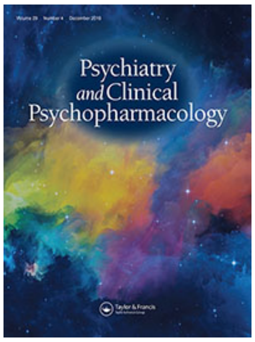


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# Vitamin B12 and haemoglobin levels may be related with ADHD symptoms: a study in Turkish children with ADHD

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**ABSTRACT**

**OBJECTIVE:** In this study, we evaluated vitamin B12 and iron parameters in Turkish children with ADHD in order to examine the relationship between ADHD symptoms and these parameters.

**METHODS:** Drug-naive 100 ADHD patients, aged between 6 and 12 years old, were included in the study. None of them had acute or chronic diseases. All patients were assessed by using the Schedule for Affective Disorders and Schizophrenia for School Age Children – Present and Lifetime Version (K-SADS-PL). Conners Parent Rating Scale (CPRS) was used for screening ADHD symptoms and symptom severity. Blood samples were evaluated for ferritin, haemoglobin, MCV, RDW, and vitamin B12 parameters.

**RESULTS:** We indicated an inverse relationship between haemoglobin levels and learning, anxiety subscale scores of CPRS. Also, vitamin B12 and psychosomatic subscale scores were found negatively related whereas the relationship was in the opposite direction for ferritin. Vitamin B12 level was negatively correlated with learning problems and psychosomatic subscales of CTRS in the combined subtype of ADHD.

**CONCLUSION:** Vitamin B12 and iron support may be useful in treatment of childhood ADHD, especially for learning problems, besides medication.

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**KEYWORDS**
ADHD; vitamin B12; haemoglobin; iron; learning problems

## Introduction

Attention Deficit Hyperactivity Disorder (ADHD) is a prevalent childhood developmental disorder, characterized by inattention, hyperactivity and impulsivity which is not appropriate for the developmental stage [1]. Although research on neurobiological basis of ADHD have increased considerably in the recent years, our knowledge about ADHD aetiology is still limited [2].

Iron deficiency is one of the nutritional factors mentioned in ADHD aetiology which is very common in childhood. Iron is an element with a role in not only basic brain functions [3,4], but also in myelination, development of oligodendrocytes and neurotransmitter synthesis [5–8]. Iron deficiency has been shown to lead structural and functional changes even in the early period and affect dopamine metabolism and myelination [9]. Also, the relationship between behavioural changes and iron deficiency has been demonstrated [10]. On the other hand, the results of the studies investigating the role of iron deficiency and anaemia in ADHD are controversial [11]. Some studies have revealed that the severity of ADHD symptoms was related with low serum ferritin levels [12–17]. However, some other studies challenged these results and claimed that brain iron should be investigated in this kind of studies instead of serum ferritin levels [18,19].

Vitamin B12 deficiency is a common nutritional deficiency and related with cognitive defects [20,21]. Bourre [22] concluded that even borderline vitamin B12 deficiency was found to impair cognitive functions in adolescents. A micronutrient supplement including vitamin B12 has also been shown to improve attention deficits in children [23] and adolescents [22]. However, this was not the case for adults [24]. Also, an animal study showed that early life was sensitive to dietary methyl donor supplementation which can alter prefrontal cortex-dependent cognitive behaviours [25]. In addition to these, findings of a genetic variation study indicated that folate-homocysteine metabolic pathway gene variants may be related with ADHD aetiology due to mild hyperhomocysteinemia and vitamin B12 deficiency [26].

ADHD is a serious public health problem with a burden to country economy. Therefore, the importance of investigating aetiological risk factors for ADHD which are common in public arises. Evidence from aetiological

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studies will also prove an insight for treatment options. As there are a few studies with conflicting results about the role of iron and vitamin B12 deficiency in ADHD, we aimed to explore the relationship of ADHD symptoms and ferritin, haemoglobin (Hb), mean corpuscular volume (MCV), red blood cell distribution width (RDW), vitamin B12 parameters which can be changed in iron or vitamin B12 deficiency.

