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ORIGINAL ARTICLE

Iron Profile in Children with Behavioural Disorders: A Prospective Study in a Tertiary Care Hospital in North India

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Abstract Iron deficiency anemia is the most frequent micronutrient deficiency in the developing countries like India especially affecting pregnant women and young children. Iron is an essential element involved in myelin formation, neurotransmitter synthesis and neuro-metabolism. Several behavioural disturbances have been reported in iron deficient children. In the present study, we determined the prevalence of iron deficiency anemia in children with behavioural disorders and assessed the improvement in terms of symptoms (by child behaviour check list), haematological parameters and iron status after treatment with oral iron. In this prospective study, 44 children in the age group of 3–12 years who were diagnosed with behavioural disorders were evaluated. Complete blood counts using automated hematology analyzer and iron parameters (serum iron, total iron binding capacity, % transferrin saturation and serum ferritin) were measured in all the patients to assess the prevalence of iron deficiency in these children. Thirty age matched controls were also studied. Iron deficiency was found in 32 (73%) children, as assessed by transferrin saturation <16% and/or serum ferritin <16 μg/l. Following treatment with iron for 100 ± 10 days, there was a statistically ($P \leq 0.05$) significant improvement in the clinical features, haematological profile and iron status. The presence of iron deficiency in children with behavioural disorders and subsequent improvement in clinical features, haematological profile and iron status suggests a possible causal relationship between iron deficiency and behavioural disorders.

Keywords Iron deficiency · Behavioural disorders · Children

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Introduction

Anemia is a frequently encountered clinical manifestation and an important public health problem, especially in developing countries. The National Family Health Survey-3 (NFHS-3) was conducted in India in 2005–2006 and covered 29 states. The data showed that the percentage of children in the age group of 6–35 months that are anemic is as high as 80% or more in Chhattisgarh, Gujarat and Punjab [1]. The figures have increased since the time of the previous survey [2]. Deficiency of iron is the most frequent micronutrient deficiency specially affecting pregnant women and young children, the estimated prevalence being 49.7 and 74%, respectively [2, 3].

Iron being a key element in the metabolism of almost all living organisms, affects growth in infancy, motor development and coordination, language development and scholastic achievement in children [4]. It is an essential element involved in myelin formation, neuro-metabolism and neurotransmitter synthesis [5, 6]. Studies on animal models and humans have examined the effects of iron deficiency and/or anemia on cognitive function and behaviour. In rodent models, both moderate and severe iron deficiency have been shown to cause alterations in activity patterns and motor development, as well as diminished cognitive performance [7–9]. Similarly, children with iron deficiency have been

reported to be irritable and disruptive. They have a short attention span and lack interest in their surroundings [10, 11].

Though iron deficiency anaemia is very frequent in developing countries like India, a search in literature did not reveal any large study which correlated iron status and behavioural disorders. This study thus attempts to explore the possible causal relationship between iron deficiency anemia and behavioural disorders.

Materials and Methods

This prospective study was conducted between December 2006 and March 2008. It was carried out on 44 children in the age group of 3–12 years with behavioural disorders (as per inclusion and exclusion criteria) in the Hematology Section of Department of Pathology in collaboration with the Department of Psychiatry, University College of Medical Sciences and Guru Teg Bahadur Hospital, Delhi. Thirty age-matched healthy controls were also included in the study. An informed written consent was taken from the parents/guardian of the patients.

Children with the following behavioural disorders (as per DSM IV-TR criteria) [12], were included in the study-Attention deficit hyperactivity disorder (ADHD), Conduct disorder, Oppositional defiant disorder (ODD), Pica and others like Phobia, Elimination disorder, Trichotillomania, Breath holding spells and Thumb sucking. These children were clinically evaluated and a check list score was given to them based on Child Behaviour Check List (CBCL) [13]. This is a 113 item behavioural problem checklist intended to evaluate pathological behaviour in children aged between 1½ and 18 years.

The following investigations were done in all patients and controls at recruitment: complete blood counts using automated hematology analyzer MS 9-3, examination of stained peripheral blood film (Wrights stain) [14], serum iron (ICSH, 1978) [15], total iron binding capacity [16] and serum ferritin (ELISA). A diagnosis of iron deficiency was made if % transferrin saturation was less than 16% and/or serum ferritin <16 μg/l.

