10, October 2024 Online ISSN: 2250-3137
Print ISSN: 2977-0122

DOI: 10.69605/ijlbpr_13.10.2024.83

Original Research

Clinic Demographic and Hematological profile of Nutritional Anemia in children

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Received Date: 16 August 2024 Accepted Date: 13 September 2024

Abstract
Aim: To study clinico Demographic and Hematological profile of Nutritional Anemia in children (6 months to 15 years of age).
Material and methods: The present prospective observational study was conducted at Jaipur National University Institute for Medical Science and Research Centre for a period of 1 year among 100 children in the age group of 6 months to 15 years with haemoglobin levels less than the cut-off values for the age, as per WHO guidelines. The following investigations were done in all cases – complete blood count, red cell indices, peripheral smear study, reticulocyte count, RDW, ESR, CRP, serum ferritin, iron indices, vitamin B12 and folic acid assay.
Results: The majority (81.0%) of subjects were between 1-10 years. Pale skin was the most common symptom, observed in 48 individuals (48.0%). Irritability was reported by 31 participants (31.0%), and lethargy was noted in 25 subjects (25.0%). Pallor was the most prevalent sign, affecting all participants 100 (100%). The majority 53 subjects (53.0%) had normocytic and normochromic red blood cells (NNA), 38 individuals (38.0%) had microcytic and hypochromic red blood cells (MHA) and 9 participants (9.0%) exhibited normocytic and hypochromic red blood cells (NHA). Measurements such as mean serum iron, TIBC, transferrin saturation, serum transferrin, serum ferritin levels, vitamin B12 and folic acid concentrations results showed no statistically significant differences ($p > 0.05$) in these values across groups with microcytic and hypochromic, normocytic and hypochromic, or normocytic and normochromic red blood cells.
Conclusion: In our study, we concluded that nutritional anemias emerged as the common type, with iron deficiency anemia being the most prevalent. The majority of children examined in this study exhibited signs of malnutrition and hailed from lower socioeconomic backgrounds.
Keywords: Clinical, Demographic, Hematological, Nutritional Anemia

Introduction:
Anemia is characterized by a reduced concentration of blood hemoglobin, indicating insufficient red blood cells or their oxygen-carrying capacity to meet the body's needs. These needs vary depending on factors such as age, sex, altitude, smoking habits, and pregnancy status.[1,2] Clinical manifestations of anemia include pallor, fatigue, irritability, and reduced exercise tolerance. If left untreated, it can progress to symptoms like rapid breathing, increased heart rate, shortness of breath during exertion, weakness, cardiac enlargement, and heart failure.[3]
According to data from the Comprehensive National Nutrition Survey (CNNS) conducted between 2016 and 2018, the prevalence of anemia was 40.6% among children aged 1-4 years, 23.5% among those aged 5-9 years, and 28.4% among those aged 10-19 years.[4] The World Health Organization (WHO) estimates that globally, 42% of children under the age of five are anemic, while in India, the prevalence of anemia among children aged 6-59 months was 53.4% in 2019.[5]
Despite efforts such as the national nutritional anemia control program and India's overall economic growth, the prevalence of anemia has increased,[6] with the most common causes being nutritional deficiencies, particularly iron, folic acid, and vitamin B12 deficiencies, along with vitamin A deficiencies.

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Hemoglobinopathies, and infectious illnesses such as malaria, TB, HIV, and other parasite infections are additional risks[7]. Risk factors commonly cited in the literature for anemia include low family income, limited maternal education, lack of access to healthcare services, poor sanitation, and inadequate dietary iron intake.[8-9]

Additionally, factors such as low socioeconomic status, early consumption of cow's milk before six months of age, low birth weight, and prematurity pose additional risks for newborns. Anemia significantly impacts a child's growth, development, overall well-being, and academic performance. It often diminishes children's appetite, leading to nutritional deficiencies and initiating a harmful cycle that exacerbates the condition. Notably, anemia is a significant comorbidity among hospitalized children, potentially prolonging hospital stays, increasing the likelihood of complications, and necessitating blood transfusions.[10]

Anemias can be categorized based on morphology or underlying physiology. Morphologically, they are classified as microcytic, normocytic, or macrocytic, determined by red cell size (MCV – mean corpuscular volume), often discernible through peripheral smear examination. Physiologically, anemias are distinguished by increased destruction or decreased production of red blood cells. However, these categories are not always mutually exclusive. Reticulocyte count aids in differentiating between the two, as a low or normal count suggests inadequate bone marrow response or ineffective erythropoiesis, while an increased count indicates normal bone marrow activity in response to red blood cell destruction (hemolysis), loss (bleeding), or sequestration.[11]

There remains insufficient information about factors influencing anemia among young children. Furthermore, there are no existing reports on the prevalence of anemia among children in the Jaipur district. Therefore, we performed a study on clinical, demographic and hematological profile of nutritional anemia in children 6 months to 15 years of age.