## Materials and method

This multicentre study included 100 outpatients with childhood ADHD who applied to the Child and Adolescent Psychiatry Departments of Bülent Ecevit University and Zonguldak Women and Children Hospital between 2014 and 2015. Parents of all patients gave informed consent to participate in the study after the study protocols had been fully explained. This study was approved by the Local Ethics Committee (IRB number: 2015-104-04/11) and adhered to the Declaration of Helsinki.

### The study procedure

After the psychiatric evaluation of patients, the laboratory records of patients who fulfilled the inclusion criteria were scanned and the ones who had recent CBC, vitamin B12, and ferritin level checks in the paediatric deparments in one-month period before psychiatric assessment were included for the study if their parents gave consent. Inclusion criteria were meeting ADHD diagnosis (F90.0 according to ICD-10), being between 6 and 12 years of age, and not receiving psychiatric treatment at that time or in the past. Those with mental retardation, psychotic disorders, autism spectrum disorders, organic mental syndromes, mood and anxiety disorders, and who had a history of acute/chronic disease or anaemia by report or according to the records were excluded from the study.

### Schedule for Affective Disorders and Schizophrenia for School Age Children – Present and Lifetime Version

To evaluate childhood ADHD and ADHD subtypes, the valid and reliable Turkish version of Schedule for Affective Disorders and Schizophrenia for School Age Children –Present and Lifetime Version (KSADS-PL) was administered to all patients [27]. K-SADS-PL is a semi-structured diagnostic interview used to assess current and past psychiatric disorders in childhood and adolescence according to DSM-IV criteria [28].

### Sociodemographic data

In the first interview, sociodemographic and clinical data forms were filled by the researchers and socioeconomic status (SES) was classified into 5 levels according to the Hollingshead-Redich Scale [29] (level 1–2 representing high, 3 representing middle and 4–5 representing low SES).

### Conners Parent Rating Scales

Conners Parent Rating Scales (CPRS) were applied to the parents of children to evaluate ADHD symptoms [30]. CPRS has subscales evaluating learning problems, hyperactivity, behaviour problems, anxiety, and psychosomatic symptoms. The valid and reliable Turkish version of CPRS was used in this study [31].

### Blood parameters

In all patients, peripheral venous blood records were evaluated by the paediatrician researcher to determine Hb, ferritin, RDW, and vitamin B12 levels. Iron deficiency was defined as ferritin < 12 ng/mL [32] and vitamin B12 deficiency was diagnosed if vitamin B12 level was <200 pg/mL according to the National Diagnosis and Treatment Guideline of Vitamin B12 Deficiency-2011 version.

### Statistical analysis

Statistical analysis was performed by using the Statistical Package for Social Sciences (SPSS) version 15.0. Kolmogorov–Simirnow test was used to determine the normal distribution of variables. Since the distribution of our variables did not fit for normal distribution, we used Spearman's correlation. Mann Whitney *U* test and *T* test were also conducted. Significance value was considered to be $p < .05$.

## Results

In our sample, 28 (28%) of 100 patients were female, and 72 (72%) patients were male. Mean age of the patients was 9.01 years. The majority of the patients (43%) were in the 4th level of SES according to Hollingshead Redlich Scale. None of the patients were in the first level, and the percentage of the patients in level 2, 3, and 5 corresponded to 4%, 24%, and 29%, respectively.

Twenty-eight (28%) of the patients had inattentive, 2 (2%) had hyperactive/impulsive and 70 (70%) had combined subtype of ADHD. No significant difference in blood parameters was found between the inattentive and combined subtype groups. However, CPRS total, oppositional, conduct problems, and psychosomatic subscale scores were found higher in the combined subtype than the inattentive subtype group. Also, we detected some differences between ADHD subtypes in the correlations of blood parameters and CPRS subscale scores. In the attention deficit subtype, RDW was found positively correlated with hyperactivity, behaviour

**Table 1.** Blood parameters of the sample.

| Blood parameters | Mean ± SD | Min–Max. |
|---|---|---|
| Haemoglobin (g/dL) | 12.93 ± 0.83 g/dL | 10.9–15.3 |
| Mean corpuscular volume (f/L) | 82.27 ± 4.86 f/L | 66.1–93.3 |
| Red blood cell distribution width (%) | 14.68% ± 1.23% | 12.3–17.9 |
| Ferritin (ng/mL) | 21.35 ± 13.59 ng/mL | 5–100.3 |
| Vitamin B12 (pg/mL) | 331.68 ± 181.19 pg/mL | 79–1258 |

Note: SD: standard deviation; Min–Max: minimum–maximum value.

problem, oppositional subscale scores. Nevertheless, ferritin was positively correlated with psychosomatic subscale scores, while vitamin B12 level was found negatively correlated with learning problem and psychosomatic subscale scores of CPRS in the combined subtype. RDW increase was also found to be associated with higher total CPRS scores in both subtypes.