All patients were given oral iron in the dose of 4–6 mg/kg in three divided doses for 100 ± 10 days. A close follow up was done to ensure compliance with iron therapy and to assess for side effects, if any. All patients were clinically evaluated at the end of iron therapy. Complete blood counts and iron parameters were also repeated at this time.

The data was subjected to statistical analysis using the SPSS statistical software (version 12). Unpaired student *t* test and Chi square test were used for comparison between cases and controls and paired student *t* test for comparison between pre and post treatment values. A *P*-value of <0.05 was taken to be statistically significant.

Results

The age of the patients ranged from 3 to 12 years with a mean ± SD of 8.7 ± 2.6 years. This was not statistically different from the controls (range 3–12 years, mean ± SD 6.9 ± 2.9 years).

The clinical diagnosis of the patients is shown in Table 1.

A checklist score was given to all patients at recruitment. The score ranged from 2 to 48 with a mean ± SD of 14.4 ± 13.4.

Complete Blood Counts (CBC)

Values of the various parameters of CBC in patients and controls are shown in Table 2.

Anemia (Hb < 11 g/dl) was detected in 28 (63.6%) patients. It was mild (Hb 8–11 g/dl) in 25 (56.8%) and moderate (Hb < 8 g/dl) in 3 (6.8%) cases.

Examination of Stained Peripheral Blood Smear

Red cells were microcytic hypochromic in 19 (43.2%), predominantly normocytic normochromic with few microcytes in 12 (27.3%) and 13 (29.5%) had normocytic

Table 1 Diagnosis of patients at presentation

Diagnosis	No.	%
ADHD	21	47.7
Conduct	5	11.4
ODD	4	9.1
Pica	9	20.5
Others	5	11.4
Total	44	100

Table 2 Complete blood counts of patients and controls

Parameter	Patients ($n = 44$)		Controls ($n = 30$)		*P*-value
	Range	Mean ± SD	Range	Mean ± SD	
Hb (g/dl)	6.1–14.8	10.7 ± 1.8	11.9–13.7	12.7 ± 0.6	0.000*
PCV %	19.0–41.0	32.1 ± 4.5	36.1–42.0	38.7 ± 1.8	0.000*
RBC ($\times 10^{12}$/l)	3.14–4.84	3.82 ± 0.49	3.57–4.52	4.04 ± 0.22	0.010*
MCV (fl)	55.6–93.2	76.7 ± 8.7	82.1–92.1	85.7 ± 3.2	0.000*
MCH (pg)	18.6–31.9	25.9 ± 3.1	28.1–32.1	29.8 ± 0.9	0.000*
MCHC (g/l)	26.0–36.0	31.2 ± 2.4	32.4–37.8	35.6 ± 1.4	0.000*
P/C ($\times 10^9$/l)	116–473	236.3 ± 78.3	168–483	281 ± 79.7	0.018*
TLC ($\times 10^9$/l)	3.4–10.2	7.1 ± 1.7	5.3–9.5	7.2 ± 1.4	0.823

(Unpaired Student *t* test) * Significant

Table 3 Iron status of patients and controls

Parameter	Patients ($n = 44$)		Controls ($n = 30$)		P-value
	Range	Mean ± SD	Range	Mean ± SD	
S iron (μg/dl)	11.0–157.0	64.6 ± 29.2	55.0–157.0	99.1 ± 27.5	0.000*
TIBC (μg/dl)	251–481	396.3 ± 48.8	234–419	329.4 ± 51.4	0.000*
% TS	5.3–35.6	17.9 ± 7.0	17.8–41.9	26.9 ± 6.7	0.000*
S ferritin (μg/l)	2.0–35.0	11.3 ± 8.5	17.0–170.0	50.9 ± 33.5	0.000*

(Unpaired Student t test)
* Significant

normochromic red cells. All the 30 controls had normocytic, normochromic morphology.

Iron Parameters

Values of serum iron, total iron binding capacity (TIBC), % transferrin saturation (%TS) and serum ferritin are shown in Table 3.

Iron deficiency was diagnosed in 32 (72.7%) patients. Of these 32 patients the deficiency was latent (serum ferritin < 16 μg/l, Hb: normal) in 4 (12.5%) patients. All 32 patients were given treatment with oral iron for 100 ± 10 days and were followed up at regular intervals. The patients were assessed again in terms of clinical and hematological improvement (CBC and iron parameters) at the end of treatment. Five patients were lost to follow up and 27 patients completed the study.

