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### STUDY OF MEAN MICRONUTRIENT LEVELS AMONG CHILDREN DIAGNOSED WITH NUTRITIONAL ANAEMIA AT A TERTIARY CARE HOSPITAL OF DISTRICT AZAMGARH

Deepak Kumar Pandey<sup>1</sup>, Rajesh Kumar<sup>2</sup>, Kamlesh Kumar<sup>3</sup>

<sup>1</sup>Assistant Professor, GMC Azamgarh, <sup>2</sup>Professor, Department of Paediatrics, GMC Azamgarh, <sup>3</sup>Department of Pathology GMC Azamgarh

#### ABSTRACT

**Introduction:** Anaemia is a significant public health problem with major consequences for human health and socioeconomic development. Anaemia is an indicator of poor nutrition and poor health developing countries carry the most significant burden of the reported cases of anaemia whose aetiology is often multifactorial. This study was undertaken to assess the mean micronutrients levels in children of nutritional anaemia having Iron, Folate, and Vitamin B12 deficiency.

**Materials and Methods:** This cross sectional study was done in Department of Paediatrics, Government Medical College Azamgarh. Children of age 6 months to 14 years admitted having signs and symptoms of anaemia were included in the study. Sample size calculated was 157. Each case was subjected to Complete Blood Count, General Blood Picture, serum iron, ferritin, folate and vitamin B12 level estimation. Data was analysed using unpaired test, ANOVA and chi square test by using SPSS software version 20 trial.

**Result:** Mean iron level was significantly low in females, rural areas, low socioeconomic status and those malnourished/underweight. Mixed iron, folate and B12 deficiency was found in 48.41%, 30.57% and 22.93% cases respectively. In 24.20% cases no deficiency was found and was classified anaemia due to some unspecified causes. Nearly 31.85% had pure iron deficiency, 12.10% had pure folate deficiency and 10.83% had pure vitamin B12 deficiency. In mixed form of anaemia, iron plus folate, folate plus vitamin B12 and iron plus B12 contributed to 8.92%, 4.46% and 2.55% cases respectively.

**Conclusion:** Nutritional deficiency anaemia is contributing to a large proportion of anaemia patients. More intensified programmes are needed especially for female children, children of rural areas, low socioeconomic status and malnutrition/underweight.

**Keywords:** Anaemia, micronutrients, iron, folate, B12, malnutrition

Corresponding author: Dr. Deepak Kumar Pandey Email id: dpandey210@gmail.com

#### INTRODUCTION

According to WHO, anaemia is a widespread public health problem with measure consequences for human health as well as social and economic development. Anaemia is functionally defined as an insufficient RBC mass to adequately deliver oxygen to peripheral tissue. According to WHO data on 'Global prevalence of anaemia 2011', anaemia affects 273.2 million children of age 6 months to 59 months, which corresponds to 42.6% of the total population of the children<sup>1,2</sup>. Nutritional deficiencies are the primary cause of anaemia. Anaemia of nutritional origin is an acquired problem caused by diet that lacks sufficient quantity

of bioavailable essential hematopoietic nutrients to meet the need for haemoglobin and red blood cell synthesis. Forty two percent of the causes of anaemia in children are attributable to iron deficiency. India has a mean haemoglobin concentration of 10.6 gram/dl in children of age 6 months to 59 months, which already comes under the category of mild anaemia<sup>1,2</sup>. WHO and UNICEF therefore re-emphasize the urgent need to combat anaemia and stress the importance of recognizing its multifactorial etiology for developing effective control programs. The aim of this study was to evaluate Iron, vitamin B12 and folate deficiency in children with anaemia and to study the mean

micronutrient levels in children of nutritional anaemia having iron, folate, and vitamin B12 deficiency.

## **MATERIAL AND METHODS**

This cross sectional study was done in department of paediatrics, GMC Azamgarh.

Children of age 6 months to 14 years admitted in department of paediatrics, having signs and symptoms of anaemia during November 2016 to March 2018 were included in the study.

