

Association of folic acid and vitamin B12 deficiency in children with iron deficiency anaemia

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Abstract

Objective: To find out the association of folic acid and vitamin B₁₂ deficiency in patients with iron deficiency anaemia in Indian children admitted to a tertiary hospital. **Design:** Observational cross sectional study. **Setting:** Tertiary-level hospital from October 2010-September 2011, for a period of one year. **Participants:** All children aged between 6 months to 13 years, admitted in paediatric ward of this hospital for any complaints, were evaluated for anaemia. All patients with Hemoglobin levels less than the WHO cut off levels for anaemia were included in the study. Those who were sick and those who were on drugs, other conditions causing bone marrow suppression were excluded from the study. **Methods:** Serum iron, serum iron binding capacity, serum ferritin, serum vitamin B₁₂, and serum folic acid analyses were conducted with autoanalyzers using commercial kits. **Results:** Out of 975 children who were admitted during the above period, 100 (34 females and 66 males) anemic children were included in the study. 2 patients were excluded as one child was diagnosed with Thalassemia minor and another patient was diagnosed as acute lymphoblastic Leukemia. Hence 98 children were taken up for study. Iron and Folic acid Deficiency observed in 6.12 % (6/98), Iron and vitamin B₁₂ Deficiency in 3.06 % (3/98), Folic acid and vitamin B₁₂ Deficiency in 1.02 % (1/98), Iron, Folic acid and vitamin B₁₂ Deficiency in 1.02 % (1/98) of patients. **Conclusions:** Iron deficiency anaemia coexist with vitamin B₁₂ and folate deficiency, but their association with iron deficiency anemia was not significant in our study.

Keywords: Anaemia, Folic acid deficiency, Iron deficiency, Vitamin B₁₂ deficiency.

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Introduction

Anaemia is a major public health problem all over the world especially in developing countries. The third National Family Health Survey (NFHS – 3) (2005-06) found that the prevalence of anaemia among under 5 children approaches 70% even though there is a national programme to control anaemia for many years [1]. The irreparable damage that anemia in childhood can cause particularly to the development of a young child on one hand and the knowledge and mechanism available for its control on the other, makes this silent morbidity completely unacceptable in modern times where we strive for millennium development Goal 4 [2]. Out of all the causes, nutritional anaemias account for the major cause of anaemias. Pregnant women, infants, young children and adolescents are at a higher

risk of nutritional anaemia as they have a high demand of nutrients such as iron, folic acid, vitamin B₁₂ and other nutrients. Nutritional anaemia is seen in almost one billion people all over the world and commonest being iron deficiency anemia [3]. Anaemia in children results in impaired cognitive performance, behavioral and language development and scholastic achievement. Anaemia is also associated with increased mortality and morbidity from infectious diseases [4]. Other causes of anemia include chronic infections, particularly malaria, hereditary hemoglobinopathies and folic acid deficiency. It is worth noting that multiple causes of anemia can coexist in an individual or in a population and contribute to the severity of the anemia.

The term 'nutritional anemia' encompasses all pathological conditions in which the blood hemoglobin concentration drops to an abnormally low level, due to a

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deficiency in one or several nutrients. The main nutrients involved in the synthesis of hemoglobin are iron, folic acid, and vitamin B₁₂. In public health terms, iron deficiency is by far the first cause of nutritional anemia worldwide. Folic acid deficiency is less widespread and is often observed with iron deficiency. Vitamin B₁₂ deficiency is far rarer and it occurs mainly in vegetarians [5]. There are various studies done to find out the prevalence of iron deficiency anaemias in paediatric population in India and other developing countries [6,7,8,9]. But there is paucity of studies using laboratory measurements to find out the exact cause of nutritional anaemia and to find out the association of folic acid and vitamin B₁₂ deficiency in patients with iron deficiency anaemia in Indian population [10]. Hence this study was done with the objective to study the association of folic acid and vitamin B₁₂ deficiency in patients with iron deficiency anaemia in Indian children admitted to a tertiary hospital.