Twenty-four per cent of the patients had iron deficiency while 21% of the patients had vitamin B12 deficiency. The mean ± standard deviation (SD) and the minimum–maximum values of the blood parameters (ferritin, haemoglobin, MCV, RDW, vitamin B12) in our study group are shown in Table 1.

When the relationship between the blood parameters and CPRS scores was checked, MCV was not found associated with any of the subscale scores. CPRS total scores were found significantly correlated with the RDW scores ($p = .007$). However, CPRS total scores were not found to be associated with haemoglobin and ferritin levels. Furthermore, we examined the relationship between the subscale scores of CPRS and blood parameters. The oppositional subscale scores did not differ in any parameters we were investigating. However, the results indicated the psychosomatic subscale scores were in a reverse relationship with vitamin B12 levels ($p = .006$).CPRS learning problem scores were found associated with low haemoglobin levels as well as CPRS anxiety subscale scores score ($p = .004$; $p = .01$, respectively). CPRS psychosomatic subscale scores were significantly associated with higher ferritin levels ($p = .028$). Higher hyperactivity and behaviour problem subscale scores were in accordance with higher RDW levels. ($p = .007$; $p = .012$, respectively).

Our results about the correlations between the blood parameters and subscales of CPRS are summarized in Table 2.

## Discussion

We aimed to investigate the relationship between ADHD symptoms and ferritin, haemoglobin, MCV, RDW, vitamin B12 parameters which can be changed in iron or vitamin B12 deficiency. To our knowledge, this is one of the few studies exploring the association between ADHD symptoms and vitamin B12 levels.

In our study, we did not find a significant relationship between vitamin B12 levels and CPRS total scores or any of the subscale scores other than the psychosomatic subscale. Also, vitamin B12 was found to be related with learning problems and psychosomatic subscales of CPRS in the combined subtype. Micronutrient supplements including vitamin B12, were shown to improve intelligence scores [33,34] and concentration deficits in children [23]. Kennedy and Haskell [35] reviewed similar cohort studies and remarked a significant negative relationship between homocysteine levels and cognitive performance. Furthermore, responders were identified as the ones who had lower amounts of micronutrients in their diets and had higher blood levels of micronutrients after supplementation [36,37]. Case control studies examining vitamin B12 levels have conflicting results. In a recent study, vitamin B12 levels of school-age patients with ADHD were found to be lower than controls [38,39]. On the other hand, there is another study conducted in Turkish adult population which did not find vitamin B12 levels of ADHD patients statistically different than controls [40].

Nevertheless, vitamin B12 has a critical role in the conversion of homocysteine to methionine which is mediated by methionine-synthase. This reaction is essential for nucleotide synthesis, genomic, and non-genomic methylation in the central nervous system [41]. B vitamins are not only required for neurotransmitter synthesis and functioning, but also for myelination of the spinal cord and brain [42]. As ADHD is a neurodevelopmental disease, vitamin B12 deficiency