Complete Blood Counts Before and After Treatment

Complete blood counts in 27 patients who completed the study are shown in Table 4.

There was a significant ($P < 0.001$) increase in the hemoglobin concentration, PCV, RBC count, MCV, MCH and MCHC in all the 27 children who received iron therapy.

However, it was observed that even after treatment with iron, hemoglobin concentration was less than 11 g/dl in 9 (33%). MCV was <80 fl in 16 (59.3%) patients and MCH was <27 pg in 12 (44.4%) patients.

Iron Status Before and After Treatment

Iron status before and after treatment of these patients is shown in Table 5.

Table 4 Complete blood counts before and after treatment

Parameter	Before treatment ($n = 27$)		After treatment ($n = 27$)		P-value
	Range	Mean ± SD	Range	Mean ± SD	
Hb (g/dl)	6.1–11.4	9.7 ± 1.2	8.6–13.7	11.4 ± 1.2	0.000*
PCV %	19.0–36.6	30.0 ± 3.8	28.2–44.2	36.2 ± 3.6	0.000*
RBC ($\times 10^{12}$/l)	3.14–4.84	3.66 ± 0.44	3.34–5.51	4.06 ± 0.59	0.000*
MCV (fl)	55.6–81.5	71.9 ± 7.3	70.2–89.5	80.1 ± 5.1	0.000*
MCH (pg)	18.6–28.3	24.5 ± 2.8	19.9–30.8	27.2 ± 2.6	0.000*
MCHC (g/l)	26.0–33.2	30.0 ± 1.9	25.4–35.3	31.8 ± 2.3	0.001*
P/C ($\times 10^9$/l)	165–473	257.2 ± 84.3	165–445	271.2 ± 81.4	0.313
TLC ($\times 10^9$/l)	3.41–9.70	6.55 ± 1.63	3.47–12.40	6.86 ± 1.82	0.275

(Paired Student t test)
* Significant

Table 5 Iron status before and after treatment

Parameter	Before treatment ($n = 27$)		After treatment ($n = 27$)		P-value
	Range	Mean ± SD	Range	Mean ± SD	
S.iron (μg/dl)	11.0–86.0	48.1 ± 15.3	34.0–135.0	80.7 ± 20.1	0.000*
TIBC (μg/dl)	301–475	416.1 ± 35.0	296–423	368.2 ± 28.4	0.000*
% TS	5.3–33.0	14.3 ± 5.3	8.1–33.2	20.6 ± 5.3	0.000*
S ferritin (μg/l)	2.0–11.0	6.2 ± 2.8	5.0–16.0	9.8 ± 3.9	0.000*

(Paired Student t test)
* Significant

There was a statistically significant rise in serum iron, %TS and serum ferritin with a statistically significant decrease in TIBC after iron therapy. Though there was a rise in the serum ferritin value, it continued to be <16 μg/l in 24 (88.9%) patients and was 16 μg/l in 3 (11.1%). % Transferrin saturation was <16% in 25 (92.6%) patients after therapy.

Clinical Evaluation After Treatment

Checklist score was applied to all 27 patients after iron therapy. There was a statistically ($P < 0.001$) significant decrease in the score to 0–43 with a mean ± SD of 10.5 ± 12.3 (pre treatment 2–48, 14.4 ± 13.4).

All children tolerated iron therapy well and did not develop any side effects or any other problem. All of them had a general feeling of well being and showed clinical improvement.

Discussion

Anemia is a frequently occurring clinical manifestation especially in developing countries. In a world census, the estimated prevalence of anemia in the world and its extrapolation to the Asian population suggests that approximately 47.5% of the anemic population lives in South Asia. Extrapolating these figures to the Indian population suggests that approximately half of the population of around one billion will be anemic [17]. Results from NFHS-3 indicate that 80% of children in India in the age group of 6–35 months are anemic. The figures in this survey have increased since the time of the previous survey [2].

Deficiency of iron is the most frequent micronutrient deficiency and 90% of all types of anemia in the world are due to iron deficiency [18]. IDA remains a major health problem in India mostly affecting young children and pregnant women [2, 3]. In a study done in North East Delhi on 545 children of 9–36 months of age, iron deficiency was detected in 87% children [19]. These figures highlight the magnitude of iron deficiency and iron deficiency anemia in India.