Materials and Methods

This study is an observational cross sectional study, conducted in paediatric ward of Vydehi Institute of Medical Sciences and Research Centre, a tertiary care hospital in southern India from October 2010-September 2011 for a period of one year. Informed consent was taken from the care takers of the patients for this study. Permission was taken from the Ethics committee of the hospital for this study. All patients admitted in paediatric ward of this hospital for any complaints were evaluated for anaemia. Those patients who were less than 6 months of age, sick patients and

those patients who were on drugs, other conditions causing bone marrow suppression were excluded from the study. All other patients with Hemoglobin levels less than the WHO cut off levels for anaemia were included in the study. WHO Expert group proposed that anaemia should be considered to exist when Hemoglobin is below the following levels in venous blood. 6 months to 6 years 11 gm / dl, less than 12 g/dL for girls from 6 to 18 yrs and boys from 6 to 14 years, and less than 13 g/dl for boys from 15 to 18 yrs of age [11].

In those patients found to have anaemia, the following laboratory investigations were done like mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), serum ferritin, serum iron, total iron binding capacity (TIBC), serum transferrin (TRF), serum folate assay and serum vitamin B₁₂ assay. 5 ml of blood was collected in a EDTA tube for the study. Serum hemoglobin was estimated by spectrophotometric method by automated machine, serum ferritin by Chemiluminescent immunoassay, serum iron by Ferrozine_{no} deproteinization, TIBC by ion exchange resin ferrozine method, serum transferrin (TRF) by rate nephelometry, serum folate assay and vitamin B₁₂ assay by Chemiluminescent immunoassay. In case they were suffering from acute infective or inflammatory conditions [12] the above investigations were done after 3 afebrile days. The normal values for various study parameters in this study were considered from Nelson Text Book of Pediatrics [13].

Results

100 patients with anaemia as defined by WHO, admitted in the paediatric ward from the period from October 2010–September 2011, were included in the study. The age of the patients ranged from 6 months to 13 years (Table 1). There were 66 (66 %) boys and 34 (34 %) girls in the study population. 2 patients were excluded as one child was diagnosed with Thalassemia minor and another patient was diagnosed as Acute lymphoblastic Leukemia. The haemoglobin levels of the study subjects are mentioned as per Table 2, which shows that 69% had Hb levels between 10 -12 g/dl, 15% had 9-10g/dl, 6% between 8-9 g/dl, 5% between 7-8g/dl and 3% less than 7 g/dl. Levels of blood folate, serum B₁₂, serum ferritin, serum Iron, TIBC and serum transferrin levels of patients studied are mentioned as per Table 3 which shows 16.3% had folic acid deficiency, 7.1% had B₁₂deficiency and 43.8% had iron deficiency. Incidence of Iron deficiency anemia in our study was 54.1%, Folic Acid deficiency was seen in 16.3% and Vitamin B₁₂deficiency anemia was seen in 13.2 % (Table 4).

Table-1: Age wise distribution of Patients under study.

Patients age in years	<1	1-2	2-5	5-10	>10
No. of cases	10	21	20	25	22

Mean Age ± SD: 6.03±4.28

Table-2: Hemoglobin levels of patients studied.