Aim And Objectives

Aim: To study clinico Demographic and Hematological profile of Nutritional Anemia in children (6 months to 15 years of age).

Objective: To assess the correlation of Red Cell Indices, Peripheral blood film levels of serum iron, Folic acid and vitamin B12 with the clinical profile of patients with Anemia.

Material And Methods: The present prospective observational study was conducted at Jaipur National University Institute for Medical Science and Research Centre for a period of 1 year among 100 hospitalized children.

Inclusion criteria: Children in the age group of 6 months to 15 years with haemoglobin levels less than the cut-off values for the age, as per WHO guidelines, were included in the study. Out of the 100 children, those with iron deficiency anemia and vitamin B12 were included in this study.

Exclusion criteria:
- Chronic diarrhea
- Children with renal disease
- Children with cyanotic heart disease
- Children with endocrine diseases
- Children with Significant blood loss
- Sickle Cell Disease
- All types of Leukemia
- IBD
- Lymphoma
- Haemolytic Anemia

Methods: A total of 100 children in the age group of 6 months to 15 years were studied and included in the study after obtaining informed written consent from their parents. The parents of the enrolled youngsters gave their informed consent. The criteria for diagnosing anemia were hemoglobin values less than 11 g/dL in children aged 6 months to 59 months and less than 11.5 g/dL in children aged 5 years to 13 years. 2 mL of venous blood was taken under aseptic conditions and immediately submitted to the lab for hematological testing in a vacutainer that contained ethylene diamine tetra acetate (EDTA). The following investigations were done in all cases – complete blood count, red cell indices, peripheral smear study, reticulocyte count, RDW, ESR, CRP, serum ferritin, iron indices, vitamin B12 and folic acid assay.

Statistical Analysis: The statistical methods used for quantitative data were descriptive statistics presented by N, Mean, Standard Deviation, and Range. For qualitative data, frequency count, N and percentage were put in tabular columns. To analyze the data, appropriate statistical tests were applied. To compare the differences between variances in the subjects, one-way ANOVA was used. To find the correlation between hemoglobin, red cell indices & iron profile, Pearson's correlation coefficient was used. $P<0.05$ was considered to be statistically significant.

Results: In the present study total of 100 subjects or patients up to 15 years were included. The majority (81.0%) of subjects were between 1-10 years 16.0% were between 11-15 years and only 3.0% of subjects were less than 1 year of age (figure 1). Out of 100, 52 (52.0%) subjects were males and 48 (48.0%) were female.

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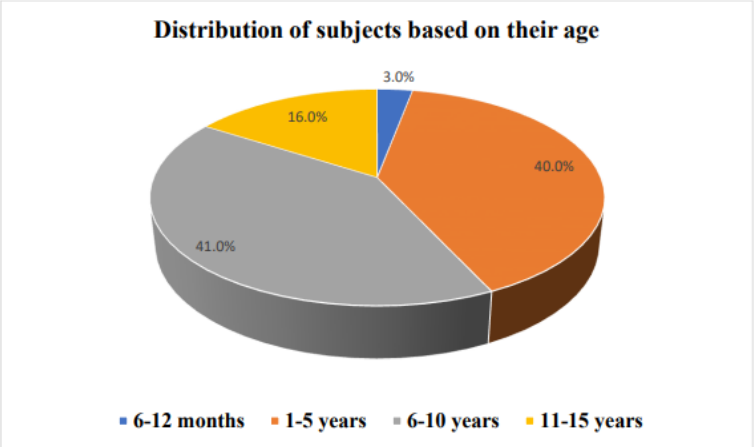

Figure 1: Distribution of the studied subjects based on age

24 individuals (24.0%) had a history of PICA, and 23 (23.0%) reported the passage of worms. Behavioural problems were noted in 9 subjects (9.0%), while 8 participants (8.0%) experienced frequent infections. Pale skin was the most common symptom, observed in 48 individuals (48.0%). Irritability was reported by 31 participants (31.0%), and lethargy was noted in 25 subjects (25.0%). Less common symptoms included cold hands and feet, experienced by 4 participants (4.0%), and abnormally rapid breathing, which was noted in 2 subjects (2.0%) as shown in figure 2.