The sample size was calculated using the formula  $n = Z^2P(1-q)/d^2$ .  $d$  is the 'allowable error of prevalence' which was taken as 10%. Prevalence was obtained from National family health survey-3 (NFHS 3) data of Uttar Pradesh<sup>2</sup>, which was 73.9%. Sample size calculated by 157. This study was approved by ethical committee of the institute. Subjects were included in the study after taking informed consent from the patient/guardian.

In this study we excluded the children who had received iron, folate vitamin B12 therapy, blood transfusion in immediate past, patient diagnosed with other pathological anaemia and seriously sick children. After taking detailed history and clinical examination, 5ml of blood sample was collected through venepuncture and the sample was divided into two parts, E.D.T.A. sample were subjected to complete blood count (CBC) and general blood picture (GBP), and the serum sample were stored at- 200 degree Celsius in cryovials for the estimation of serum iron, ferritin, folate and vitamin B12. Iron estimation was done using calorimetric method with ferrozine without deproteinization. Ferritin estimation was done by particle enhanced immunoturbidimetric assay. Vitamin B12 estimation was based on competitive test principle using intrinsic factor specific for vitamin B12. Folate assay based on a competitive test principle using natural folate binding protein (FBP) specific for folate. Cut of value for serum iron, ferritin, folate and vitamin B12 are 30 microgram/dl, 15 ng/ml, 5.0ng/ml and 200pg/ml respectively. If the serum sample were not having any deficiency of iron, ferritin folate and vitamin B12 were diagnosed as anaemia due to some unspecified causes. We had employed the student unpaired test and analysis of variance (ANOVA) for quantitative data and chi

square test for quantitative data analysis by using SPSS version 20 trial.

## **RESULTS**

Socio-demographic profile of this patient has been shown in table-1. Among these 157 children, 52.87% were males and 47.13% were females. The percent proportion of anaemia was more among toddlers (27.39%) and lowest among adolescents (10.83%). Majority belonged to urban areas were 65.6% as compared to 34.4% of rural area. A higher proportion belonged to middle (47.13%) followed by lower (35.67%) socioeconomic status as per Kuppuswamy's classification. Maximum cases belonged to severe (31.84%) followed by moderate (28.66%) and mild (19.05%) grade of undernutrition or underweight as per WHO classification of malnutrition/underweight. Out of all anemic causes, 57.96% cases were of moderate grade of anaemia, 38.21% were of severe and 3.82% were of mild grade of anaemia. In general blood picture maximum were of microcytic hypochromic (31.85%) followed by microcytic hypochromic (24.20%), dimorphic (23.57%) and normocytic normochromic (20.83%). Iron deficiency was present in 90% of the cases having microcytic and 83.7% cases of dimorphic general blood picture. Folate deficiency was observed in 47.37% cases, which was of microcytic general blood picture and 40.54% cases of dimorphic general blood picture. National vitamin B12 deficiency found in 48.65% dimorphic followed by 36.84% macrocytic, 25% normocytic and 16% microcytic general blood picture cases.

## **DISCUSSION**

There was decrease in prevalence of anaemia with the increase in age and these findings are similar to the study done by Rajaratnam et al in Tamil Nadu<sup>3</sup>. This is probably due to the reason that early age group children were maximally dependent on their care providers for their nutrition.

In present study, males were slightly more than females. This is similar to the study done by Gomber et al<sup>4</sup>. Iron and ferritin levels were significantly low in females along with low level of folate and vitamin B12. It can be because of poor attitude towards female child health and nutrition in