Haemoglobin levels in gms/dl	<7	7-8	8-9	9-10	10-12
No. of cases	3	5	6	15	69

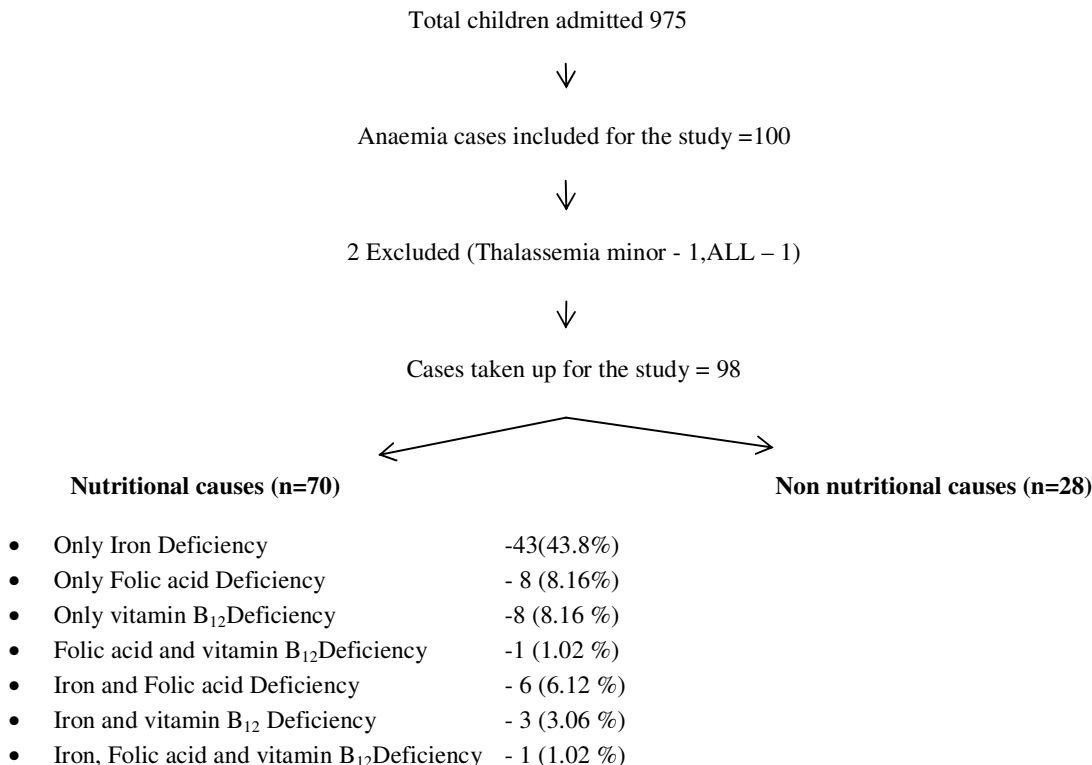


Table-3: Levels of Blood folate, serum B12 serum ferritin, Serum Iron, TIBC and Serum transferrin levels of patients studied.

Hematological parameters	No. of subjects	Range	Distribution			
Blood Folate pg/ml	98	2.4 - > 20	< 4 16 (16.3)	5- 10 60(61.2)	10 – 15 20(20.4)	>15 2 (2.04)
Serum B12 ng/ml	98	82 - >1500	<140 7 (7.1)	140 – 202 6 (6.1)	203 – 800 83 (84.7)	>800 2 (2.04)
Serum Iron mcg / dl	98	7 – 284	<22 29 (29.6)	22 – 50 36 (36.7)	>50 33 (33.7.1)	
TIBC mcg/dl	98	187 – 636	<250 8 (8.2)	250 -400 46 (46.7)	>400 44 (44.9)	
Serum Ferritin mcg / l	98	2.2-711	<7 13 (13.3)	7-140 74 (75.5)	>140 11(11.2)	
Serum Transferrin mg/dl	75	17 – 578	<95 2 (2.7)	95-385 58 (77.3)	>385 15 (20)	

(Numbers in parenthesis are the percentages)

Serum transferrin could be done in only 75 patients because of the non-availability of kits during the study period.

Table-4: Incidence of Iron deficiency, Folic Acid levels, and Vitamin B12 levels of patients studied.