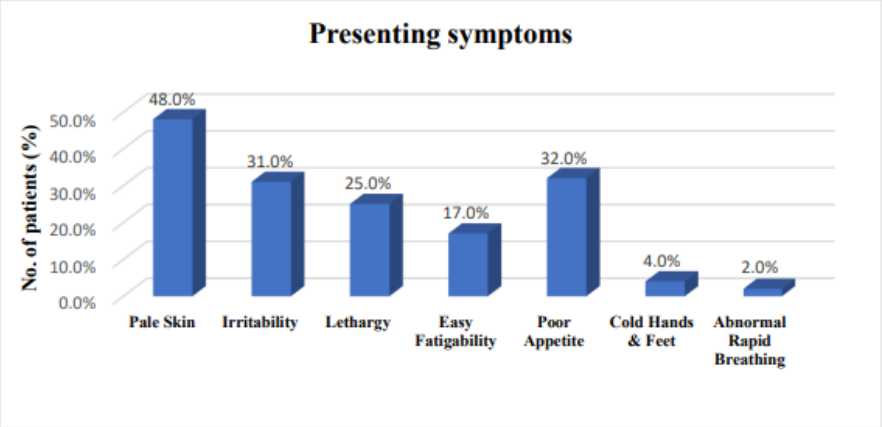

Figure 2: Presenting symptoms among the study subjects

Pallor was the most common sign, affecting all participants 100 (100%). Icterus (yellowing of the skin), lymphadenopathy (swollen lymph nodes), and edema (fluid buildup) were identified in a smaller percentage of subjects 8 (8.0%), 3 (3.0%), and 6 (6.0%), respectively (table 1).

Table 1: Presenting signs among the study subjects

Signs	Frequency (n=100)	Percentage
Pallor	100	100%
Icterus	8	8.0%
Lymphadenopathy	3	3.0%
Edema	6	6.0%
Clubbing	2	2.0%
Cyanosis	1	1.0%
Koilonchyia	5	5.0%
Glossitis	1	1.0%
Chelitis	12	12.0%

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Out of 100 participants, the majority, 53 subjects (53.0%), had normocytic and normochromic red blood cells, 38 individuals (38.0%) had microcytic and hypochromic red blood cells and 9 participants (9.0%) exhibited normocytic and hypochromic red blood cells (figure 3).

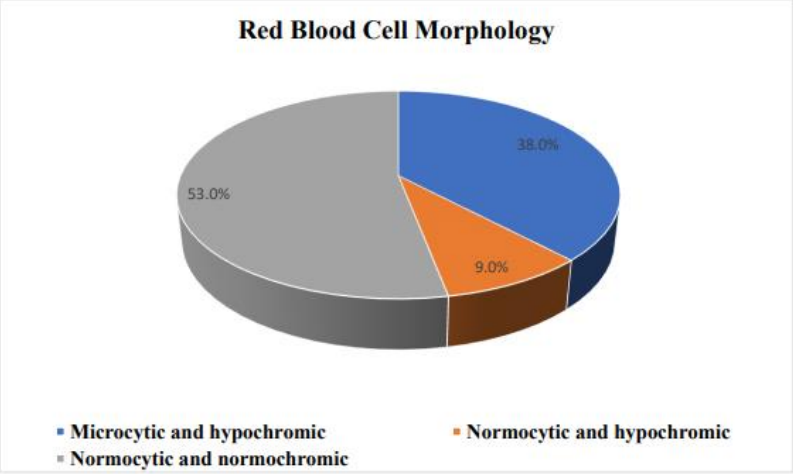

Figure 3: Frequency of red blood cell morphology among study subjects

Table 2: Comparison of Haematological indices with red blood cell morphology

Peripheral Smear		Hb	TLC (cells/cu.mm)	RBC (millions/cu.mm)	PCV (%)	MCV (fL)	MCH (pg)	MCHC (%)
Microcytic and hypochromic	Mean	8.3	10326.3	4.5	27.0	59.0	19.8	30.8
	SD	1.2	4579	0.6	3.7	11.7	6.7	2.1
Normocytic and normochromic	Mean	9.8	10611.1	4.6	32.4	69.9	21.8	37.3
	SD	0.5	3797	0.4	3.9	5.2	1.1	16.8
Normocytic and hypochromic	Mean	9.8	11181.1	4.3	31.8	71.6	23.0	31.3
	SD	0.9	5773	0.5	3.8	11.9	2.8	1.9
F value		24.5	0.306	0.980	18.6	13.8	4.95	5.87
p-value		**<0.001**	0.737	0.379	**<0.001**	**<0.001**	**0.009**	**0.004**