our society. In this study almost two third of cases belonged to urban area. Health care facilities are usually easily accessible in urban areas and this can be attributed to the increase in the number of urban patients. This is in accordance with several studies done in the past<sup>5,6</sup>. Mean micronutrient levels were compared between urban and rural subgroup. Rural population has statistically significant iron deficiency. Though the level of ferritin, vitamin B12 and folate were also low in rural population but difference was not statistically significant. This can be attributed to the poor nutritional care in the rural population. In our study anaemia was more prevalent among cases of middle and low socioeconomic status. Several studies done in South East Asia also showed similar results<sup>7,8</sup>. Prevalence of anaemia was graded on the basis of specific nutrient deficiencies in different socioeconomic groups and it was found that iron deficiency anaemia was more in low and middle socioeconomic status. Cobalamin and folate deficiency was also more prevalent in the low and middle socioeconomic status group. Mean iron level was significantly low in low socioeconomic status, though the mean micronutrient level of ferritin, folate and B12 were also low in the low socioeconomic status but were not statistically significant. This finding is supported by the study on adolescent girls in Korea, where there was a relationship between household income and ferritin level for iron deficiency anaemia.<sup>9</sup> In our study 80.25% anaemic cases had some grade of malnutrition/underweight and only 19.75% of cases had normal nutrition, this is in accordance with study done in Northern Himalayan state India and Bihar where anemic cases suffered from different grades of malnutrition/underweight.<sup>10,11</sup> Mean micronutrient level for iron was significantly low in different grades of malnutrition/underweight compared to children with normal nutritional status. The level of ferritin, folate and vitamin B12 were also low in different grades of malnutrition/underweight but difference was not statistically significant. Among 157 anaemic cases, prevalence of moderate anaemia was highest followed by severe and mild. Similar results were found in other studies done in past. The reason behind this may be that these studies included only

hospitalised cases.<sup>6</sup> In present studies maximum percentage of cases were of microcytic hypochromic general blood picture (31.85%) followed by microcytic (24.20%), dimorphic (23.57%) and normocytic normochromic blood picture (20.38%). The finding is similar to other study done in past in which they found that in maximum cases general blood picture was of microcytic hypochromic type<sup>10</sup>. In the present study, pure or mixed iron deficiency had the highest prevalence (48.41%). Prevalence of iron deficiency was commonest in studies done in past<sup>12,4</sup>. Pure or mixed folate deficiency was around 30.57% and contributed to the second most common cause of nutritional deficiency anemia and this was in accordance with Mamobolo et al who found folate deficiency anemia was the second most common cause of nutritional deficiency anemia<sup>10</sup>. In present studies vitamin B12 or cobalamin (22.93%) deficiency was least common cause of nutritional deficiency anemia. The study was similar to other studies in which they found vitamin B12 deficiency was the least common cause of anemia<sup>14</sup>. Variation in the causes of anemia and micronutrient levels in different studies may be contributed to either selection of age group or demographical and geographical reasons. Children with unspecified anemia (24.20%) in whom deficiency of iron, folate and vitamin B12 was found, maximum number belonged to high and middle socioeconomic status had moderate to severe grades of anemia and normocytic normochromic blood picture. This may be due to coexisting disease such as malaria, worm infestation, hemoglobinopathies and intake of haematinics, any undiagnosed chronic disease, haemolytic, thyroid disorders, liver disorders and renal disorders<sup>15,16,17</sup>.

## **CONCLUSION**

Nutritional deficiency anemia is still observed among a large proportion of the anemic patients. In spite of large scale supplementation with iron and folate, this deficiency is still prevalent, so the strengthening of same is required. Vitamin B12 deficiency is also common in children. Large proportion of the pediatric population is vegetarian so supplementation and fortification of vitamin B12 is also required to reduce the prevalence of anemia.