Iron is an important component of haemoglobin, myoglobin and enzymes which perform vital functions in the body [4]. Deficiency of iron thus affects various tissues, organs and systems in our body in addition to causing anemia. The brain has a higher concentration of iron than any other metal [20]. It is an important nutrient and an essential element involved in myelin formation and neurotransmitter synthesis [4].

Deficiency of iron is reported to cause several behavioural disturbances in children. These children are irritable, disruptive, have a short attention span and lack interest in the surroundings. It has been shown that neurologic development in infants and scholastic performance in older children may be impaired in iron deficiency [11].

In the present study, iron status was assessed in 44 children with behavioural disorders. Of these, 32 (73%) were found to be iron deficient (% transferrin saturation <16% and/or serum ferritin <16 μg/l). These findings are in concordance with the study conducted by Konofal et al. who demonstrated abnormally low serum ferritin levels in children with ADHD [21]. In their study, serum ferritin levels were twice as low in children with ADHD as in controls.

Several other authors have also found an association between iron deficiency anemia and poor cognitive and motor development and behavioural problems [6, 10, 11]. It has also been seen that the severity of these changes is proportional to the severity and duration of anemia [22, 23].

An interesting observation in our study was that, all the nine patients with pica, were iron deficient. The association of pica with iron deficiency is well documented in literature [24, 25]. Singhi et al. studied 50 children with iron deficiency anemia with pica and found that the plasma iron level (mean ± SD) in children with pica (42.7 ± 9.2 mg/dl) was about 20% lower than that in controls (51.5 ± 10.0 mg/dl, P value < 0.001) [26].

It was interesting to know that there were four children in this study in whom iron deficiency was latent and they still presented clinically with various behavioural disorders. Similar results have also been shown in other studies, where it was found that iron deficiency even in the absence of anemia, resulted in biochemical alterations that impaired behaviour in children [7, 27].

All patients were given oral iron therapy for 100 ± 10 days. Hematological parameters and iron status were measured at the end of therapy. Twenty-seven patients completed the study and five were lost to follow-up. Following treatment with iron, there was a statistically ($P \leq 0.001$) significant improvement in the haematological parameters. The mean ± SD hemoglobin levels increased from 9.7 ± 1.2 to 11.4 ± 1.2 g/dl after treatment with iron. PCV, RBC count, MCV, MCH and MCHC also showed a significant ($P \leq 0.001$) increase (Table 4). There was a statistically ($P < 0.001$) significant increase in serum iron, %TS, serum ferritin and a decrease in TIBC after iron therapy.

Nine (33%) children still had a Hb of < 11 g/dl at the time of follow-up. These children had taken iron regularly and had tolerated it well. This suggests that a longer period of iron therapy is required to alleviate anemia.

Clinical re-evaluation of patients with IDA using the checklist score in the present study showed that there was a statistically ($P < 0.001$) significant decrease in the post

treatment values of the CBCL. The mean ± SD before and after treatment were 14.4 ± 13.4 and 10.5 ± 12.3, respectively. Previous studies in Indian children have also shown that children with iron deficiency anemia had lower levels of attention and concentration and that there was significant improvement with iron supplementation [28, 29].

The degree of response was variable in children with different diagnoses. However, it could not be assessed statistically as the number of patients in each group was small. Patients with pica showed drastic improvement in symptoms post therapy.

The presence of iron deficiency in children with behavioural disorders and subsequent improvement in clinical features, haematological profile and iron status suggests a possible causal relationship between iron deficiency and behavioural disorders.

An area of concern is the observation by some authors that iron deficiency induced changes are not completely reversed even after iron repletion. Lozoff et al. studied 191 children with IDA in infancy that were followed up at 5 years of age to check for developmental status. The authors concluded that children with IDA during infancy are at risk for long lasting developmental disadvantages compared to iron replete infants [30].

Though children with behavioural disorders in this study showed clinical improvement with iron therapy, some of them continued to remain iron deficient. Longer follow-up on a larger number of children is required to fully ascertain to what extent the behavioural disorder was reversed.

In view of these findings and the magnitude of IDA in our country, it is imperative that iron deficiency be treated expeditiously and effectively to prevent a cohort of population existing with reduced mental and psychomotor development. With some of the changes extending into adulthood it will have a significant impact on productivity and ultimately on the economy of the world.

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