*: statistically significant

Subjects with microcytic and hypochromic red blood cells (smaller and paler than normal) had a statistically significant ($p<0.01$) lower mean hemoglobin (Hb) and hematocrit (PCV) compared to the other groups. MCV (mean corpuscular volume) was also significantly lower ($p<0.01$) in this group, confirming their microcytic nature. Interestingly, there were no statistically significant differences in TLC (total leukocyte count) or RBC (red blood cell count) between the groups. While both normocytic and normochromic (normal size and color) and normocytic and hypochromic groups had similar MCV, the latter group had a significantly lower MCH (mean corpuscular hemoglobin, $p=0.009$) indicating less hemoglobin per red blood cell despite being normal sized. Overall, the data suggests a strong correlation between red blood cell morphology and certain hematological indices, particularly Hb, PCV, MCV, and MCH.

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Table 3: Comparison of Haematological indices with red blood cell morphology

Peripheral Smear		RDW (%)	Platelet Count	Reticulocyte Count
Microcytic and hypochromic	Mean	17.6	3.7	0.91
	SD	4.3	1.1	0.81
Normocytic and hypochromic	Mean	14.5	3.0	0.47
	SD	2.1	1.2	0.22
Normocytic and normochromic	Mean	15.0	4.8	0.86
	SD	3.4	8.7	0.36
F value		6.06	0.54	2.1
p-value		**0.003**	0.581	0.122

*: statistically significant

Individuals with microcytic and hypochromic red blood cells exhibited a higher mean red cell distribution width (RDW) of 17.6% compared to those with normocytic and hypochromic (14.5%) or normocytic and normochromic cells (15.0%). This difference was statistically significant ($p<0.05$). Similarly, the mean reticulocyte counts for microcytic and hypochromic (0.91) and normocytic and normochromic (0.86) groups did not show a statistically significant difference ($p>0.05$) as shown in table 3.

Individuals with microcytic and hypochromic red blood cells had a mean serum iron concentration of 41.1 ug/dl, a total iron-binding capacity (TIBC) of 362.9 umol/L, transferrin saturation of 13.3%, a serum transferrin level of 404.0 mg/dl, and a serum ferritin concentration of 48.6 ug/L, with corresponding standard deviations (SD) of 37.8 ug/dl, 127 umol/L, 18.4%, 99.3 mg/dl, and 56.5 ug/L, respectively. However, none of these differences were statistically significant, as indicated by the ($p>0.05$). Similar findings were observed for individuals with normocytic and hypochromic or normocytic and normochromic red blood cells. The F values ranged from 0.019 to 1.49, with corresponding p-values ranging from 0.229 to 0.981, suggesting no statistically significant differences across the groups. Individuals with microcytic and hypochromic red blood cells had a mean vitamin B12 concentration of 460.2 pg/ml and a mean folic acid concentration of 13.4 ng/ml, with standard deviations (SD) of 177 pg/ml and 4.8 ng/ml, respectively. Those with normocytic and hypochromic or normocytic and normochromic red blood cells exhibited different mean concentrations for both vitamin B12 and folic acid, but these differences were not statistically significant, as indicated by the ($p>0.05$) as shown in table 4.

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Table 4: Comparison of Vit. B12 (pg/ml), Folic acid (ng/ml) with red blood cell morphology

Peripheral Smear		Vit. B12 (pg/ml)	Folic acid (ng/ml)
Microcytic and hypochromic	Mean	460.2	13.4
	SD	177	4.8
Normocytic and hypochromic	Mean	361.7	11.9
	SD	116	4.3
Normocytic and normochromic	Mean	449.7	15.9
	SD	212	14.1
F value		0.967	0.883
p-value		0.384	0.417

Discussion:

Iron deficiency anemia represents the most prevalent type of nutritional deficiency globally, contributing significantly to illness burden, diminished productivity, heightened mortality, and morbidity rates. Even within developed nations, iron deficiency, with or without anemia, persists among infants, toddlers, adolescent females, and women of childbearing age. It continues to stand as the predominant hematologic ailment affecting infants and children worldwide.[12] Considering the significant long-term repercussions and widespread occurrence of iron deficiency, its prevention in early childhood emerges as a crucial public health concern. Hematology clinics frequently encounter patients with hemoglobinopathy syndromes, with thalassemia emerging as the most prevalent disorder in India.[6]

In our study, of 100 children, the majority were in the age group ranging from 6 to 10 years (41.0%) followed by 1 to 5 years (40.0%), 11 to 15 years (16.0%), and the least were below 1 year of age (3.0%). There was a slight male predominance 52 (52.0%) than females (48.0%). Our findings were comparable to the findings of Kanth RK et al[13] who reported that data was collected from 100 children aged between 1-12 years The mean age was 5.54±3.24. The gender distribution, 54 boys and 46 girls were in the study population. In the study conducted by Ramawat P et al[10], Among the children surveyed, 8.4% were under six months old, 30.2% were aged between six months and one year, 40.7% were aged between one and five years, 13.2% were aged between five and ten years, and 8% were aged between ten and fourteen years. In the study conducted by Ramawat P et al[10] 51.3% were boys and 48.6% were girls. Reddy KS et al19 reported that 61.1% cases were male and 38.9% were females whereas the majority of cases were in age group 6 to 10 years (36.7%) followed by 1 to 5 years (21.1%) and 11 to 17 years (17.8%). In contrast to the present study Mishra G and Dwivedi R[2] reported that among the 300 patients studied, comprising individuals aged one to 18 years, 148 (49.3%) were males, and 152 (50.7%) were females.

In our study, pallor was the most prevalent sign, affecting all participants 100 (100%). Icterus (yellowing of the skin), lymphadenopathy (swollen lymph nodes), and edema (fluid buildup) were identified in a smaller percentage of subjects 8 (8.0%), 3 (3.0%), and 6 (6.0%), respectively. Clubbing (fingertip enlargement), cyanosis (bluish skin), and koilonychia (spoon-shaped nails) were even less frequent 2 (2.0%), 1 (1.0%), and 5 (5.0%), respectively. Glossitis (inflamed tongue) and cheilitis (inflamed lips) were only observed in a single subject each 1 (1.0%). Our findings were in accordance with the findings of Kanth RK et al[13] who reported that Cardiovascular symptoms such as tachycardia were prevalent in 96% of cases, with ejection systolic murmurs present in 18% and hemic murmurs in 2%. Organomegaly, including hepatomegaly, was observed in 18% of cases, splenomegaly in 12%, and hepatosplenomegaly in 14%. Other general signs included Koilonychia in 8% of cases, rash in 6%, bone pain in 6%, and petechiae in 2%. Among the common symptoms noted, the majority (56%) experienced easy fatigability, followed by pallor in 36%, generalized weakness and irritability in 20% each, and 22% reported PICA.

Additional symptoms included lethargy in 14%, seizures in 8%, and icterus, edema, and palpitations each reported in 6% of cases. According to Reddy KS

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et al[14] the clinical signs and symptoms were pallor (91.1%) followed by Weakness and fatigability (78.8%), fever (54.4%), icterus (16.6%), and shortness of breath (4.4%) whereas cough, history of PICA, splenomegaly, petechiae, vomiting, koilonychia, hyperpigmentation, tremors were major symptoms. Upadhyay P and Kanetkar SR[15] reported that pallor was the commonest sign noted in all cases (100%) followed by tachycardia (84.09%), signs of malnutrition (80.68%), Hemic murmur (44.88%), Edema (23.87%), splenomegaly (16.47%), Hepatomegaly (9.09%), Lymphadenopathy (6.81%).