**Table 2.** The relationship between the blood parameters and CPRS subscale scores.

| Blood parameters | LP | H | O | A | PS | BP | Total |
|---|---|---|---|---|---|---|---|
| Hg (g/dL) | $r = -0.286$ | $r = 0.105$ | $r = -0.053$ | $r = -0.257$ | $r = -0.057$ | $r = 0.020$ | $r = -0.125$ |
| | $\mathbf{p = .004}$ | $p = .301$ | $p = .603$ | $\mathbf{p = .010}$ | $p = .573$ | $p = .847$ | $p = .215$ |
| MCV (f/L) | $r = -0.134$ | $r = 0.053$ | $r = 0.000$ | $r = -0.061$ | $r = -0.081$ | $r = 0.028$ | $r = 0.050$ |
| | $p = .184$ | $p = .598$ | $p = .998$ | $p = .544$ | $p = .424$ | $p = .783$ | $p = .624$ |
| RDW ((%) | $r = -0.088$ | $r = 0.270$ | $r = 0.151$ | $r = 0.157$ | $r = 0.072$ | $r = 0.251$ | $r = 0.268$ |
| | $p = .386$ | $\mathbf{p = .007}$ | $p = .134$ | $p = .119$ | $p = .474$ | $\mathbf{p = .012}$ | $\mathbf{p = .007}$ |
| Ferritin (ng/mL) | $r = -0.012$ | $r = -0.170$ | $r = 0.012$ | $r = 0.036$ | $r = 0.220$ | $r = -0.053$ | $r = 0.024$ |
| | $p = .902$ | $p = .091$ | $p = .909$ | $p = .720$ | $\mathbf{p = .028}$ | $p = .602$ | $p = .813$ |
| Vit B12 (pg/mL) | $r = -0.182$ | $r = -0.10$ | $r = -0.067$ | $r = -0.109$ | $r = -0.272$ | $r = -0.010$ | $r = -0.114$ |
| | $p = .070$ | $p = .922$ | $p = .505$ | $p = .281$ | $\mathbf{p = .006}$ | $p = .923$ | $p = .259$ |

Notes: Vit B12: Vitamin B12; LP: learning problem subscale of Conners Parent Rating Scale (CPRS); H: hyperactivity subscale of CTRS; O: oppositional subscale of CTRS; A: anxiety subscale of CTRS; PS: psychosomatic subscale of CTRS; BP: behaviour problem subscale of CTRS; Total: total score of CTRS. $p < .05$ is indicated in bold.

may be one of the factors that worsen ADHD symptoms and it may be a reasonable attitude to keep the vitamin B12 levels high in ADHD patients.

According to parameters related to iron, CPRS total scores were not found associated with the Hb and ferritin levels in our study. Likewise, Donfrancesco et al. [18], Millichap et al. [43] and Menegassi et al. [19] showed no significant relationship between ferritin levels and ADHD symptom scores in ADHD patients. However, Oner et al. [14] found lower ferritin levels and higher CPRS, Conners' Teacher Rating Scale (CTRS) total scores in accordance. We found a positive correlation between psychosomatic subscale of CPRS and ferritin. Other studies reported the relationship between the ADHD hyperactivity scores on Conners scale and low ferritin levels in their ADHD group [15–17]. Moreover, the results of the case control studies searching ferritin levels differ from each other. Donfrancesco et al. [18], Menegassi et al. [19] and Perçinel et al. [17] did not find significant differences in ferritin levels between groups. On the contrary, some other studies detected lower ferritin levels in ADHD subjects [12,44,45]. These different results may be due to the methodological differences between studies. The effect of comorbidities in the psychiatric disorder groups, and being on medication as well as the number of the subjects may have influenced the results.

In this study, haemoglobin was found to be negatively associated with learning problems and anxiety subscales of CPRS. As we did not include any anaemic patients to the study, it may be thought that adverse cognitive effects of iron deficiency may have a synergistic effect on ADHD cognitive symptoms before the haemoglobin reaches the anaemic level. Therefore, it may be favourable to support iron levels for ADHD patients besides medication.

One of the limitations of our study is the lack of a control group. Secondly, we only used CPRS to evaluate the symptoms of ADHD and their severity. Also, the effect of comorbidity was not investigated in this research. These factors may have influenced our results. On the other hand, we selected a homogeneous patient group according to SES, age and gender with a reasonable number. We did not include any patients on psychotropic drugs considering appetite changes. In addition, none of the patients had acute or chronic infection. As ferritin and B12 levels are related with food intake and infections, these including criteria seem important.

In conclusion, especially low B12 and haemoglobin levels may be related with learning problems in children with ADHD. As vitamin B12 and iron deficiency are seen commonly in our developing country, we suggest that vitamin B12 and iron support may make a difference in treatment of cognitive symptoms in ADHD besides medication. Further investigation is needed in this field.

## Disclosure statement

No potential conflict of interest was reported by the authors.

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