Iron deficiency	No. of patients	Percent
Yes	53	54.1
No	45	45.9
Total	98	100.0
Folic Acid		
<4	16	16.3
4-15	80	81.6
>15	2	2.04
Total	98	100.0
Vitamin B₁₂		
<203	13	13.2
203-800	83	84.7
>800	2	2.04
Total	98	100.0

Discussion

The results of the present study showed that out of 98 children with anaemia, nutritional anaemia was noted in 71.4% (70/98) of the children. Pure iron deficiency anaemia (IDA) was detected in 43.8 per cent (43/98) of anaemic children. Vitamin B₁₂ deficiency alone or in combination with iron was diagnosed in 8.16% (8/98) and 3.06% (3/98) anaemic children respectively. Similarly folate deficiency alone or in combination with IDA was 8.16% and 6.1 % (6/98) respectively. 1% (1/98) had all, iron, folic acid and Vitamin B₁₂ deficiency. Pasricha et al [14] in 2011, studied the micronutrients such as vitamin B₁₂, folate, iron and vitamin A concentrations of 396 children in the age group of 12 – 23 months in rural Karnataka in south India. They found that 65.6% had at least one micronutrient deficiency and those children between 1-2 years who are breast feeding should be targeted during micronutrient supplementation programs. Ahmed F et al [15] in 2008 studied the prevalence of selected micronutrient deficiencies amongst anaemic adolescent school girls in rural Bangladesh, and they found that 28% of the girls had depleted iron stores, 25% had folic acid deficiency, 89% had vitamin B₂ and 7% had vitamin B₁₂ deficiencies.

They concluded that there is coexistence of micronutrient deficiencies among anaemic adolescent girls in rural Bangladesh, although they do not suffer from energy deficiency. Of all micronutrients only iron and vitamin B₂ concentrations were found to be related to the Hb concentration. Gadowsky SLet al [16]

estimated the prevalence of biochemical iron, folate, and vitamin B₁₂ depletion among a group of Canadian pregnant adolescents. They found that 22% of the pregnant adolescents had anaemia, 78% had depleted iron stores and 25% had plasma vitamin B₁₂ values in the sub-optimal range. Metz J [17] reviewed the prevalence of anemia based on biochemical evidence. He found that the overall contribution of vitamin B₁₂ deficiency to the global burden of anemia was not significant, except in women and their infants and children in vegetarian communities.

He also found that it was unlikely that folate deficiency makes a major contribution to the burden of anemia in developing countries. Iron-deficiency anemia may coexist with vitamin B₁₂ and especially folate deficiency, and may confound the hematological features of the vitamin deficiencies whose prevalence would then be underestimated. Van der Westhuyzen Jet al [18] investigated the prevalence of anaemia and deficiencies of iron, folate and vitamin B₁₂ in 140 rural black preschool children aged 3-5 years living in five different villages in the Letaba area, near Tzaneen. 39.2% were anaemic having hemoglobin levels below 11.1 g/dl.

Approximately 10% were considered to be iron-deficient. On the basis of subnormal red cell folate values, 1 in 4 children was folate-deficient, and suggested the need for intervention at the community level such as enrichment of the staple foodstuff, maize

meal with folic acid. Some limitations in our study should be considered. Firstly, this study was conducted in hospitalized patients and hence the results in this study are not a true representative of the general population. Secondly, this study did not measure vitamin A. There are studies which showed that children with iron deficiency anemia had more frequently low serum retinol [14,19] but its potential contribution as a cause of nutritional anemia in this study was not estimated. Thirdly, quantitative estimation of CRP was not done, which would have identified false positive elevation of serum ferritin. It is possible that ferritin values, especially those in the neighborhood of the cut-off point were increased by acute inflammation [20,21].

Fourthly, the measurements of metabolites such as homocysteine and methylmalonic acid may have identified the functional deficiencies of vitamin B 12 and folic acid. Finally the sample size of this study is small and hence the results of this study could not be generalized to the whole population even though our hospital catered to patients from all over the country. Hence further studies are recommended to formulate a policy in treating those patients with nutritional anaemias.

Conclusion

Anaemia continues to be a public health program in children in India. Iron deficiency either alone or in combination is the commonest nutritional cause of anaemia. Iron deficiency anemia and its association with folic acid deficiency and vitamin B₁₂ deficiency were not significant in our study. Pure or mixed folic acid and vitamin B12 deficiency are important but not commonly recognized cause of anaemia in these children.

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