Out of 100 participants, the majority, 53 subjects (53.0%), had normocytic and normochromic red blood cells, 38 individuals (38.0%) had microcytic and hypochromic red blood cells and 9 participants (9.0%) exhibited normocytic and hypochromic red blood cells. Our findings align with previous studies. Kanth RK et al[13] reported various peripheral smear appearances of RBCs, with microcytic and hypochromic appearance in the majority (52.0%), followed by normocytic hypochromic (24%), normocytic normochromic (20.0%), and macrocytic (4%). They also noted sickling in 6.0% and malaria parasite in 4.0% of cases. Similarly, Reddy KS et al[14] observed microcytic hypochromic picture in 56%, normocytic hypochromic anaemia in 18.8%, normocytic normochromic in 12.2%, dimorphic anaemia in 7.7%, and macrocytic in 4.4% cases. However, Zhao A et al[16], found microcytic hypochromic picture in 49%, dimorphic in 24%, normocytic normochromic in 22%, and macrocytic in 4% cases. Chhabra et al[17], demonstrated that microcytic hypochromic picture was common across all age groups and significantly associated with iron deficiency anemia. Subhani PS and Prasad KN[18] reported that most children had microcytic hypochromic or iron deficiency anemia, while normocytic normochromic anemia was also common. Upadhyay P and Kanetkar SR[15] reported various types of anemia, with microcytic hypochromic being the most prevalent (47.73%), followed by normocytic normochromic (21.59%), normocytic hypochromic (13.07%), macrocytic hypochromic (10.79%), and macrocytic normochromic (6.82%). Similarly, Kumar A et al[19] found microcytic hypochromic anemia to be the most prevalent, followed by macrocytic, dimorphic, and normocytic normochromic anemias. Additionally, Maiti D et al[20] reported microcytic hypochromic anemia (71.3%) as the most common morphological type across all age groups, with macrocytic anemia being the least common.

Overall, the data suggests a strong correlation between red blood cell morphology and certain hematological indices, particularly Hb, PCV, MCV, and MCH. Our findings were similar to Subhani PS and Prasad KN[18] who reported that the average MCHC in the MH group was 27.8±5.3g/dl, while the mean MCV in the NN group was 34.9±7.8 g/dl. There was a notable difference in MCV, MCH, and MCHC between the MH and NN groups. This was consistent with the results of Ramana Sastry CPV[21], Maheshwari BK et al[22] and Upadhyay P and Kanetkar SR[15], Maiti D et al[20] reported that the average hemoglobin level was notably lower (P = 0.004) in the microcytic hypochromic anemia group compared to the normocytic group. However, there were no significant differences observed in the mean MCV and mean MCHC values between the normocytic normochromic and microcytic hypochromic groups. On the other hand, the mean RDW was significantly higher (P = 0.005) in the micocytic hypochromic group compared to the normocytic group.

In the present study, individuals with microcytic and hypochromic red blood cells exhibited a higher mean red cell distribution width (RDW) of 17.6% compared to those with normocytic and hypochromic (14.5%) or normocytic and normochromic cells (15.0%). This difference was statistically significant (p0.05). Similarly, the mean reticulocyte counts for microcytic and hypochromic (0.91) and normocytic and normochromic (0.86) groups did not show a statistically significant difference (p>0.05). Our findings were supported by Mishra G and Dwivedi R[2] who reported that the variance in Red Cell Distribution Width (RDW) between iron deficiency and vitamin B12 deficiency anemia was notable, with a p-value of less than 0.001. The mean RDW value for Vitamin B12 deficiency anemia stood at 31.1±10.1%, significantly higher than that of IDA patients, which was recorded at 17.2±3.5%. Aulakh R et al[23], conducted a study involving 151 children with microcytic anemia (MCV<0.001).

Limitations: The present study had limitations as it was a single-center, hospital-based study conducted in Jaipur. While the hospital attracts a broad spectrum of patients from the state, the findings may not be generalized to the aggregate of Rajasthan or the country due to relatively smaller sample size.

Strengths: This study stands as a robust indicator of the prevalence of anemia in urban healthcare facilities. Its findings are poised to enlighten clinicians and primary healthcare practitioners on the clinico-epidemiological landscape of anemia, aiding in more accurate diagnosis, treatment, and the formulation of preventive strategies moving forward.

Conclusion: In our study, we concluded that nutritional anemias emerged as the common type, with iron deficiency anemia being the most prevalent. The majority of children examined in this study exhibited signs of malnutrition and hailed from lower socioeconomic backgrounds. Proposed interventions encompass early detection, efficient management, and treatment of anemia. We advocate for heightened awareness regarding sanitation practices to mitigate infections and nutritional guidance for parents regarding the consumption of iron-rich foods and iron supplementation to prevent anemia among young

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children, particularly those from low-income and socioeconomically disadvantaged communities. Early screenings followed by diagnostic assessments facilitate timely diagnosis and appropriate treatment.

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