**Table-1 Demographic profile and micronutrient deficiency in children with anemia**

Demographical Factors		N=157	Iron Deficiency N=76	Folate Deficiency N=48	Vitamin B12 Deficiency N=36
Age Group	Infant	18(11.47%)	6(7.89%)	4(8.33%)	6(16.67%)
	Toddler	43 (27.39%)	23(30.26%)	14(29.17%)	8(22.22%)
	Pre-School	38(24.20%)	19(25%)	12(25%)	10(27.78)
	Adolescent	17(10.83%)	8(10.53%)	8(16.67%)	3(8.33%)
Sex	Male	83(52.87)	43(56.87%)	24(50%)	21(58.33%)
	Female	74(47.13%)	33(43.42%)	24(50%)	15(41.67%)
Residency	Urban	103(65.6%)	33(43.42%)	18(37.50%)	10(27.78%)
	Rural	54(34.4%)	43(56.58%)	30(62.50%)	26(72.22%)
Socioeconomic Status	Upper	27(17.20%)	3(3.95%)	5(10.42%)	5(13.89%)
	Middle	74(47.13%)	33(43.42%)	16(33.33%)	18(50%)
Underweight/ Undernutrition	Mild	31(19.75%)	15(19.74%)	5(10.42%)	6(15.67%)
	Moderate	45(28.66%)	22(28.95%)	18(37.5%)	14(38.89%)
	Severe	50(31.84%)	34(44.74%)	19(39.58%)	11(30.55%)
	Normal	31(19.75%)	56.58	6(12.5%)	5(13.89%)
Grades of Anemia	Mild	6(3.82%)	3(50%)	2(33.33%)	1(16.67%)
	Moderate	91(57.96%)	47(51.65%)	28(30.77%)	23(25.27%)
	Severe	60(38.21%)	26(43.33%)	18(30%)	12(20%)
General Blood Picture	Microcytic Hypochromic	50(31.85%)	45(90%)	0	8(16%)
	Macrocytic Hypochromic	38 (24.20%)	0	18(47.37%)	14(36.84%)
	Dimorphic	37(23.57%)	31(83.78%)	15(40.54%)	18(48.65%)
	Normocytic Normochromic	32(20.83%)	0	3(9.38%)	8(25%)

**Table-2: Mean Micronutrients Level and Their Correlation with Demographic Profile.**

Demographical factors		Iron Deficiency Anemia				Folate Deficiency Anemia		Vitamin B12 deficiency Anemia	
		Iron Mean±SD (mg/L)	P Value	Ferritin Mean±SD (meg/dl)	P Value	Folate Mean ± SD (meg/dl)	P Value	B12 Mean±SD (eg/dl)	P Value
Age Group	Infant	0.19±0.1	.333	19.79±32.56	.682	3.41±1.72		54.84±23.50	0.284
	Toddler	0.15±0.08		7.29±4.68		3.91±2.83		55.08±24.34	
	Pre-School	0.32±0.51		33.30±120.18		3.64±1.57		44.69±21.34	
	School Going	0.19±0.09		10.43±10.51		3.75±1.16		52.17±26.63	
	Adolescent	0.18±0.10		8.38±0.10		4.02±2.44		48.11±16.78	
Sex	Male	0.25±0.16	0.0094	21.18±81.74	0.0001	4.22±2.44	0.1253	51.59±20.32	0.8861
	Female	0.18±0.17		9.28±8.66		3.31±1.46		50.43±25.87	
Residency	Urban	0.19±0.10	0.0415	10.33±22.3	0.4916	3.03±1.66	0.0674	58.43±25.87	0.6776
	Rural	0.25±0.1		13.32±12.33		4.07±2.15		67.85±51.34	
Socioeconomic Status	Upper	0.27±0.05	0.031	20.84±4.05	0.956	4.04±1.52	0.079	48.41±18.18	0.976
	Middle	0.23±0.08		10.35±15.44		3.86±0.94		46.36±22.10	
	Lower	0.19±0.07		9.87±83.03		2.53±2.52		45.80±25.03	
Nutrition Status Under-Weight Or Undernutrition	Mild	0.18±0.09	0.006	12.90±22.37	0.486	3.48±1.39	0.603	62.40±28.37	0.516
	Moderate	0.17±0.18		10.38±111.93		3.45±1.56		52.64±20.20	
	Severe	0.12±0.08		7.93±4.65		3.23±2.76		49.52±25.16	
	Normal	0.32±0.08		14.33±3.82		4.55±0.72		64.20±8.35	

**Table-3: Etiological distribution of anemia among the study participants.**

Type of anemia	Number of children (n=157%)	95% confidence interval%
Pure Iron deficiency	50(31.85%)	24.16-40.54
Pure Folate deficiency	19(12.10%)	9.24-15.34
Pure Vitamin B12deficiency	17(10.83%)	9.12-13.45
Iron plus Folte deficiency	14(8.92%)	7.01-10.45
Folate plus Vitamin B12 deficiency	7(4.46%)	3.55-5.45
Iron plus B12 deficiency	4(2.55%)	1.22-6.45
Iron plus Folate plus Vitamin B12 deficiency	8(5.10%)	3.67-5.96
Unspecified	38(24.20%)	15.78-38.87
Total	157(100%)	



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